Structured Notes According to

MICROBIOLOGY

Revision friendly **Fully Colored Book/Structured Notes**

For Best results, watch the video lectures along with reading notes

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CONTENTS

MICROBIOLOGY

LIST OF IMPORTANT TOPICS

BACTERIOLOGY

- Strep/Staph Classification
- Staph Toxins
- Legionella: Case scenario
- Typhoid toxins
- E.coli, Salmonella , Cholera, Pseudomonas : Lab tests, toxins
- Clostridium: Toxins
- Tuberculosis: Lab Ix most important
- Rickettsia, Syphilis, Leprosy
- Leptospira,Brucella: Case based Qs

PARASITOLOGY

- Malaria: Image based Qs, stages
- Amoebiasis, Giardiasis, Nematodes/Trematodes Transmission And Host
- Toxoplasmosis
- Cysticercosis-Esp. NCC stages
- Haemoflagellates
- Coccidian Parasites
- Cestodes

VIROLOGY

- Classification of viruses
- Influenza: ShiG vs DriG
- Hepatitis: Types, Hep B serology tests most important
- HIV: Opportunistic, AIDS-deterministic infections, Mx
- Herpes Group : Types and infections
- General Virology
- Recent updates in Virology

MYCOLOGY

- Classification Of Fungi
- Dermatophytes
- Endemic Mycoses
- Cryptococcus
- Madura Mycosis: Case based, Radiology
- Candida , occupational fungal diseases

IMMUNITY (To be done with Pathology)

- Immunoglobins
- Immunodeficiency Disorders
- Antigen , Antigen antibody reactions

LEARNING OBJECTIVES

UNIT 1: INTRODUCTION

- Introduction to microbiology
  - Basics of microbiology
  - Stains
  - Motility
  - Culture media, images of various culture media

1 INTRODUCTION TO MICROBIOLOGY

BASICS OF MICROBIOLOGY

00:05:20

- All Cocci are Gram +ve Except
  - Meningococci
  - Gonococci
  - Veillonella
  - Moraxella
- All Bacilli are gram -ve Except
  - **M** - **M**ycobacterial species
  - **A** - **A**nthracis bacillus
  - **C** - **C**lostridium species
  - **D** - **D**iphtheriae Corynebacterium
  - **O**
  - **N** - **N**ocardia
  - **A** - **A**ctinomyces
  - **L** - **L**isteria
  - **D** - **D**iphtheroids (Normal commensals of throat)

How to remember

- MAC DONALD

STAINS

1. Gram stain

00:12:40

- Discovered by Christian gram
- Sequence of Bacterial Smear
  - **M**ethyl - **M**ethyl violet /crystal violet/gentian violet (primary stain)
    ↓
  - **G** - **G**ram's Iodine (Mordant)
    ↓
  - **A** - **A**cetone (Decolourizer)
    ↓
  - **S** - **S**afranin (Counter stain)

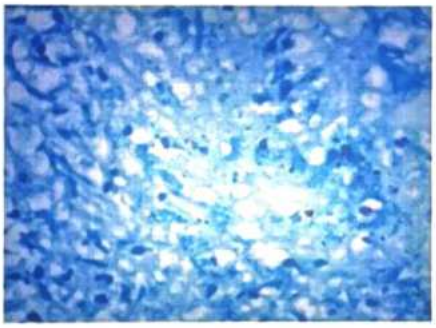
How to remember

- Methyl GAS

- Gram positive: Purple to violet
- Gram negative: Pink
- All steps are done for 1 minute except decolorizer step which is done for 2- 3 seconds
- Factors governing gram staining
  - Thickness of Peptidoglycan Layer
    → Most important factor governing gram staining
    → Thick Peptidoglycan layer: Gram positive feature
    → Thin Peptidoglycan layer: Gram negative feature
  - Dye - iodide complex
    → Strong: Gram positive
    → Weak: Gram negative
  - LPS [Lipopolysaccharide]
    → Feature of gram-negative microorganism
    → Dissolved by acetone

2. Acid fast staining (ZN staining)

00:24:30



- Gram stain and acid fast stain are differential stains
- Gram stain differentiates between Gram +ve & Gram -ve
- AF stain differentiates between Acid Fast & Non Acid Fast
- Acid fastness
  - It is the ability to resist decolorization
  - It is due to the presence of mycolic acid
  - More the mycolic acid more will be the acid fastness
  - Mycobacterium tuberculosis : 20% acid fast
  - Mycobacterium Leprae: 5% acid fast
  - Nocardia: 1% acid fast
  - 0.5 to 1% acid fast: Cryptosporum, Cyclospora, ...spora
    → These are Coccidian parasites which cause Diarrhea in HIV +ve individuals
  - 0.5 to 1% acid fast:
    → Hooklets of egg of T. Saginata
    → Scolex of E.Granulosus
  - Spermatic head, bacterial spores are also acid fast
  - Species of legionella that is acid fast: Legionella micdadei

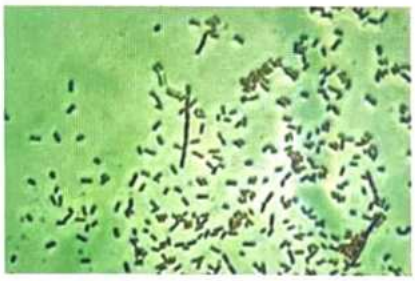
- Kinyoun method of staining

KINYOUN STAINING

  - Modification of ZN staining
  - Cold method of staining (phenol concentration is ↑sed for fixation)
  - The heating part is not done

3. Albert stain

00:32:21



Chinese letter pattern

Albert stain A	Albert stain B

• Made up of 3 important components
  ○ **G** - Glacial Acetic acid
  ○ **M** - Malachite Blue
  ○ **T** -Toluidine Blue
	• It is composed of
	  ○ Iodine
	  ○ Potassium iodide

How to remember

- GMT

- It stains: C. Diphtheriae
  - Green
  - Cuneiform arrangement / Chinese letter pattern

4. Bipolar stain

00:35:01

- Poles are stained and remaining spared
- Gives safety pin appearance
- Bipolar stained organisms include
  - Yersinia pestis
    → Causes Plague aka black death
  - Hemophilus ducreyi
    → Causes chancroid (painful)
  - Donovani granulomatis / Klebsiella granulomatis
    → Leads to granuloma inguinale
    → It is having a Beefy red ulcer
  - Vibrio parahemolyticus
    → It is a Halophilic Vibrio (7-8%)
    → It leads to Seafood poisoning
  - Burkholderia mallei
    → Leads to Glanders disease (Disease of Horse)
    → It is Non-Motile
  - Burkholderia pseudomallei
    → Leads to Melioidosis
    → It is Motile
  - Pasteurella species
  - Francisella species

Capsulated organism

00:43:44

- **P** - Pneumococcus, pseudomonas
- **A** - Anthracis bacillus
- **K** - Klebsiella species
- **I** - Influenza Hemophilus
- **Y** - Yersinia species
- **B** - Bordetella species
- **M** - Meningococci
- **C** - Clostridium perfringes [C. welchii], Cryptococcus neoformans
- **V** - Vibrio parahaemolyticus

How to remember

- PAKIYBMCV

Composition, Demonstration & Functions of Capsule

00:47:14

- Capsule is mainly made of polysaccharide
- Exception: Bacillus Anthracis capsule is made of polypeptide
- Demonstration of the capsule is done by: Quellung & Mc Fadyean's Reaction

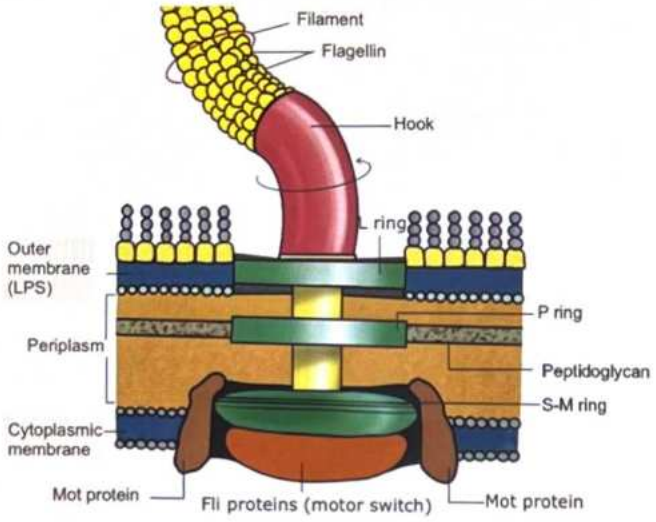
Quellung Reaction	MC Fadyean's Reaction
• Ag- Ab Reaction ○ Capsular Ag and preformed Capsular Ab's are mixed up (Serological reaction) resulting in Capsular swelling/delineation • Done for Pneumococcus, H. influenza	• Culture + Loffler's methylene blue: Capsule stains blue • Done for bacillus Anthracis

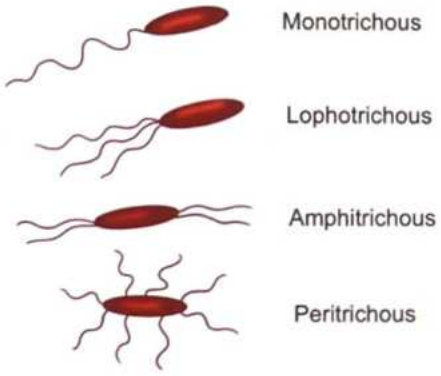
- Functions of capsule
  - Acts as a virulent factor by
    - → Avoids opsonization & thus prevents phagocytosis
    - → Biofilm formation. It is responsible for Antibiotic resistance

MOTILITY

00:53:40

- It is due to flagella
- Flagella is made up of Flagellin which is found to be antigenic
- Types of flagella
  - Monotrichous: Vibrio cholera & Pseudomonas
  - Amphitrichous: Alcaligenes faecalis
  - Lophotrichous: Spirillum minus
  - Peritrichous: E. coli, Proteus, Listeria salmonella





Type of motility	Organism
• Tumbling motility	• Listeria monocytogenes
• Darting motility	• V. Cholera
• Swarming Motility	• Proteus
• Stately motility (slow form of swarming)	• Clostridium species
• Gliding motility	• Mycoplasma
• Corkscrew motility	• T. pallidum
• Lashing motility	• Borrelia
• Falling leaf-like motility	• Giardia lamblia
• Twitching motility	• Trichomonas vaginalis
• Rotatory motility	• B. coli (ciliate)

- Spirochetes have endoflagella which arise from periplasmic space
- Methods to demonstrate motility
  - Hanging drop method
  - Semi-solid agar method: Done using Mannitol motility agar [mc used]
  - Phase contrast microscopy
  - Dark ground microscopy: For spirochetes

CULTURE MEDIA

01:07:51

Refer Table 1.1

Previous Year's Questions

Q. A child presented with bloody stools and abdominal pain. Which of the following enrichment media will be used to culture the following organism? (NEET 2021)

A. Selenite f broth
B. Alkaline peptone water
C. Robertson cooked media
D. BHI

3. Selective Media

- Media which inhibits the growth of the unwanted organism and promote the growth of an organism of interest
- Mostly solid
- Wilson & Blair: S. typhi
- Potassium tellurite agar: C. diphtheria
- Mannitol salt agar: S. aureus

Mac-Conkey Agar

- Media that is Indicator, Differential & Selective: Mac-Conkey agar
  - Indicates the color change
  - Differentiates between lactose fermenting & Non-lactose fermenting organisms
    - → Lactose fermenting: Changes color to Pink
    - → Non-lactose fermenting: No change

- Selective for gram –ve bacilli
- Contents
  - → P – Peptone
  - → L – Lactose
  - → A – Agar
  - → N – Neutral red (Indicator dye)
  - → T – Taurocholate sodium (selective agent)

How to remember

- PLANT

- Organism which ferments Lactose
  - E. coli
  - Klebsiella
  - S. Sonnei

CLED (Cystine Lactose Electrolyte Deficient Media)

- CLED is a better culture media than Mac-Conkey agar because
  - It is Non-selective
  - Prevents swarming of proteus
  - Differential: Differentiate between lactose fermenters and Non-lactose fermenters
  - Indicates color change

Previous Year's Questions

Q. CLED media is better than Mac - Conkey media. Why? (AIIMS – Nov - 2019)

A. It stimulates the growth of staph and Candida as it is non-selective

B. Inhibits swarming of proteus

C. Differentiates between Lactose fermenter & non lactose fermenter

D. Sodium Taurocholate is used as selective agent

4. Transport media

- Maintains viability but no growth

Transport media	Organism
• VR media	• V. cholera
• Cary Blair Media	• V. cholera
• Pike's media	• S. Pyogenes
• Stuart's and Amie's media	• Gonococci and Meningococci
• Sach's Buffered Glycerol saline	• Shigella and Salmonella

Anaerobic media

- Robertson Cooked Meat Media (RCM): Clostridium species

Clostridium species	
Proteolytic	**Saccharolytic**
• Turns meat black	• Turns meat pink
• Eg: C. Tetani	• Eg: C. Perfringens

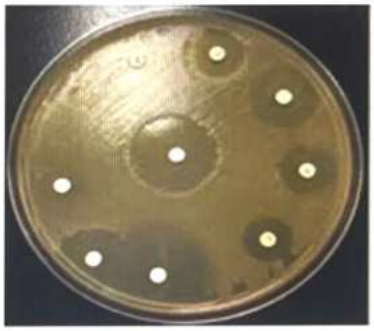
- Thioglycollate broth

Sugar Media

- Glucose powder, peptone water (containing Andrade's indicator) and culture growth is added to a test tube, after one day it turns from colorless to Pink. This suggests that the organism have fermented Glucose
- Andrade's indicator: indicates the colour change

Antibiotic sensitivity testing

- Media used: Mueller Hinton Agar (MHA)
- Method
  - Kirby Bauer Disc Diffusion Testing (KBDDT)
    - → M/c used
    - → Zone of inhibition present: Sensitive
    - → Zone of inhibition absent: Resistant



KBDDT

  - Stoke's method of antibiotic testing
    - → Better method, as controls are used

IMAGES OF VARIOUS CULTURE MEDIA

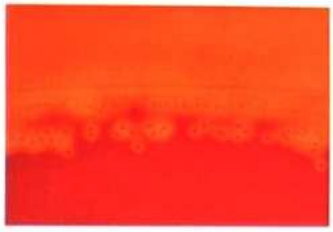
01:36:12

1. Nutrient agar

- Intended use
  - It serves as the base for the other media preparation
  - Isolation of the non-fastidious organism
  - NA slant is used for the short-term preservation of the bacteria.
  - Performing serological and biochemical tests.
  - It is used to study bacterial colony morphology and demonstrating the swarming & pigment production.
  - Semisolid NA (0.2-0.4%) is used for the demonstration of bacterial motility and preservation of bacteria. An additional of 6% in NA makes it firm agar that prevents swarming.

Organism	Image
• Growth of P. Aeruginosa on NA	
• Growth of S. aureus on NA	
• Swarming of proteus on NA	
• Motility in Semisolid NA	

2. Blood agar

Alpha Hemolysis	Beta Hemolysis
	

3. Chocolate agar

- Intended use
  - It is more nutritive than Blood agar.
  - H. influenza only grows on CA as it requires X (hemin) factor and V (NAD) factors for their survival, which are only present in CA
  - Isolation of Neisseria

4. Mac Conkey agar

- Media Type: Differential or Indicator media or low selective media.
- Content of Mac – Conkey agar
  - **P** - Peptone
  - **L** - Lactose
  - **A** - Agar
  - **N** - Neutral red (selective)
  - **T** - Taurocholate Sodium
- Method for sterilization: Autoclaving
- Colour of prepared medium: Red with purplish tinge coloured clear to slightly opalescent gel forms.

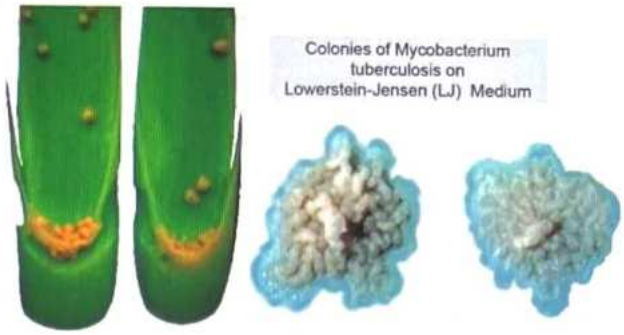
- Intended uses
  - It is used for the isolation of gram-negative bacilli
  - It differentiates lactose fermenting (LF) bacteria (E. coli, Klebsiella) from non-LF bacteria.
  - LF Bacteria- Pink colored colonies
  - NLF bacteria - Pale or colorless colonies

5. Cystine lactose electrolyte deficient agar

- Intended use
  - It is used for the isolation of uropathogens
  - It differentiates LF bacteria (yellow colonies) from non-LF bacteria
    - → It is less inhibitory to gram-positive bacteria
    - → Prevents swarming of Proteus

6. Lowenstein Jensen Medium

- Intended uses
  - It is used for the isolation of mycobacterium species.
  - Colonies appear dry, wrinkled, rough, buff, and tough.



Colonies of Mycobacterium tuberculosis on Lowerstein-Jensen (LJ) Medium

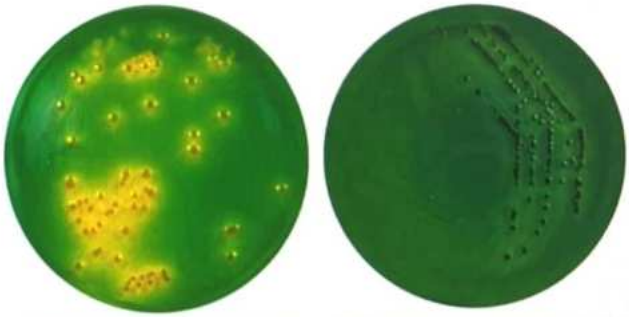
7. Loeffler serum slope medium

Loeffler Medium

- Intended Use
  - Early detection (6-8 hrs) of C. Diphtheriae.
  - Enhances the production of metachromatic granules and appearance suggestive of Chinese-letter formation in methylene blue stain.
  - Growth of Corynebacterium appears as minute, and cream colored colonies with slightly raised centers.

8. Thiosulfate citrate bile salts sucrose (TCBS) agar

- Intended Use
  - V. cholera produces large colored colonies after fermenting sucrose.



Vibrio cholerae on TCBS Agar

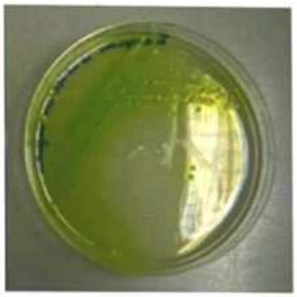
Vibrio parahaemolytics on TCBS Agar

- Non-sucrose fermenter produces green colored colonies.

9. Potassium tellurite agar (PTA)

- Intended use
  - It is used for the isolation of C. Diphtheriae
  - C. Diphtheriae produced black colored colonies on PTA
  - Best selective media for C. Diphtheriae

10. Cetrimide agar



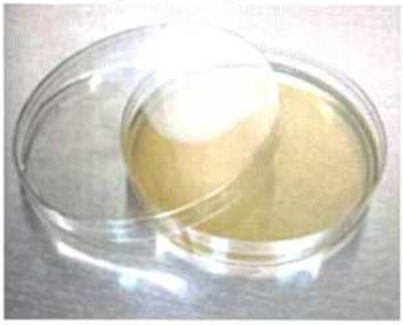
- Indented use
  - Best media for pseudomonas
  - Isolation of P. Aeruginosa

11. Robert son's cooked meat (RCM) medium

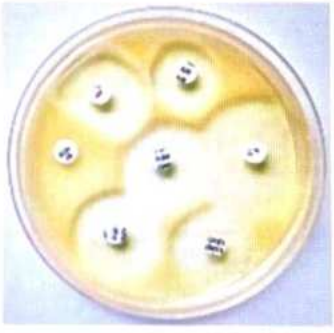
- Intended use
  - Used for the isolation of Anaerobic bacteria
  - It can be also used as an Anaerobic transport medium
  - Demonstration of Proteolytic and Saccharolytic activities

  - Proteolytic bacteria: Blackening of meat particles
    - → C. Tetani
    - → C. Botulinum A, B, F.
  - Saccharolytic Bacteria: Pinking of meat particles
    - → C. Perfringens
    - → C. Difficile
    - → C. Botulinum C, D, E.

12. Mueller Hinton Agar (MHA)



- Intended use
  - To perform Antibiotic susceptibility testing (AST) against isolated pathogenic bacteria from clinical specimens



  - To perform MIC by E. test (Epsilometer test)

13. Brain heart infusion (BHI) broth

- Intended use
  - Used as blood culture media
  - Used for anaerobic bacteria cultivation and the recovery of fastidious bacteria
  - Useful for isolation and cultivation of fungi

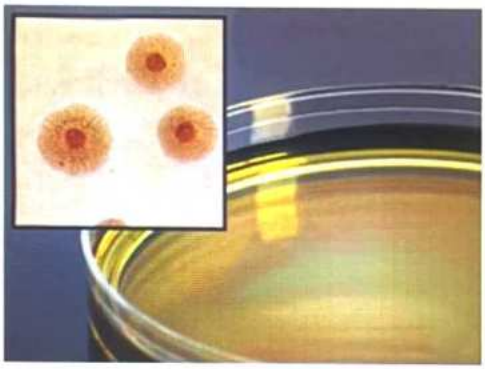
14. Middle brook 7H9 broth

- Media type: Special media for Mycobacterium

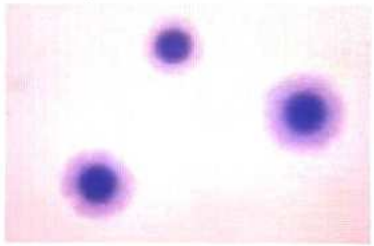
15. Buffered charcoal yeast extract (BCYE) agar

- Media Type: Selective medium
- Intended use
  - This medium is to be used for the isolation and presumptive identification of Legionella.

16. Pleura-pneumonia like organism (PPLO) agar

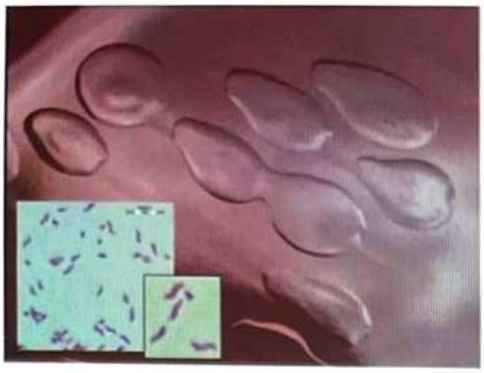


- Appearance: "Fried Egg" colonies on PPLO Agar
- Intended use
  - Recommended for isolation and cultivation of Mycoplasma species.



17. Skirrow's medium

- Intended use
  - Selective media for the isolation of Campylobacter Jejuni
  - Positive oxidase test & Gram stain: Gull wing-shaped Gram negative rods



18. Pike's media

- Media type: Transport medium
- Intended use: Cultivation of streptococcus Pyogenes

19. Amie's medium

- Media type: Transport media
- Main constituents: Charcoal, Sodium Thioglycollate
- Intended use: For collection, transport and preservation of microbiological specimens

20. Stuart medium

- Media Type: Transport medium
- Main constituents: Sodium glycerophosphate, Thioglycollate
- Intended use: Recommended for the preservation and transportation of Neisseria species.

21. Cary-Blair medium

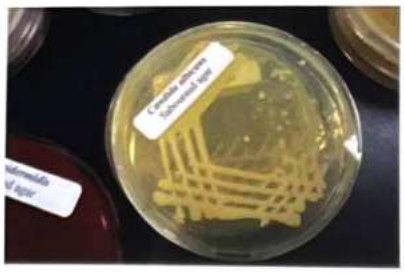
- Media Type: Transport medium
- Used for Vibrio cholera

22. VR medium

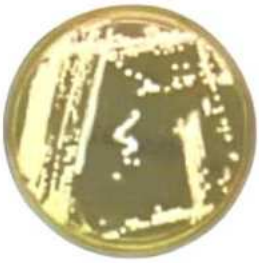
- Media Type: Transport medium
- Main constituents: Crude Sea salts, Peptone, and pH-8.6 is maintained

23. Sabouraud dextrose agar (SDA)

- Main constituents: Dextrose, Peptone, Low pH (5.6), Chloramphenicol
- ntended use
  - Isolation and identification of fungi



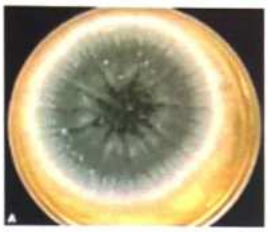
Candida



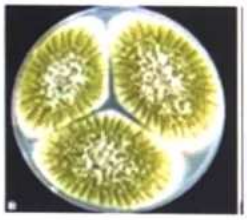
Cryptococcus

Aspergillus colonies on SDA

- A. Fumigatus



- A. Flavus



- A. Niger

Bacteria and their alternate names 01:47:17

Bacteria	Alternate name
Bordetella	Bordet Gengou bacillus
Clostridium Tetani	Nicolair's bacillus
Corynebacterium diphtheria	Klebs-Loeffler's bacillus
Corynebacterium pseudo tuberculosis	Preisz-Nocard bacillus
Hemophilus aegyptius	Koch-Weeks bacillus
Hemophilus Influenzae	Pfeiffer's bacillus
Klebsiella pneumonia	Friedlander's bacillus
Klebsiella ozaenae	Abel's bacillus
Klebsiella Rhinoscleromatosis	Frisch's bacillus
Mycobacterium tuberculosis	Koch's bacillus
Mycobacterium intracellulare	Battey's bacillus
Mycobacterium paratuberculosis	Johne's bacillus

Plasmid borne toxins

- **T-** Tetanospasmin of C. Tetani
- **P-** Pertussis toxin of B. Pertussis
- **H-** Heat-labile toxin (LT) of ETEC
- **A-** Anthrax toxin of B. Anthracis

How to remember

- TPHA

Chromosomal mediated toxins

- **P-** Pyrogenic toxin B of S. Pyogenes
- **P-** Pertussis toxin of B. Pertussis
- **E-** Exotoxin A of P. Aeruginosa
- **Se-** Shiga toxin of S. dysenteriae
- **Covered-** Cholera toxin (CT) of V. cholera

How to remember

- PPE Se Covered

Phage encoded toxins

- **B-** Botulinum toxin of C. Botulinum
- **V-** Verotoxin (Shiga-like toxin) of EHEC
- **D-** Diphtheria toxin of C. Diphtheriae

How to remember

- Bottle Vich Drinks Paa

Intracellular bacteria

- **Y -** Yersinia
- **R -** Rickettsia
- **M -** Mycobacterium species
- **B -** Brucella, Bordetella
- **C -** Chlamydia
- **S -** Salmonella
- **M -** Meningococcus
- **L -** Legionella
- **D -** Donovani Granulomatis

How to remember

- Yaar Ritesh mere koe Bru Coffee Sale main La De

Obligate intracellular bacteria

- M. Leprae
- Rickettsiae
- Chlamydia
- Coxiella burnetti
- T. pallidum

Non-sporing anaerobes 01:56:02

- Gram positive
  - E - Eubacterium
  - L - Lactobacillus
  - M - Mobiluncus
  - B - Bifidobacterium
  - A - Actinomyces
  - P – Propionibacterium

How to remember

- Engineers love MBA Program

- Gram negative
  - B- Bacteroids
  - P- Prevotella, porphyromonas
  - L- Leptotrichia
  - F- Fusobacteriums

How to remember

- BPL Fuse

Oxidase positive organisms

- A - Aeromonas
- L - Legionella micdadei
- V - Vibrio cholera
- C - C. Jejuni
- P - Pseudomonas
- N - Neisseria meningitides and gonorrhea
- H - Hemophilus influenzae
- Oxidase reagent is Tetra methyl para phenylene diamine dihydrochloride

How to remember

- AL VC have PNH

Catalase positive bacteria

- M – Meningitides Neisseria, gonococci
- Y – Yersinia
- L – Legionella, Listeria
- I – Influenzae H.
- P – Pseudomonas, Pasteurella
- S – Staph
- R - HelicobacteR
- S – Shigella (except Shigella dysenteriae Type- 1)
- E (resembles C) – Coliforms
- A – Atypical Mycobacteria
- L – Listeria

How to remember

- MY LIPS are (R) SEALED

Organism and Their Infective Doses 02:04:35

Organism	Infective dose
• ETEC (Entero Toxic E. Coli)	• $10^6 - 10^{10}$
• EIEC (Entero Invasive E. Coli)	• $10^8 - 10^{10}$
• EHEC (Entero Hemorrhagic E. Coli)	• 10 – 100
• Shigella	• 10 – 100
• Salmonella	• $10^3 - 10^6$
• C. Jejuni	• 10^4
• Y. enterocolitica	• $10^8 - 10^9$
• Vibrio	• 10^{10} (water source) • $10^2 - 10^4$ (food)

Pleomorphic organism

- M - Mycoplasma
- C - Clostridium
- H - H. influenzae
- V - V. cholera
- V - V. parahaemolyticus
- P – Proteus

How to remember

- MCH Very Very Progressive

Classification Based on O_2 Requirement 02:11:27

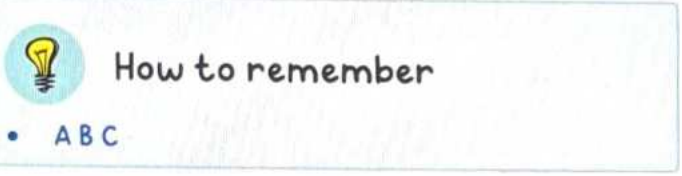
1. **Obligate Aerobes**

- Needs desperate O_2

- Example
  - MTB
  - Pseudomonas
  - Brucella
  - Bacillus

2. Obligate Anaerobes

- Does not need O_2 at all
- Example
  - A - Actinomycetes
  - B - Bacteroids
  - C - Clostridium species



3. Facultative anaerobes

- Changed anaerobe
- Aerobe + anaerobe
- Most of the medically important bacteria
- Example
  - Staphylococcus
  - Streptococcus species

4. Microaerophiles

- Need 5% O_2
- Example
  - Campylobacter
  - Helicobacter

5. Capnophiles

- CO_2 loving
- Examples
  - Brucella abortus
  - Pneumococcus
  - H. Influenzae

Classification Based on Temperature Requirement

02:19:56

- P - Psychrophile: -10° C to +10° C
- M - Mesophile: 20° C to 40° C [medically important bacteria]
- T - Thermophile: > 45° C, > 55° C, > 60° C (Better Option)
  - Can handle >100°C
  - Thermos Aquaticus: Used in PCR
  - Father of PCR: Karry B Mullis

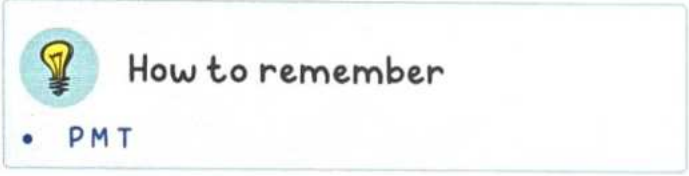


Table 1.1

1. Simple media		2. Complex media	
Nutrient agar	**Nutrient broth**	**Enriched media**	**Enrichment media**
• Have peptone and meat extract • 2% agar • It's a solidifying agent and doesn't provide any energy	• Liquid culture media	• Solid media • Eg: Blood agar and chocolate agar • Blood agar contains 5 to 10% sheep blood • Heat up blood agar causes Lysis of RBC releasing Haemin and NAD, which gives a chocolate color and media changes into chocolate Agar	• Always liquid culture media • Examples o Selenite-F-broth for Salmonella, Shigella o Tetrathionate broth for Salmonella

CLINICAL QUESTIONS

Q.1 Robert Koch was performing an experiment where he wanted to study some properties exhibited by bacteria, fungus etc for which he was using gel culture media. But he could not get the desired results hence, Robert Koch's assistant advised him to use agar instead of gelatine for solidifying culture media for the cultivation of bacteria. Why is agar preferred over gelatine?

A. Agar has more nutrients
B. Gelatin melts at 37°C
C. Gelatin is not easily available
D. Agar is cheaper

Solution:

- Gelatin was the substance originally used to gel culture media. However, gelatin is liquid at 35°C., the optimum temperature for many bacteria.
- In addition, some bacteria are capable of liquifying the gelatin, making it an unsuitable solidifying agent.
- Agar has neither of these drawbacks.
- **Agar is preferred over gelatin, as it is bacteriologically inert, melts at 98°C and usually solidifies at 42°C.**

Q.2 A 38-year-old homosexual man with HIV presented with multiple extensive necrotic lesions with an abundant purulent secretion that covered his face, lips, scalp, and torso. The doctor suspected a coinfection and used Smith Noguchi's media to culture. Smith Noguchi's media is used for:

A. Salmonella
B. Klebsiella
C. Spirochetes
D. Bacillus

Solution:

- Smith Noguchi's media is used for **Spirochetes.**

Smith-Noguchi Medium (by using living tissue) –

- Anaerobic media.
- Used for Cultivation of nonpathogenic treponemes
- E.g. Reiter strain of Treponema phagedenis - **Spirochetes)**

Answers to other options:

- **Salmonella - Wilson and Blair bismuth sulphite** medium is best selective medium.
- **Klebsiella** - On **MacConkey** Agar, these are lactose fermenter.
- **Bacillus - PLET** media is best for culture.

LEARNING OBJECTIVES

UNIT 2: SYSTEMIC BACTERIOLOGY

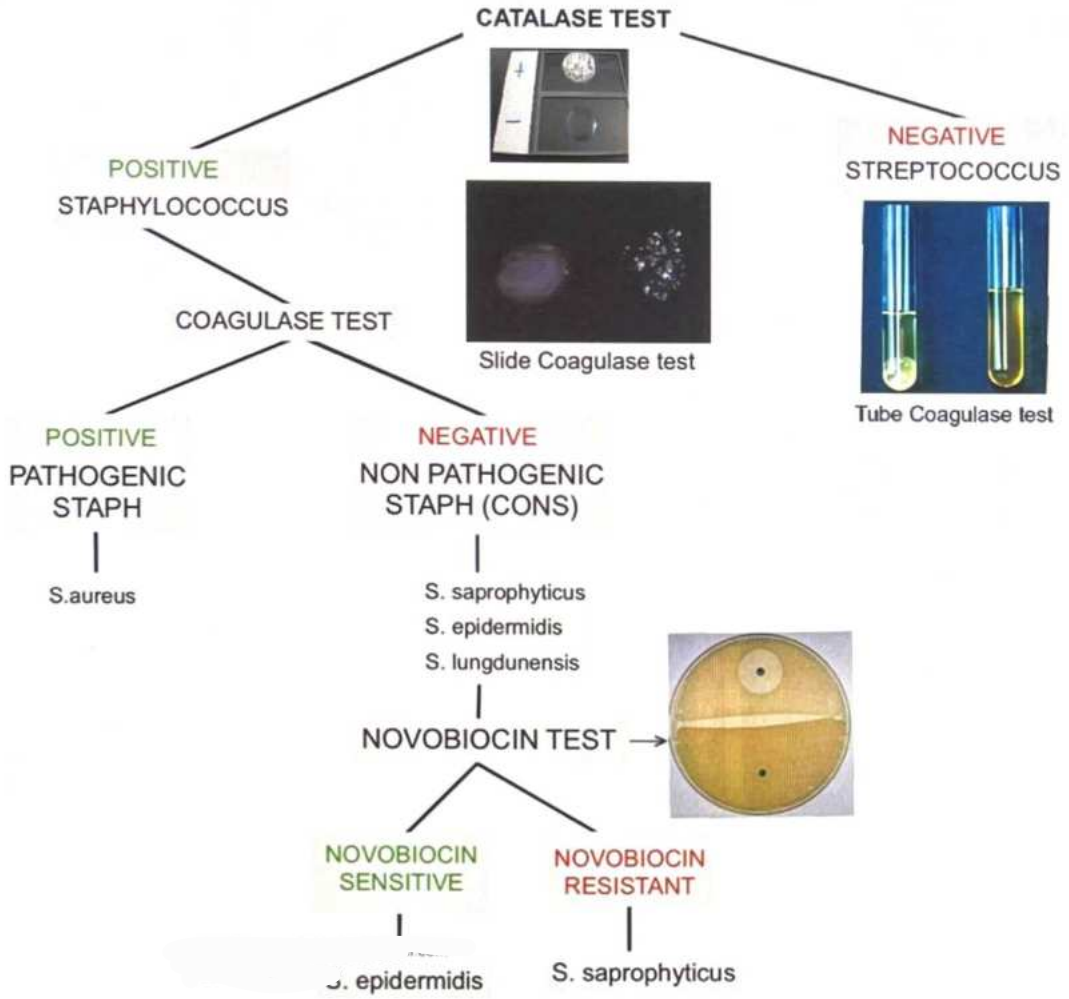
- Gram +ve and Gram -ve Cocci
  - Gram positive cocci
    - → Coagulase negative staph
    - → Staph aureus
    - → Streptococcus
  - Meningococci
  - Gonococci
- Gram positive bacilli
  - Corynebacterium Diptheria
  - Bacillus Anthracis
  - Bacillus Cerus
  - Clostridium: perfringens, Tetani, botulinum, Difficle
  - Mycobacterium TB, Leprae
  - Nocardia and Actinomycetes
  - Listeria Monocytogenes
- Gram negative bacilli
  - Enterobacteriaceae
    - → E. Coli, Klebsiella, Proteus, Salmonella, Shigella
  - Lactose Non fermenters
    - → Pseudomonas, Burkholderia Mallei, Burkholderia Pseudomallei, Acinetobacter Baumanii, Stenotrophomonas Maltophilia
  - Vibrio, Hemophilus, Bordetella, Brucella, Yersinia, Legionella and Spirochetes
  - Leptospira, Rickettsiae, chlamydia, campylobacter, Helicobacter
  - Mycoplasma, Bacteroides

2 GRAM POSITIVE AND GRAM NEGATIVE COCCI

GRAM POSITIVE COCCI

00:00:14

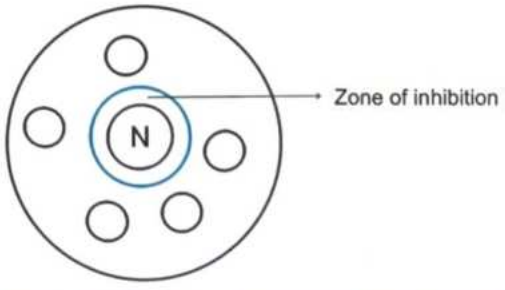
- They are staphylococcus and streptococcus



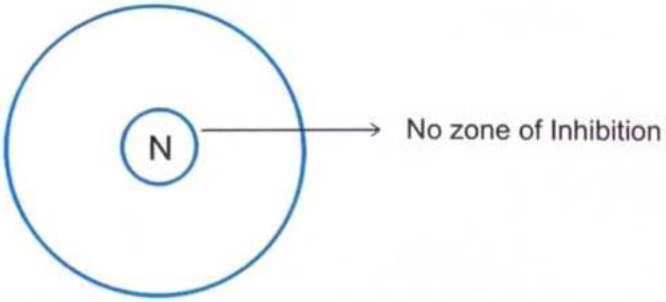
- Novobiocin test: Done for Antibiotic testing
- Slide coagulase test: to determine bound form
- Tube coagulase test: to determine free form
- 3% H_2O_2 + Staph. Colonies: Bubbles (Catalase Positive)

AST [Antibiotic Sensitivity Testing]

- Eg:Novobiocin sensitivity test
- KBDDT method
- Done on
  - MHA [**M**uller **H**inton **A**gar]
  - NA [**N**utrient **A**gar]
- 6 antibiotic discs can be put in an average petridish
- **K**irby **B**auer **D**isc **D**iffusion Testing (KBDDT)
  - Zone of inhibition present: Sensitive [Staph. Epidermidis]



  - Zone of inhibition absent: Resistant [Staph. Saprophyticus]



- Stoke's method of antibiotic testing: Better method but costly

COAGULASE NEGATIVE STAPH (CONS) 00:07:18

- MC organisms/group involved in Early Prosthetic Valve Endocarditis: CONS
- Single most cause for Early Prosthetic Valve Endocarditis: S. aureus

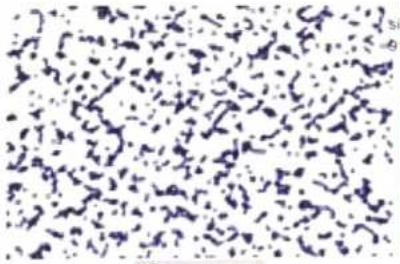
1. Staph Epidermidis 00:07:59

- Normal Commensal of skin Epidermis
- A/w infection due to interventions
  - IV cannula, CSF Shunts, prosthetic valve surgery, catheters
  - Causes early prosthetic valve endocarditis [< 1yr]
- Produces Biofilm which
  - Helps in attachment of organism to various surfaces
  - Contribute to Antibiotic resistance
- Novobiocin sensitive: zone of inhibition is present

2. Staph Saprophyticus 00:11:44

- Young female develops UTI M/c GPC involved - S. Saprophyticus
- Young female develops UTI MC cause overall - E. Coli
- Novobiocin resistance: no zone of inhibition

STAPH AUREUS 00:15:25


S. aureus

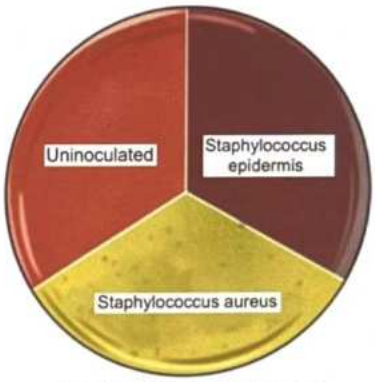

Blood agar

α, β, γ Hemolysis

- Gram positive cocci
- Catalase +ve
- Coagulase +ve
- Gram staining: Grape like clusters (purple to violet)
- Various culture medias used for Staph Aureus
  - NA [Nutrient Agar]: Golden yellow pigmented colonies which enhances on Milk Agar


Golden Yellow Colonies on NA

  - BA [Blood Agar]: It produces β haemolytic colonies on Blood Agar (Complete zone of Hemolysis)
  - Mannitol salt agar



MANNITOL SALT AGAR

    → Best Selective Media
    → In presence of Staph. Aureus the agar changes from pink to yellow
  - Ludlam's media [2nd best media]
  - Salt Milk Agar

- Virulence Factors
  - The thickness of the Peptidoglycan layer
  - Teichoic acid
  - Clumping factor
  - Protein A
    → Is present in COWAN-1 strain of Staph. aureus
    → Plays a role in co-agglutination
    → Has a property of binding with the Fc portion of IgG
  - Haemolytic Factors
    → α Haemolysins: It shows paradoxical action that is it Inactivates at 70° C and Reactivates at 100° C

→ β Hemolysins: It shows Hot & Cold Phenomenon
- In S. Aureus β Hemolysis starts at 37° C but it is best seen at 4° C K/a Hot and Cold Phenomenon

→ γ Haemolysin: γ Haemolysin + PV Toxin [Panton Valentine Toxin] and produces synergohymenotropic action [↑virulence of Staph. aureus]

- PV Toxin / Leucocidin
  - → PV Toxin is aka Leucocidin
  - → It kills Leucocytes

- Enzymes
  - → DNAase
  - → Thermo nuclease
  - → Coagulase [plays the most important role in pathogenicity & virulence]
    - Tube coagulase: Free form of coagulase, Better & more confirmatory
    - Slide coagulase: Bound form of coagulase
  - → Hyaluronidase
  - → Lipase
  - → Phosphatase

Toxin Mediated Illnesses 00:30:28

1. Food Poisoning

- Incubation period of S. aureus is 1-6 hr
- Caused due to consuming contaminated meat and milk products
- D/t preformed Enterotoxin
  - Enterotoxin-A (M/c)
  - Heat stable
  - Vagomimetic action
- Incubation period of S. Aureus Resembles Bacillus Cerus Emetic strain
  - S. Aureus: contaminated meat & milk products
  - B. Emetic: contaminated Chinese fast food

Previous Year's Questions

Q. A 12-year-old boy presents with vomiting within 3 hours of consumption of food at a party. What is the likely causative agent responsible for the symptoms? (NEET - Jan - 2020)

A. Staphylococcus aureus
B. Salmonella
C. Clostridium botulinum
D. Clostridium perfringens

Previous Year's Questions

Q. Few friends had late night dinner. Next morning, one started to vomit followed by another. Both of them had pastry in restaurant. Causative organism is? (NEET 2021)

A. Vero toxin
B. Bacillus cereus
C. Staph aureus
D. None of the above

2. SSSS [Staphylococcal Skin Scalded Syndrome] 00:35:08

- D/t Exfoliative / Enterotoxin A & B
- In children SSSS is K/a Ritter's disease
- In adults its K/a TEN (Toxic Epidermal Necrolysis)

3. TSS [Toxic Shock Syndrome]

- D/t TSS Toxin aka Enterotoxin F and it is also called Pyrogenic Exotoxin C
- TSS Toxin is a super antigen
- Super antigen activates > 20% lymphocytes Causing massive Lymphokine release resulting in Shock
- Treatment of shock
  - I.V fluids + Clindamycin (Clindamycin is having anti toxin action in this case)
- Cause: D/t usage of contaminated vaginal tampons during menses

4. Other Diseases associated with S. aureus 00:40:40

- Cellulitis
- Bullous Impetigo
- Carbuncles, Furuncles
- Acute osteomyelitis
- Acute mastitis
- Post op. parotitis
- Epidural / subdural abscess
- MCC of early prosthetic valve endocarditis: S. aureus
- MCC of native valve endocarditis: S.aureus
- MCC of Rt sided valve endocarditis in I/V drug abusers: S. aureus
- Associate with Pneumonia (have Pneumatocele)
- Botryomycosis: Mycetoma like lesion
- Tropical pyomyositis

MRSA [Methicillin Resistant Staph Aureus] 00:45:35

- DOC: Vancomycin (a/w Red Man Syndrome)
- Mechanism of resistance
  - Change in PBP to PBP_{2a} that is an alteration of the Transpeptidase sites by Mec A gene acquisition

→ Mec A gene 1, 2, and 3 associated with Nosocomial MRSA
→ Mec A gene 4, 5, and 6 associated with Community-acquired MRSA
→ Community-acquired MRSA is more dangerous than Nosocomial MRSA
→ Nosocomial infection: infection occurring after > 48 hr of hospitalisation
- Production of β Lactamases (also termed as Pencillinases)

- Lab diagnosis
  - Screening test using two Antibiotics: Oxacillin and Cefoxitin (Cefoxitin screening test better than Oxacillin)
    → Method
      - KBDDTon MHA (Muller-Hinton Agar) / NA
      - Add 4% NaCl
      - Keep the temperature of incubation at <37° C [30° -35°]
    → If S. aureus growth is Oxacillin resistant: 90% MRSA
    → If S. aureus growth is Cefoxitin resistant: 100% MRSA
  - ELISA for MEC A gene
  - PCR for MEC A gene [best screening test for MRSA]
  - Latex Agglutination Test for PBP2a

- Best way to avoid Nosocomial spread of MRSA: Handwashing
- Mupirocin ointment or Bacitracin ointment indicated for nasal carriers
- Transduction
  - M/c method of resistance transfer in S. aureus
  - Bacteriophage mediated

- Phage Typing 00:58:26
  - 23 types of phages are present which are specific for S. aureus strains
  - 80/ 81A: MC phage (Most commonly responsible for outbreaks in hospital)

STREPTOCOCCUS 01:05:55

Classification

Refer Table 2.1

Refer Table 2.2

- CAMP TEST: Christie Atkins Munch Peterson Test
- HHT: Hippurate Hydrolysis Test

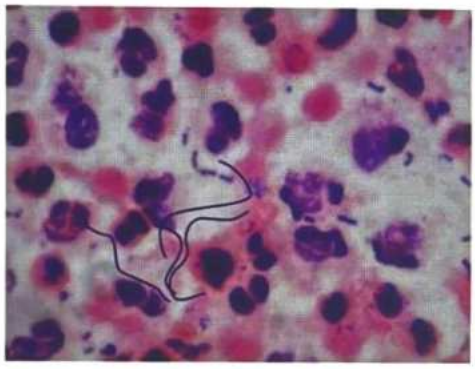
Previous Year's Questions

Q. Identify the organism on Bacitracin sensitivity disk test given below? (AIIMS -May - 2018)

A. Streptococcus pyogenes
B. Staph aureus
C. Clostridium
D. Corynebacterium

BETAHEMOLYTIC STREPTOCOCCI

1. GROUP- A STREPTOCOCCI: Streptococcus Pyogenes 01:13:36



- Gram positive cocci
- Catalase -ve
- Arranged in violet colored chains on gram staining
  - S. salivarius: Has the Longest chain
- Culture media: CVBA[Crystal Violet Blood Agar]
  - Inhibits staph aureus
  - Produce β Hemolysis
  - Pinpoint size colonies
- Transport media: Pike's media
- Diseases 01:16:59
  - Cellulitis
  - Impetigo (Honey colored crust)
  - Erysipelas (Lymphatics involvement)
  - Puerperal sepsis
  - Necrotising fasciitis because of which it's also termed as flesh eating bacteria
  - Acute pharyngitis
  - Quinsy / Peritonsilar abscess
- Super antigen of S. Pyogenes: Scarlet fever toxin or Erythrogenic toxin A, B & C or Pyrogenic toxin A, B & C
  - Activates > 20% lymphocytes causing shock

- Non Suppurative Complications of S. Pyogenes 01:19:48
  - Acute Rheumatic Fever
  - Acute Glomerulonephritis

Acute Rheumatic Fever	Acute Glomerulonephritis
• H/O sore throat always • Any group A strain • Immune response marked • Complement levels are normal • Lifelong prophylaxis of penicillin required • ASO Titers:> 200 Todd units	• H/O pyodermic infection / Sore throat • 49, 53-55, 59-61 • Mild to moderate immune response • Complement level decreases and then becomes normal • Spontaneous resolution • Anti DNAase B:> 300-350 units

2. GROUP- B STREPTOCOCCI: Streptococcus Agalactiae 01:23:47

- Normal Commensal of the birth canal
- MCC of Neonatal Meningitis worldwide
- Diseases
  - Early Neonatal meningitis
    - → Time period: 0-6 days
    - → Factors responsible: Prematurity / prolonged labour
    - → Initially pneumonia later on meningitis
    - → High mortality (4.7%)
    - → Serogroups: 1_a, 1_b, 2, 3, 4
  - Late Neonatal meningitis
    - → 7-90 days
    - → Less mortality (2.8%)
    - → Serogroups: Type 3 (MC)
  - Pneumonia later on leads to meningitis
  - Bacteremia later on leads to meningitis

- CAMP test +ve: Enhanced β Hemolysis
- HHT [Hippurate Hydrolysis Test] is +ve

3. GROUP-D STREOPTOCOCCUS 01:33:39

- Contains Enterococcus and Non-Enterococcus

Non-Enterococcus

- S. bovis
- S. gallolyticus (Associated with Colon cancer)

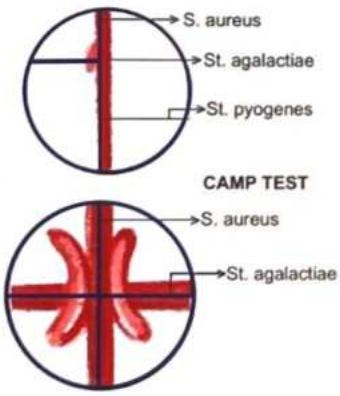
Enterococcus

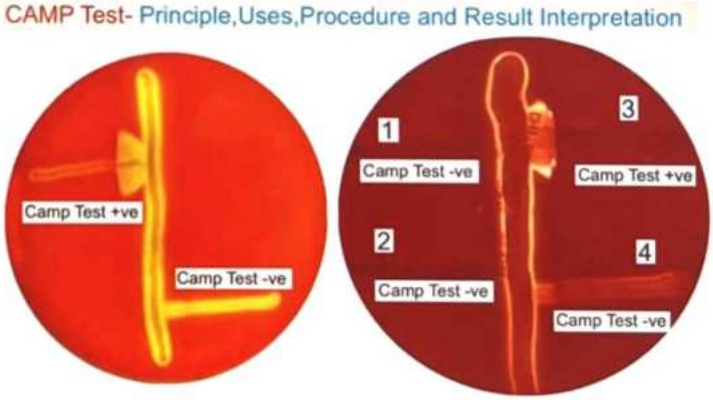
- Include
  - E. Faecalis
  - E. Faecium
- E. Faecalis and E. Faecium resist
  - 6.5% NACL
  - PH = 9.4
  - Can grow in 40% Bile
  - Temperature > 60° C minutes for minutes
- Normal commensals of gut
- Causes Sx wound infection
- Lt. sided valve endocarditis
- DOC
  - Pencillin + Aminoglycoside
  - Vancomycin [2nd line]
- Vancomycin resistant Enterococci
  - Doc: Linezolid
  - They became resistant by van gene acquisition [Van A-Van H]
  - They change the structure of the cell wall from D-alanine-D-alanine to D-alanine-D-lactate or D-alanine-D-serine
- Culture media 01:39:26
  - Bile Aesculin Agar: Black Colored Colonies
  - On Gram Staining: Spectacle like appearance is seen

ALPHA HEMOLYTIC STREPTOCOCCUS

- Contains a viridians group which includes

Camp Test 01:28:30





Enhanced/ Butter Fly Zone of Hemolysis

- St. Viridans: leads to SABE, Late-Onset Prosthetic Valve Endocarditis
- St. Sanguis
- St. Mutans

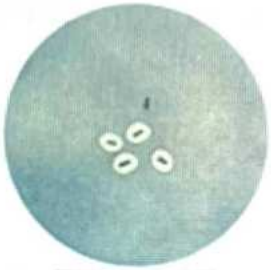
- Dental caries caused by St. Mutans > St. Sanguis

1. St. Viridians

- Gram Positive Cocci
- BIO negative
  - **B**- **B**ile Soluble negative
  - **I** - **I**nulin fermenter negative
  - **O**- **O**ptochin sensitivity negative

2. Streptococcus pneumoniae [pneumococcus]

01:44:10



Diplococci

- Gram positive Diplococci
- Lanceolate [Flame Shaped]
- Virulence Factors
  - Capsular polysaccharide
  - Autolysin aka Amidase enzyme
  - Pneumolysin: membrane damaging toxin
    → Autolysin and pneumolysin play a role in making the Carom coin appearance of colonies aka Draught Man Appearance
  - PSPA: Pneumococcal Surface Protein A
  - IgA protease
  - Pneumococcal surface protein C
  - Adhesins
- On the basis of capsular polysaccharides its divided into 90 types and this typing is k/a SSS typing [Specific Soluble Substance Based Typing]
- Biochemical reaction
  - **B** - **B**ile soluble +ve
  - **I** - **I**nulin fermenter +ve
  - **O** - **O**ptochin sensitive +ve

Important Information

- BIO

- Diseases
  - **C** - **C**onjunctivitis
  - **O** - **O**titis Media (MC)
  - **M** - **M**eningitis (MCC of meningitis in adults)
  - **P** - **P**neumonia
  - **S** - **S**inusitis

Important Information

- COMPS
  - Arthritis
  - MCC of community Acquired Pneumonia (Lobar Pneumonia, Type 3 causes most virulent type of pneumonia)

Previous Year's Questions

Q. An HIV +ve patient presented with fever, chills. His CD4 count is < 55. Examination findings of consolidation in right intra-scapular area with X-ray showing right lower consolidation. Most probable diagnosis of the infection is? (NEET 2021)

A. Staphylococcus aureus
B. Pneumococcus
C. Pseudomonas aeruginosa
D. Pneumocystis jiroveci

- Vaccines
  - 23 Valent Polysaccharide Vaccine
    → Used for splenectomised individuals as they are prone to capsulated organism infections
  - 13 Valent Conjugate Vaccine: preferred for Children
- Pneumococcal vaccine Indicated:
  - Before doing a cochlear implant
  - Individuals undergoing CSF shunting
- DOC for Pneumococcus: Penicillin
  - Penicillin resistance is due to PBP changing to PBP_{2b}

GRAM NEGATIVE COCCI

01:58:57

Refer Table 2.3

MENINGOCOCCI

02:05:51

- Gram –ve cocci
- Catalase +ve, oxidase +ve
- IP: 3-4 days
- Capsulated
- On the basis of capsule polysaccharides Meningococci is divided into13 Serogroups
  - A, B, C, Y, W, E, X: most important Serogroups
  - A, B, C: most infective Serogroups

- Virulence factors
  - Capsular polysaccharide
  - IgA Protease
  - Lipo Oligosaccharide
  - OMP [outer membrane Proteins]
  - Pili
- 5-15 % persons are normal nasopharyngeal carriers
- These nasopharyngeal carriers Spreads the infection by Droplet spread which has a Pulmonary involvement and then through Haematogenous route it goes into Meninges causing Meningitis

Meningococcal meningitis

- Clinical features
  - Fever
  - Vomiting (Projectile)
  - Headache
  - Neck rigidity (Kernig sign and Brudzinski sign)
  - Rash present
- Lab diagnosis
  - CSF examination: 3 samples are taken one for the Microbiology department, Pathology (for cytology), and Biochemistry (for Proteins & Sugars) respectively
  - Culture on
    - → MHA
    - → Thayer Martin Media (Modified Chocolate Agar)
  - Gram staining shows
    - → Pink coloured cocci
    - → Arranged extra/ intracellular
    - → Lens shaped
  - Blood Culture
  - Latex card agglutination test
  - PCR for meningococci (CSF/ Blood)
- Other manifestation
  - Non blanching rash >80%
  - Septicemia: because of Endotoxic activity of Lipo Oligo Saccharide
  - Water house Friedrichsen Syndrome:
    - → DIC causing B/L adrenal hemorrhagic necrosis + purpuric rashes
    - → Later on leads to death if not well managed
    - → It is the Most dreadful complication
- In C_5- C_9 (MAC) complement deficiency: Struggles from Recurrent Neisserial infections
- Treatment
  - DOC for carriers: Ciprofloxacin > Rifampicin
  - DOC for cases: Penicillin > 3rd gen cephalosporin
- Vaccines 02:20:05
  - MCV-4 (Meningococcal Conjugate Vaccine)
    - → Active against A, C, Y, W
    - → No proper vaccine is available For group B because Group B capsule is Poorly immunogenic

GONOCOCCI 02:23:54

- Catalase & Oxidase +ve
- Typing
  - Pili based typing
  - AUXO typing [Nutrient based]
- IP: 2-7 Days
- Causes: Gonorrhoea
- Virulence factors
  - **C** - **C**apsular polysaccharide
  - **I** - **I**g A_1 protease
  - **P** - **P**ili
  - **L** - **L**ipo Oligo saccharide
  - **O** - **O**MP [outer membrane proteins], Opacity associated protein

Important Information

- CIPLO
  - Transferrin and Lactoferrin

- Diseases
  - In Male
    - → Acute Urethritis causing Purulent discharge which looks like semen (flow of seeds)
    - → Lateron, it leads to Water-Can Perineum
  - In Females
    - → It causes Cervicitis having as Mucopurulent discharge
    - → In a Pregnant female having gonococcal infection, the newborn can present with Ophthalmia neonatorum
- Lab diagnosis
  - 3 Type of swabs are taken
    - → Urethral Swab
    - → Endo cervical Swab
    - → Eye Swab
  - Culture Media: Thayer Martin Media and Modified New York Agar
  - Gram Staining: Pink colored cocci, kidney shaped, Arranged mostly intracellular
  - RCUT: Ferments only glucose
  - PCR: For gonococci
- Complications 02:33:24
  - Water can perineum
    - → Occurs in males (Urethritis)
    - → Multiple fistulas in perineal region
  - Fitz Hugh Curtis Syndrome
    - → Perihepatitis
    - → Also a/w Chlamydia
  - Polyarthritis which later on changes to Suppurative arthritis
  - Deep Gonococcal Infection (DGI)

- Treatment
  - Doc : 3rd gen cephalosporin
    → This organism recruit Plasmid thereby ↑ Resistance against 3rd gen cephalosporins
- Ophthalmia neonatorum is more a/w Chlamydia than gonococcus

Table 2.1

α Hemolysis	β Hemolysis (β-HS)	γ Hemolysis

• S. Pneumoniae
• S. Viridans
	• Based on carbohydrate 'C' in the cell wall Classified into A to V K/a Lancefield Classification
	• A is divided into 80-100 groups based on 'M' protein termed as Griffith typing (Mnemonic: Lucknow City Good Morning)
	• Groups
	  ○ Group A: S. Pyogenes
	  ○ Group B: S. Agalactiae
	  ○ Group C: S. Equisimilis
	  ○ Group D: Enterococcus/ NE
		• Enterococcus
		• Faecalis
		• Faecium

Table 2.2

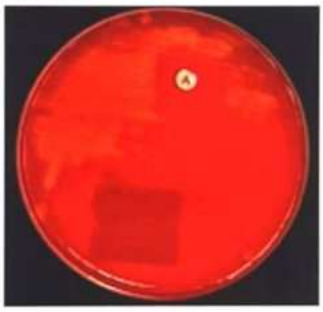
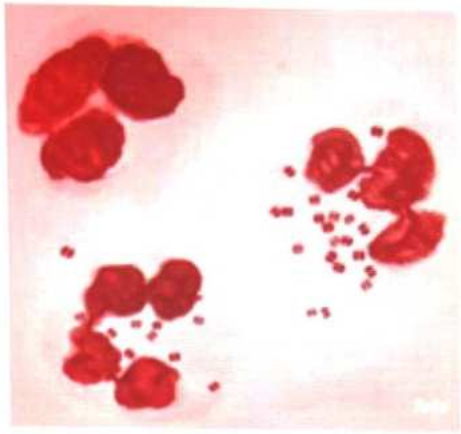
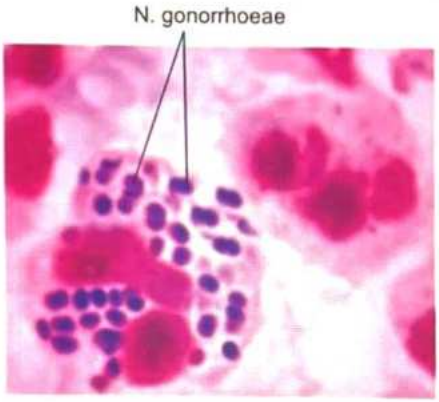
Tests	Group A Streptococci	Group B Streptococci
• Bacitracin test	• Sensitive 	• Resistant
• CAMP test	• Negative	• Positive
• HHT	• Negative	• Positive
• PYR Test	• Positive	• Negative

Table 2.3

Meningococci	Gonococci

• Capsulated
• Lens shaped
• RCUT (Rapid carbohydrate utilization test)
  ○ Ferments Glucose & maltose
• Does not recruit Plasmid
• Can be both Extra/Intracellular
	• Non capsulated but Some strains are capsulated
	• Kidney shaped
	• RCUT (Rapid carbohydrate utilization test)
	  ○ Ferments only Glucose
	• Recruits plasmid for resistance
	• Mainly intracellular

	
• Colony on Thayer martin media: Mostly circular • Natural habitat: Nasopharynx	• On Thayer martin media colony have: Irregular margins • Genital tract

CLINICAL QUESTIONS

Q. 1 A patient presented with complains of red sore throat, cough, hoarseness and redness of eyes. The condition was diagnosed as pharyngitis which is caused by Streptococcus pyogenes. C-carbohydrate in Streptococcus pyogenes contributes to which of the following property?

A. Lancefield classification
B. Phagocytic inhibition
C. Toxin production
D. Haemolysis

Solution:

- C carbohydrate in Streptococcus pyogenes contributes to **Lancefield classification.**
- **Option 1-** β-Hemolytic Streptococci are classified by Lancefield serological classification into groups **based on nature of "C carbohydrate antigen**:
- **Lancefield groups** (A-V excluding I, J):
- Group A: Streptococcus pyogenes
- Group B: Streptococcus agalactiae
- Group C: Streptococcus equisimilis
- Group D and so on.
- **Streptococcus pyogenes** is classified into serotypes by **Griffith typing: Based on M protein**. More than 100 serotypes are present.
- **Option 2 -** C-Carbohydrate antigen **doesn't have any anti-phagocytic role**.
- **Option 3-** C-Carbohydrate antigen doesn't have any correlation with toxin production.
- **Option 4-** On the basis of **hemolysis**, Streptococci can be divided into 3 groups:-
  - **α** Hemolytic : S. pneumoniae, S. viridans
  - **β** Hemolytic: Divided further on the basis of carbohydrate C antigen of cell wall into **Lancefield groups** (A-V excluding I, J). Eg: S. pyogenes
  - **γ** Hemolytic: Enterococcus

Q.2 A 32-year-old presented with acute, painful, edematous, and erythematous skin lesions of his lower back, that rapidly progressed to the right knee. The patient underwent surgery, and a diagnosis of necrotizing fasciitis was confirmed by tissue biopsies. It is usually caused by:

A. Beta hemolytic streptococci
B. Staphylococcus aureus
C. Clostridium perfringens
D. Pneumococcus

Solution:

- The spectrum of infections of the deep soft tissues ranges from localized bacterial, viral and parasitic lesions to rapidly spreading, tissue destructive infections such as necrotizing fasciitis and myonecrosis.

A. **Pyomyositis:-** It is common in the tropics, therefore also called tropical pyomyositis. It is a localized infection of skeletal muscles. It is caused most commonly by staphylococcus aureus.

B. **Necrotizing fasciitis:** It is an infection of the deeper layers of skin and subcutaneous tissues, easily spreading across the fascial plane within the subcutaneous tissues. There are two types of necrotizing fasciitis:

- **Type 1:-** It is a polymicrobial infection, i.e. mixed aerobic and anaerobic infection. It occurs most commonly after surgical procedures in diabetic patients or in those who have peripheral vascular disease.
- **Type 2:-** It is caused most commonly by streptococcus pyogenes (Group A beta-hemolytic streptococci).

C. **Clostridial myonecrosis (Gas gangrene):-** It is characterized by rapid and extensive necrosis of muscle accompanied by the gas formation and systemic toxicity. It is caused by C. perfringens (most common), C. novyi, C. septicum and C. histolyticum. It is also called type III necrotizing fasciitis.

Q.3 A 23-year-old male presents with fever and altered sensorium for two days with the following rash on legs. Her BP is 70/50 mm Hg and Neck stiffness is present. Lumbar puncture reveals cloudy CSF with 4200 cells/uL, Protein level 198 and Glucose of 21mg/dL. Which of the following correctly describes the organism causing this condition?

A. Gram-negative diplococci (kidney shaped), oxidase positive
B. Gram-negative diplococci, ferments glucose and maltose
C. Gram-positive cocci catalase negative, bacitracin sensitive
D. Gram-positive diplococci (lanceolate), catalase negative, optochin sensitive

Solution:

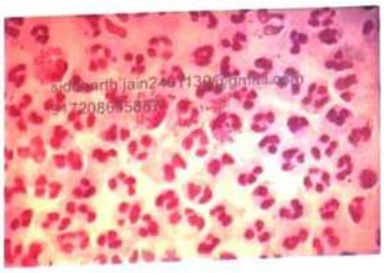
- Gram-negative diplococci, ferments glucose and maltose correctly describe the organism causing this condition.
  - The above history and clinical presentation is suggestive of **Meningococcal Meningitis.**
  - Meningococcal Meningitis - caused by **Neisseria meningitidis.**

Option 1

Neisseria gonorrheae -

- Gram-negative diplococci
- kidney shaped
- oxidase positive
- Manily non capsulated
- Ferments glucose only
- Mainly intracellular

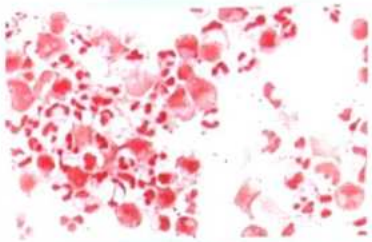
Causes urethritis, cervicitis, opthalmia noenatorum.



Option 2

Neisseria meningitidis -

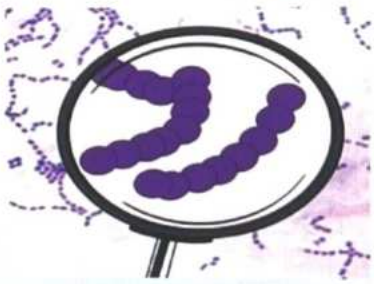
- Gram-negative diplococci
- Capsulated
- Lens shaped
- Ferments glucose and maltose
- Extracellular/intracellular



Option 3

Streptococcus pyogenes:

- Gram-positive cocci
- in chains
- catalase negative
- bacitracin sensitive



Option 4

Streptococcus pneumoniae:

- Gram-positive diplococci
- lanceolate or flame shaped
- catalase negative
- optochin sensitive

3 GRAM POSITIVE BACILLI PART-1

CORYNEBACTERIUM DIPHTHERIA 00:00:19

- Also termed as Kleb Loffler's Bacillus
- Having Metachromatic granules/ Polymetaphosphate granules/ Volutin Granules/ Babes Ernest granules
  - These are Energy deposits

Stain

- P-Ponders Stain
- A-Albert's Stain
- N-Neisser Stain

How to remember

- PAN

Chinese letter pattern

- On Gram Staining / Albert Staining: Cuneiform arrangement can be seen

Biotypes

Refer Table 3.1

Disease: diphtheria

- Characterized by formation of a Pseudo membrane (Greyish white membrane)
- MC site: Faucial
- Most dangerous: laryngeal
- Complication :Bull Neck (Neck edema)
- Chronic complication: polyneuropathies and myocarditis(more dangerous)

Previous Year's Questions

Q. Which is correct about Diphtheria membrane? (FMGE - June - 2019)

A. Grey membrane that bleeds on removal
B. Grey membrane that does not bleeds on removal
C. Grey pseudo membrane that bleeds on removal
D. Grey pseudo membrane that does not bleeds on removal

Lab Diagnosis

- Take Throat swab: gram stain and culture
  - Culture on
    - → LSS (Loffler's Serum Slope) For early diagnosis (6-8 hrs)
    - → PTA: Definitive diagnosis best selective media
  - Other culture media's
    - → H-Hoyle's media
    - → M-Macleod's media
    - → T-Tins dale media

How to remember

- HMT

- Other tests for lab diagnosis
  - Elek Gel Precipitation Test
  - PCR For tox gene
  - ELISA For toxin Production
  - Schick test: It determines susceptibility of an individual to Diphtheriae
    - → If positive: needs immunization
    - → If negative: already immunized
    - → Neutralization test
- Diphtheria is Toxemia not Bacteremia

Diphtheria toxin

- Strong Exotoxin
- Made of 2 component A and B
  - B: Helps in Binding to cell
  - A: ADP Ribosylation of EF - 2 which causes inhibition of protein synthesis leads to cell death

- Needs iron for its action 0.1mg/dl
- Lysogenic Conversion: Every corynebacterium Diphtheriae is not pathogenic, But made pathogenic by - phage tox gene

Cutaneous diphtheria 00:20:59

- Punched out ulcerative lesion
- Not toxin mediated

- Rx
  - ADS (Anti Diphtheric Serum)
    → In severe cases: 80,000-1,00,000 units
  - Antibiotic
    → Penicillin or Erythromycin
    → Better to give in initial stage of disease i.e., before toxin production
    → For carriers: Erythromycin

- Protective Titers against Diphtheria
  - Antitoxin titer 0.01 unit/ml (Following vaccination)

Previous Year's Questions

Q. Diphtheria vaccine is type of? (AIIMS - June - 2020)

A. Live attenuated vaccine
B. Killed vaccine
C. Toxoid
D. Polysaccharide vaccine

Non corynebacterium Diphtheriae 00:24:36

- C. jeikum
  - MDR isolate
- C. minutissimum
  - Causes Erythrasma which is having a Coral red fluorescence
- C. urealyticum
  - Produces Pyelonephritis
  - Leads to formation of Struvite stones
- C. parvum
  - It is an Immuno modulator

BACILLUS ANTHRACIS 00:29:11

- GPB [Gram Positive Bacilli], Capsulated
- Non motile
- Polypeptide
- Spore bearing (Box Car Appearance)
- Non-Bulging spore
- Used as Bioterrorist weapon Category A

Previous Year's Questions

Q. Bacillus Anthrax is? (FMGE - June - 2019)

A. Gram positive cocci in cluster
B. Gram positive rods with square ends
C. Gram positivebacilli with spherical ends
D. Gram negative cocci in cluster

Anthrax toxin

- Made of Tripartite Toxin
  - **E** - Edema Factor
  - **P** - Protective Factor
  - **L** - Lethal Factor

How to remember

- EPL

- Acts Synergistically: ↑CAMP

Capsule

- Made of Polypeptide or Polyglutamate
- Mc Fadyean's Reaction
  - Demonstrates capsule
  - Culture growth+ Loffler'sMethyl Blue→ capsular stains blue

Culture media

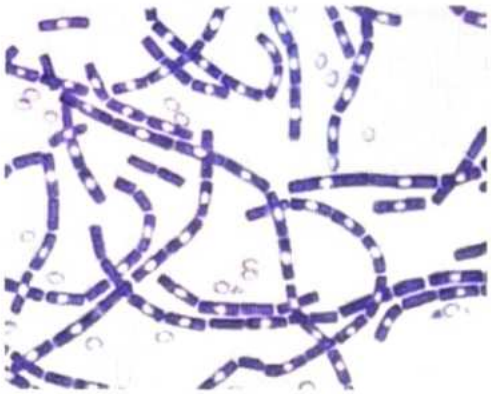
- **PLET** medium
  - **P** - Polymyxin
  - **L** - Lysozyme
  - **E** - EDTA
  - **T** – Thallus acetate

Lab diagnosis

- **M** -Medusa head appearance
- **I** -Inverted fir tree appearance (seen in Gelatin stab culture)
- **S** -String of pearl appearance (seen in Penicillin agar)
- **B** -Bamboo stick appearance (seen in Gram stain)

How to remember

- MIS Bamboo



Bamboo Stick Appearance on Gram Staining

Diseases

a. Cutaneous anthrax
- Hide porter's disease
- Black Eschar surrounded by Edema
- Later on it turns into pustule with non pitting edema K/a Malignant Pustule

b. Pulmonary anthrax
- Wool sorter's disease
- Occupational Hazard
- Hemorrhagic Pneumonia which is also termed as hemorrhagic mediastinitis
- Prevented by duckering that is Pre treat the wool by Formaldehyde

c. Intestinal anthrax
- Spread through eating contaminated wild animal meat ingestion

Tests for Bacillus Anthracis
- Ascoli's Thermo precipitation test: For Anthrax antigen
- PCR: Latest test for B. Anthrax

Treatment
- DOC: Ciprofloxacin & Clindamycin x 60 days

BACILLUS CERUS

00:44:46

Emetic strain	Diarrheal strain
• IP: 1 to 6 hrs (mimics IP of S. aureus food poisoning) • Chinese fried rice • Heat stable toxin • In action it mimics heat stable toxin of E. Coli • MOA: ↑ CGMP	• IP: 8-16 hrs • Contaminated meat & milk • Heat labile toxin • Mimics Heat Labile toxin of E. Coli • MOA: ↑ CAMP

Culture media
- MYPA [Mannitol Yolk sac Polymyxin Agar]

CLOSTRIDIUM

00:53:11

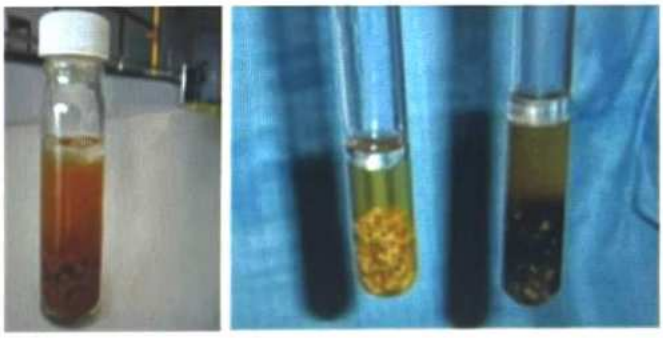
- GPB, Bulging spore
- Non capsulated
- Motile: stately [Slow Form of swarming]
  - Exception: Cl. perfringes [capsulated, non motile]
- It's an Obligate anaerobe

Important Information

Obligate Anaerobes
- A - Actinomyces
- B - Bacteroides
- C - Clostridium

Culture media

Cl. perfringens | Cl. tetani



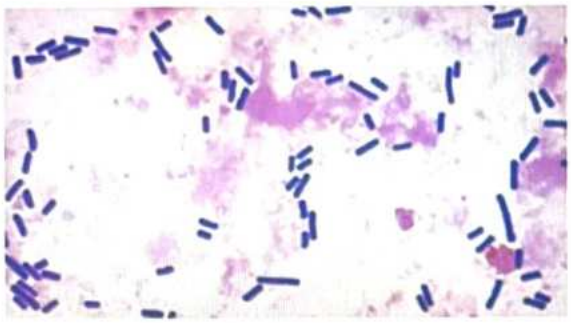
- Robertson Cooked Meat Media
  - If meat pieces turn PINK: Sacchrolytic
    - → Cl. Perfringens
    - → Cl. Novy
    - → Cl. Septicum
    - → Cl. Botulinum C, D & E
  - If meat pieces turn BLACK: Proteolytic
    - → Cl. Tetani
    - → Cl. Botulinum A, B & E

Methods of Anaerobiosis
- McIntosh and Filde's jar
- Closed jar with candle (Obsolete)
- Gas pak (latest)
  - Chemical method of Anaerobiosis
  - MC used

Spore arrangement
- Terminal & round spore aka Drumstick Appearance: Cl. Tetani

- Sub terminal spore: Most clostridia
- Central spore: Cl. bifermentans

1. CLOSTRIDIUM PERFRINGENS/ WELCHII



Clostridium Perfringens

- 13 major & minor toxins are present
- α, β, epsilon, theta, iotaare more important
- On the basis of these toxins we divide them into Type A - E
- Most infections are caused by combination ofType A; α toxin

Disease

a. Gas Gangrene/ Clostridial Myonecrosis 01:03:44

- IP: 1-2 days
- C/F
  - Pain
  - Crepitus: due to Gas in the deep muscle compartments (because of low oxidation reduction potential)

- Treatment
  - For Initial lesion: Surgical debridement & Antibiotics
  - For advanced case: Amputation of the limb & Antibiotics

Important Information

Gas Gangrene caused by

- P - Perfringens (Brick shaped)
- N - Novyii
- S - Septicum (Boat shaped aka citron bodies)

- Gram staining
  - Brick shaped
  - Citron bodies

- Lab diagnosis
  - RCM: Sacchrolytic
  - PCR for Cl. Perfringens

b. Food Poisoning 01:09:52

- Type A; toxin
- D/t contaminated meat products
- IP: 8-24 hrs
- Involves Heat labile Enterotoxin but spores are Heat Resistant

c. Pigbel 01:11:25

- D/t uncooked/ contaminated pork consumption
- Causes necrotizing enteritis
- D/t toxin Type C

Reactions Concerned with Cl.perfringes

- **S**enior - **S**enior fermentation of litmus milk
- **N** - **N**agler's reaction
- **T** - **T**arget Hemolysis (Double zone of Hemolysis)
- **R** - **R**everse camp test

How to remember

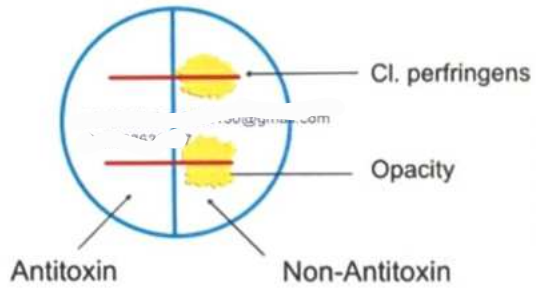
- Senior NTR

Target zone Hemolysis

Stormy Ferm-entation

Target Zone Hemolysis Stormy Fermentation

a. NAGLER's reaction

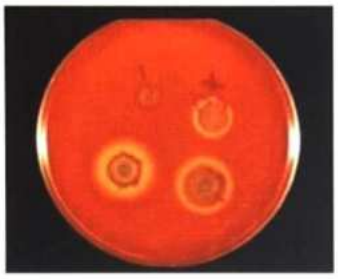


- D/t Toxin -Lecithinase or Phospholipase
- It is a neutralization reaction (toxin- Antitoxin reaction)
- Done on Egg Yolk Agar

b. Stormy fermentation in litmus milk

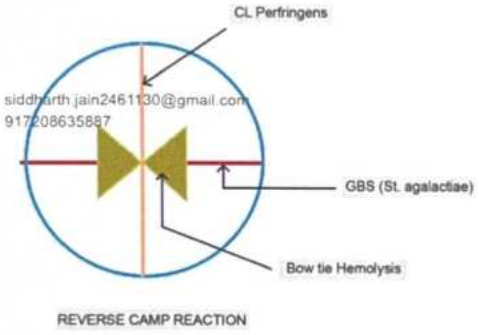
- Immense fermentation of litmus milk

c. Target Hemolysis (Double zone of Hemolysis)



- Inner zone: d/t Theta-toxin
- Outer zone: d/t -toxin

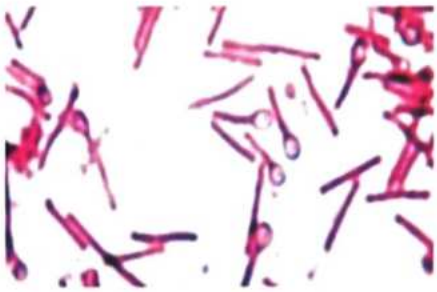
d. Reverse CAMP test



- Bow Tie Hemolysis with St. agalactiae

2. CLOSTRIDIUM TETANI

01:20:02



- Tetanus (8th days disease)
- IP: 6-10 days
- If IP < 6 days: Grave prognosis

Toxins

a. Tetanolysin
- Hemolysis
- O_2 labile
- Resemble to toxin of S. Pyogenes& S. pneumoniae

b. Tetanospasmin
- More dangerous
- Neurotoxin
- Causes presynaptic inhibition of Glycine/ GABA (inhibitory neuro- transmitters) leads to Constant stimulation causing Lock Jaw (Trismus)
- If not controlled it will lead to Opisthotonus Posture (Bad prognosis) and there will be ↑Deep tendon reflexes

Neonatal Tetanus
- India is free of neonatal tetanus d/t
  - Clean practices
  - Antenatal immunization
  - Hospital deliveries

Lab Diagnosis
- Excised tissue fragment + RCM: Meat pieces turn Black
- PCR for Cl. Tetani
- Mouse toxigenicity testing

Prevention of tetanus after injury

	Simple wound	Other wound
Category A	Nothing required	Nothing required
Category B	Toxoid 1 dose	Toxoid 1 dose
Category C	Toxoid 1 dose	Toxoid 1 dose + HTIg
Category D	Complete Toxoid dose	Complete Toxoid dose + HTIg

- Category A: TT/Booster in last 5 yrs
- Category B: Complete course in last 5-10 yrs
- Category C: Complete course beyond 10 yrs
- Category D: Unknown status

Previous Year's Questions

Q. A30-year-old man reports four hours after acquiring a clean wound without laceration. He has received TT vaccination 10 years back. What do you advice regarding tetanus prophylaxis? (NEET - Jan - 2020)

A. Single dose of TT
B. Full case of TT
C. I dose TT and TIG
D. No vaccination needed

Treatment

- DOC for Tetanus: Metronidazole
- Antitoxin Titers: > 0.01 units/ml

3. CLOSTRIDIUM BOTULINUM

01:33:33

- Produces the most potent toxin on earth

Toxins

- Neurotoxins (main): most potent toxin on earth
- Botulinum Toxin
  - Zn dependent protein having a Heavy Chain and Light Chain
  - It has an Anticholinergic action which leads to Flaccid paralysis
  - Acts on
    - → Most common nerve terminals
    - → NMJ, Post ganglionic parasympathetic nerve ending
    - → Peripheral ganglia
  - BT binds to Ach receptor and blocks release of Ach which leads to Flaccid Paralysis
  - Used in: Strabismus, Blepharospasm, Myoclonus
  - BT can be produce by
    - → **B** - **B**utyricum
    - → **B** - **B**aratti
    - → **A** – Argentimense

How to remember

- BBA

- C_2 (Enterotoxin)
  - A/w canned food poisoning
  - Associated findings
    - → Dysphagia
    - → Dysarthria
    - → Diplopia
  - Only food poisoning associated with constipation
  - Decrease in DTR

Infant botulism

01:40:02

- A/w Sporulated honey ingestion
- Clinical Features: Flaccid paralysis so its K/a Floppy Infant Syndrome

Lab Diagnosis

- RCM: Meat pieces will turn both Pink and Black because it is both Proteolytic as well as Saccharolytic
- PCR for Cl. Botulinum
- Cultured on blood agar under anaerobic conditions

4. CLOSTRIDIUM DIFFICLE

01:42:28

- Difficult to isolate
- Clostridium Difficle Associated Diarrhea (CDAD)
  - If not Rx in time, leads to Pseudomembranous colitis
  - Normal commensals of gut (Cl. Difficle)
  - A/w long term used of 3rd generationcephalosporin, Clindamycin
  - When Normal flora destroyed, Cl. Difficle attaches to gut and it irritates and lead to Diarrhea
  - If it's not managed properly will lead to Pseudomembranous colitis which can present with bloody diarrhea
  - DOC: Oral Vancomycin

Lab diagnosis

- Toxin Demonstration in stool [Entero toxin (A) + Cytotoxin (B)]
  - Demonstrated on Hep 2 cell lines (commonly used for viral isolation)
- PCR for Tox gene
- ELISAfor toxin production
- Culture on CCFA (Cefoxitin Cycloserine Fructose Agar)
- Sigmoidoscopy / Colonoscopy to visualize the ulcers

Previous Year's Questions

Q. Diagnosis of Cl difficile infection is made by which of the following methods? (AIIMS - Nov - 2017)

A. Toxin gene detection by polymerase chain reaction (PCR)
B. Culture
C. Enzyme – linked immunosorbent assay (ELISA)
D. Nagler's reaction

Table 3.1

		Colonies on PTA	Toxigenic strains	Virulence	Occurrence
1. Gravis	Daisy's	Daisy Head Appearance	100%	Severe	Epidemic
2. Intermedius	Frog eggs are	Frog Egg Appearance	95-99%	Moderate	Epidemic
3. Mitis	Poached	Poached Egg Appearance	80-85%	Mild	Endemic

CLINICAL QUESTIONS

Q.1 A microbiologist has received two culture plates of Corynebacterium diphtheriae and Corynebacterium pseudotuberculosis. He has to culture and differentiate among them. Which of the following test should be conducted in order to biochemically differentiate C. diphtheriae from C. pseudotuberculosis?

A. Catalase test
B. Urease test
C. Oxidase test
D. Tellurite Reduction

Solution:

- C. pseudotuberculosis is **urease positive and** is thus differentiated from C. diphtheriae.
- C. pseudotuberculosis causes caseous lymphadenitis in sheep and goats.

NON-CORYNEBACTERIUM DIPHTHERIAE:

1. C. parv[illegible]mmuno modulator
2. C. jeokum → MDR isolate
3. C. minutissimum → causes Erythrasma, coral red fluoresscene is seen.
4. C. pseudotuberculosis → causes animal diseases, also known as **PREISZ NOCARD BACILLI.**

Optioin 1: Corynebacterium - catalase positive.

Option 3: Oxidase positive organisms are:

- Pseudomonas aeruginosa
- Burkholderia species
- Vibrio
- Pasteurella multocida
- Brucella
- Neisseria
- Haemophilus spp
- Campylobacter

Option 4: Potassium tellurite agar (PTA) is the selective media used for **C. diphtheriae.**

Q.2 A 12-year-old boy presented to the clinic with stiff and painful muscles, jaw cramping, trouble in swallowing, jerky movements, and fever. On processing the sample and staining, the microscopy showed spores that had a drum stick appearance. The mechanism of action of the toxin produced by the pathogen involved in this condition is:

A. Inhibition of GABA release

B. Inhibition cAMP

C. Inactivation of Ach receptors

D. Inhibition of cGMP

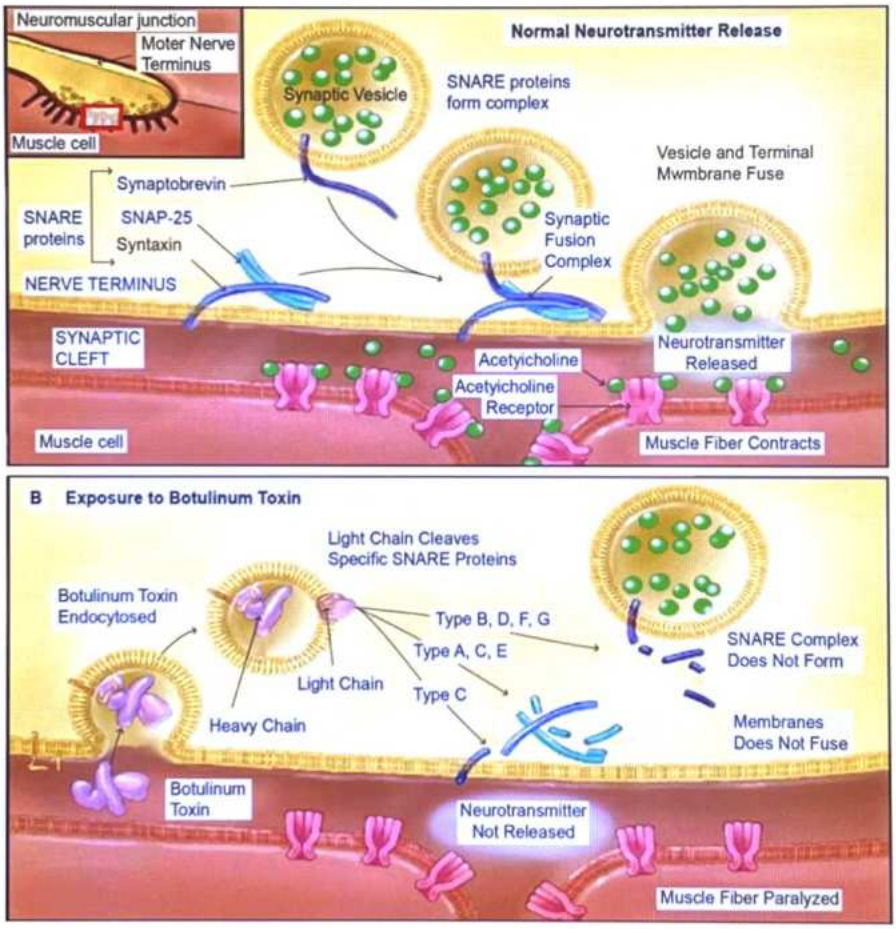
Solution:

- Mechanism of action of toxin is **Inhibition of GABA release.**
- **Tetanospasmin** produced by Cl. tetani is responsible for the above clinical condition.
- **Cl. tetani produces:-**

1. **Tetanolysin:** heat labile, oxygen labile hemolysin. It plays no role in the pathogenesis.
2. **Tetanospasmin** or tetanus toxin (TT) : neurotoxin responsible for the pathogenesis of tetanus:
   - It is oxygen stable but heat labile; coded by plasmid.
   - **Mechanism of action**: Toxin acts **pre-synaptically** at the inhibitory neuron terminals and prevents release of inhibitory neurotransmitter GABA and glycine → leads to **spastic muscle contraction.**

Clinical features:

- Lock Jaw (**Trismus**)
- **Opisthotonus** Posture (Bad prognosis)
- ↑ Deep tendon reflexes
- **Strychnine** poisoning has a similar mechanism except that it acts **post-synaptically.**
- **BT (Botulinum Toxin) blocks the release of acetylcholine** in neuromuscular junction, which leads to **flaccid paralysis.**



4 GRAM POSITIVE BACILLI PART-2

MYCOBACTERIUM TUBERCULOSIS 00:00:14

- 20% Acid fast
  - Ability to resist decolorization
  - Depends on Mycolic Acid content
- Virulence Factors
  - Cord factor
  - Lipo arabino mannan
    - → Helps in attachment
    - → LAM Ag detected in urine
    - → These 2, prevent phagolysosomal fusion
  - HSP (Heat Shock Protein)
- Pathogenesis
  - Human (Pulmonary TB)
  - Milk source
- Mode of Transmission: Inhalational mode
  - By droplet nuclei
  - Generated during coughing & sneezing

Previous Year's Questions

Q. Lipoarabinomannan (LAM) assay in urine is used for screening of? (AIIMS - Nov - 2017)

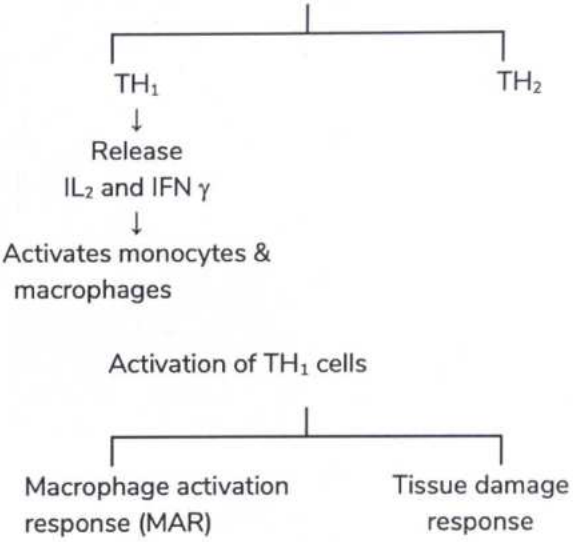
A. Mycobacterium tuberculosis
B. Pneumocystis Jiroveci
C. Histoplasma Capsulatum
D. Cryptococcus neoformans

Sequence of Pathological Events

- Droplet nuclei (Tubercle bacilli) are inhaled→Adhesion to macrophages→ LAM present on Mycobacterial Surface → binds to complement Receptor & Mannose receptor (Present on macrophages) because of which Internalization of bacilli happens
- Phagocytosis: C_3b mediated enhancement (opsonization of bacilli)
- Survival Inside the Macrophages: LAM and Cord factor prevent phagolysosomal fusion
- Macrophages Rupture and Release Bacillary content which infect other phagocytes & cycle continues

Host immune response 00:10:47

- Macrophages presentMTB Ag to T helper cells (TH cell)



- Macrophage Activation Response
  - Majority Show resistance to infection (able to contain bacilli)
  - INF activates resting alveolar macrophages because of them Killing & digesting of Tubercle bacilli starts
  - Activated macrophages aggregate around centre of lesions & forms granuloma
  - Activation can happen in two forms of Tubercle
    - → Hard Tubercle
      - Initial stage: Hard
      - Central zone: activated macrophages, Epitheloid& giant cells
      - Peripheral zone of lymphocytes & fibroblasts
    - → Soft Tubercle: Central part undergoes Caseous necrosis (central part containing Cheese like Necrotic material)
- Tissue damaging response 00:17:19
  - Happens in minority
  - MAR is weak andbacilli is more virulent because of this there is an Immense delayed Hypersensitivity leading to Lung destruction
  - Caseous necrosis happens and it Liquefies which contains Large no of bacilli and they sheds off by
    - → Directly drain into the airways
    - → Lymphatic spread
    - → Hematogenous spread to various organs

Primary Pulmonary TB 00:27:30

- Exogenous infection with tubercle bacilli
- Age group: Children
- Production of Sub pleural lesion which affects the upper part of the lower lobe or lower part of upper lobe K/a Ghon's focus(Fibrotic nodular lesion)
- Ghon's focus + Hilar LAP: Ghon's complex orPrimary Complex
- Calcified Hilar LAP: Ranke's complex
- Mostly they are Asymptomatic

Post Primary / Secondary Pulmonary TB

- Due to exogenous re-infection
- Reactivation of primary lesion
- Presence of calcified nodules: Assman's focus(Mostly infraclavicular)
- Hematogenous spreadin apex of lung K/a Simon's focus
  - Involves apical or posterior segments of upper lobe
- Lesions undergo necrosis and there is Tissue destruction leading to Cavity formation

Extra Pulmonary Involvement 00:34:35

- MC extra pulmonary involvement: Lymph nodes > Pleural > Genito Urinary Tract

- Skin TB
  - MC primary lesion: Lupus vulgaris 'Apple Jelly Nodules'
  - 2nd MC lesion: Scrofuloderma (underlying Cervical LN burst up)

- MC site for GI TB: ileo caecal junction
- Skeletal TB: Pott's spin(there can be a Para vertebral cold abscess)

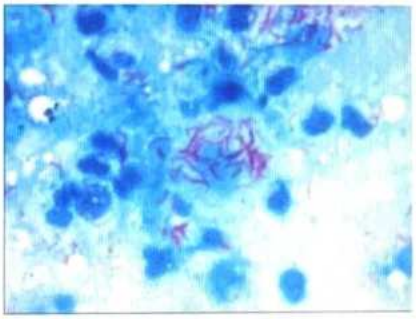
- CNS TB: Can Manifest as
  - T B meningitis
  - Tuberculoma

LAB Diagnosis

- Sputum Examination: 2 sputum samples taken
  - On spot sample
  - Early morning sample

- Then do Concentration of sputum:Petroff's method
  - Thick sputum + (NaOH + Hcl) / NALC: Liquified sputum
  - Concentration is done to increase the yield of bacteria

- From concentrated sputum make ZN smear and culture on LJ media
  - LJ media: Green colour media due to malachite Green dye



  - Rough, Tough, Buff Colonies
    - → Rough: Irregular colonies
    - → Tough: Difficult to remove
    - → Buff: Yellowish Brown

LJ Media

  - Takes 6-8 wks

- For Bacilli to be visible on:
  - ZN smear: > 10,000 bacilli/ml of sputum
  - Culture media: 10-100 bacilli/ml of sputum

- ZN STAINING: readymade sputum smear and add → Carbol fuschin → Intermittent heating → 20% H_2SO_4 (Decolouriser)→Methylene Blue→ Against Blue background can observe Pinkish to red colored AF Bacilli

- Modification of acid last stain: Kinyoun method of staining
  - Cold method of staining
  - More amount of Carbol fuschin is used here & phenol concentration is high and Duration is increased

- Auramin & Rhodamine Staining 00:49:49
  - Florescent staining
  - Done in cases of more load
  - Drawback: false positive Rates are high

Automated liquid culturemedia 00:51:09

- BAC TEC or BACT alert
- Use liquid broth: Middle brook 7H9
- Principle of BAC TEC: O_2 sensitive fluorescent compound in broth

- In the Initial Phase: O_2 is more and Fluorescence is quenched
- In MTB: O_2 is less and Fluorescence is more

Molecular Methods 00:54:45

- PCR based: Target on IS6110 gene
  - Turnaround time: 2 hrs
  - Can't differentiate between live/ Active/ Latent TB
- CBNAAT /Gene xpert Rif
  - Cartridge based Nucleic acid amplification test
  - Rifampicin drug resistance is also detected (based on rpoB gene)

Previous Year's Questions

Q. Gene expert detects resistance due to mutation in following gene? (AIIMS - June - 2020)

A. rpoB
B. pnc A
C. Kat G
D. inh A

- LPA
  - Line Probe Assays
  - PCR based + MDR detection
- MGIT
  - Mycobacterium Growth Indicated Tube
  - Can be used for detection of TB and drug resistance
- Serological Test: Banned in India (No ELISA & no card test)

Diagnosis of latent TB

- IGRA- Interferon Gamma Release Assay (Quantiferon TB gold assay)
  - Look for sensitized T cells on patients Blood sample
  - ESAT-6, CFP-10, TB 7.7 are the artificial mycobacterial antigens by which we challenge the sensitized T cells
  - Sensitized T cells release Interferon
  - Interferon activity ismeasured by ELISA and Prior exposure will be known

- Tuberculin Test/ Mantoux Test 01:08:06
  - 0.1 ml PPD (Purified Protein Derivative)Into flexor aspect of forearm
  - After 3 days()48 to 72 hrs), measure Induration (Hardness Diameter)
    - → < 5mm: Negative
    - → 5 – 10 mm: Equivocal
    - → > 10 mm : positive
  - Erythema should not to be measured
  - False Positive in
    - → Recent BCG vaccination
    - → Atypical Mycobacteria infection
  - False negative (in those with impaired CMI)
    - → HIV +ve (Advanced Stage)
    - → Miliary TB
    - → Malignancy
    - → Immunosupression

MDR TB 01:11:54

- Resistance against H & R
- Resistance against H R Z E S
- But, resistance against Z E S not MDR TB
- For MDR TB Resistance against H & R should be present
- H= Isoniazid , R=Rifampicin , Z= Pyrazinamide, E= Ethambutol and S= Streptomycin

Previous Year's Questions

Q. Which of the following is 2nd line antitubercular drug? (AIIMS - June - 2020)

A. Streptomycin
B. Ethionamide
C. Vancomycin
D. Acyclovir

Previous Year's Questions

Q. Which of the following Antitubercular drug is not bactericidal? (AIIMS - June - 2020)

A. Isoniazid
B. Rifampicin
C. Kanamycin
D. Thiacetazone
E. Ethambutol

XDR TB

- Resistance against 1st line drugs (HRZES) + One of the quinolones in 2nd line drugs+One injectable drug resistance

TDR

- Total Drug Resistance
- No Rx

ATYPICAL MYCOBACTERIA

01:16:09

Runyon's Classification

1. Photochromogens
2. Scotochromogens(Mnemonic)
3. Nonchromogens: MAC, M. xenopi, M. ulcerans
4. Rapid Growers: M. cheloni, M. fortuitum

1. Photochromogens

- Grow in light
- **M** - Mycobacterium **M**arinum
- **A** - Mycobacterium **A**siaticum
- **S**– Mycobacterium**S**imiae
- **K** - Mycobacterium **K**ansasii

How to remember

- MASK

2. Scotochromogens

- Grow in darkness
- **S**–Mycobacterium**S**zulgai
- **S**–Mycobacterium**S**crofulaceaum
- **G**–Mycobacterium**G**ordonae

How to remember

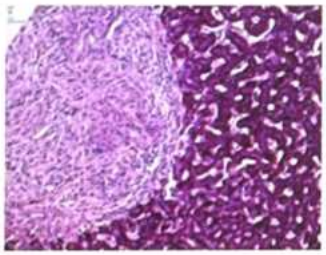
- SSG

3. Non-Chromogens

- Neither light nor darkness required
- MAC (Mycobacterium Avium Complex)/ Mycobacterium Intracellulare aka Battey'sBacillus
- M. xenopi
- M. ulcerans

Previous Year's Questions

Q. Identify organism? (All[illegible]



A. Mycobacterium avium
B. Nocardiasis
C. Actinomycetes
D. Pneumococci

4. Rapid Growers

- M. cheloni
- M. fortuitum
- M. smegmatis
- M. abscessus

Important Information

- M. fortuitum, M. cheloni, M. abscessus are associated with Post injection abscess

Diseases

- Swimming Pool Granuloma/ Fish Tank Granuloma: d/t M. Marinum
- Buruli Ulcer: d/t M. Ulcerans
- Atypical Mycobacteria which involves lungs and mimics MTB in presentation: M. Kansasii
- Another ATM involving lungs: MAC / MAI

Important Information

- MIP (Mycobacterium Indicus Pranii) is being used for preparation of MW Vaccine
- MW vaccine is a Vaccine of leprosy
- This vaccine is an Immunomodulator

Important Information

- In HIV positive individual if CD 4 < 50, infections that can happen are
  - MAC
  - CMV Retinitis
    → Rx: Ganciclovir

MYCOBACTERIUM LEPRAE

01:26:53

- Cause Leprosy akaHansen's Disease
- Doesn't Follow Koch Postulates: M. Leprae, T. pallidum
- Not cultured in Pure culture media
- Best cultured:Foot Pad of Armadillo(9 Banded)
- Also Grown in Foot Pad of Mice

Nine Banded Armadillo

Ridley – Jopling Classification 01:36:12

Refer Table 4.1

- In the above table CMI decreases from left to right

TT (Paucibacillary)	LL (Multibacillary)
• Bacteriological Index: 0 to 1+ • Hypo pigmented annular macules present • Early nerve involvement	• Bacteriological Index: 4 to 6+ • Multiple nodules are present • Nerve lesions appear late

Previous Year's Questions

Q. Which of the following is the most likely cause in case granuloma with positive AFB?
(FMGE - June - 2019)

A. Cat Scratch disease
B. Trench fever
C. Leprosy
D. Syphilis

Lepromin Test 01:38:04

- Type IV HSR
- Lepromin Test is a prognostic test, not a diagnostic test
- Early Lepromin: Fernandez reaction
- Late Lepromin: Mitsuda reaction

Early Lepromin reaction	Late Lepromin reaction
• Read after 3 days • Measures Induration Diameter	• Reads after 3 wks • Measured Nodular Diameter
• Induration > 10mm: +ve	• Nodule > 5 mm:+ve

Lepra Reactions

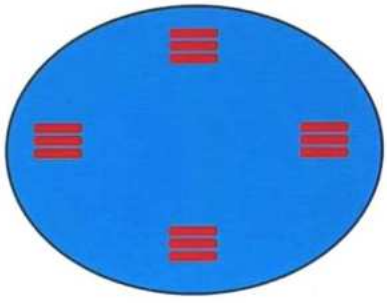
- Can occur spontaneously or after the treatment

Type 1 Lepra Reaction	Type 2 Lepra Reaction
• Type IV hypersensitivity • Also termed as Down grading & up grading reaction • Mc Feature: Edema • Mc nerve involved: Ulnar • Happens in BB cases • TH1 response release IFN & IL-2	• Type III hypersensitivity reactions • Also termed as ENL (Erythema Nodosum Leprosum) • Mc Feature: crops of painful nodules over the skin [d/t release of TNF] • Happens in BL, LL cases • TH2 response release IL-6, 8

- Rx: Lepra reaction is a case of emergency
  - DOC for both Type 1 and 2: Glucocorticoids
- Thalidomide can be used in Type 2 Lepra reaction

Lab Diagnosis

- Slit Skin Smear Examination
- Taken from
  - B/L ear lobes
  - Forehead
  - Chin, cheeks
  - Buttocks
- Nasal mucosal swabs + tissue smear taken and stained
- ZN Staining
  - Add Carbol fuschin, do intermittent heating
  - Add 5% H_2SO_4
  - Add Methylene Blue
  - Blue background
  - Cigar bundle shape/ Globi like arrangement of Leprae seen



Cigar bundle leprae

- For Prognosis of leprosy: Morphological Index (Better)
  - Measures % of Solid stained bacilli (Lice bacilli)

Treatment 01:43:02

- Paucibacillary: D + C + R (As per new WHO Update)
  - Dapsone + Clofazimine + Rifampicin
  - For 6 months
  - In India: (D + R) x 6 months

- Multi Bacillary:D + C + R
  - Dapsone + Clofazimine + Rifampicin
  - For 12 months
  - In India: (D + C + R) x 1 yr

Grading of Smear 01:51:10

- 1-10 bacilli/ 100 OIF: 1 +
- 1-10 bacilli/ 10 OIF: 2 +
- 1-10 bacilli/ 1 OIF: 3 +
- 10-100 bacilli/ OIF: 4 +
- 10-1000 bacilli/ OIF: 5 +
- > 1000 bacilli/ OIF: 6 +
- Based on this grading, Bacteriological index is measured

NOCARDIA & ACTINOMYCETES 01:54:40

Refer Table 4.2

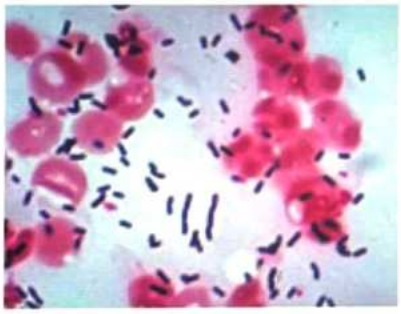
Previous Year's Questions

Q. Swollen foot with sinuses in a former?
(INICET - Nov - 2020)

A. Mycetoma
B. Sporotrichosis
C. Melanoma
D. SCC

LISTERIA MONOCYTOGENES 02:04:07

- Gram Positive Bacillus (GPCB)
- Tumbling motility [Peritrichous Flagella]
- Shows differential motility based on temp
  - At 37°C: Non motile
  - At 20-25°C: Motile



L. monocytogenes

- Cultureon
  - Blood Agar
  - Chocolate Agar
  - PALCAM Agar
- Cold Enrichment Method: at 4° C this organism love to grow on Blood Agar, Chocolate Agar, and PALCAM Agar

Important Information

- In pregnancy avoid refrigerated food because it May contain Listeria which can lead to PROM and abortion

Reactions positive for Listeria

- Anton Test: Putting the Listeria culture into eyes of Rabbit and it develops Keratoconjunctivitis
- CAMP Test
- Catalase Test

Diseases

- Early Neonatal phase: Granulomatosis Infantiseptica
- Late Neonatal phase: Meningitis
- In Pregnancy: Chorioamnionitis which leads to PROM which can lead to Preterm Births and Abortions
- In immunosuppressed patients: Sepsis and Meningitis

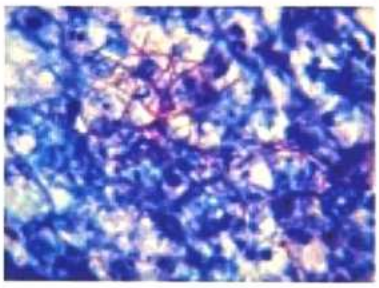
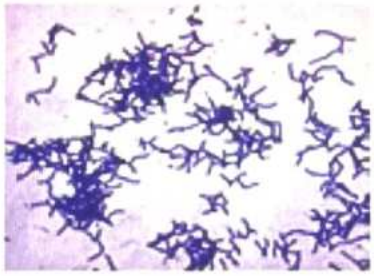
Treatment

- DOC for Listeria:Ampicillin
- It promotes Actin polymerization for intracellular movement

Table 4.1

TT	BT	BB	BL	LL
T. Tuberculoid		Boderline		Lepromatous Leprosy
Paucibacillary (Good prognosis)			Multibacillary (Poor Prognosis)	
• Early involvement of nerve • Granuloma formation	• Granuloma formation • Nerve abscess is common		• No Granuloma formation	• Late nerve involvement • Leonine facies • Lucio phenomena aka Periarteritis • No Granuloma formation

Table 4.2

Nocardia	Actinomycetes
	
Nocardia	**Actinomycetes**

• Strict (Obligate) aerobe
• 1% Acid fast
• Soil (Exogenous)
• MC site: Pulmonary (Pulmonary Nocardiosis)
• MC extra pulmonary site: Brain
	(Antler like branching / Filamentous branching)
	• Strict (Obligate) anaerobe
	• Mainly non acid fast
	• Mouth (Endogenous flora)
	• MC site: Angle of Jaw
	• Disease: Oral-cervical actinomycosis
	• Triad of Oral-cervical actinomycosis:
	  ○ Sinus
	  ○ Swelling
	  ○ Sulfur granules: Crush them & see under microscope, Sun ray appearance can be observed

Sulphur Granule

- A. Israeli
  - ○ Acid fast
  - ○ Leeds to PID
  - ○ Occurs in IUCD users

- Nocardia & Actinomyces in combination causes: Actinomycetoma
  - Multiple sinuses can be observed
  - It is called as Mycetoma foot

Mycetoma foot

- Culture: Paraffin bait technique
- DOC: Cotrimoxazole

- Spider molar tooth appearance on solid media
- Fluffy ball on bottom in liquid media
- DOC: Penicillin

CLINICAL QUESTIONS

Q.1 A patient presented to the OPD complaining of cough from 3 weeks, chills, fever and extreme weight loss. The blood sample was sent to the laboratory for testing. In the laboratory, interferon gamma release assay was used for the diagnosis. Which of the following statement regarding the Interferon-gamma release assays used for the diagnosis is correct?

A. 1st generation Quantiferon-TB does not use ESAT-6
B. 2nd generation Quantiferon-TB (gold) used ESAT-6 and CFP-10
C. These tests can distinguish between M. tuberculosis and M. bovis
D. None of the non-tubercular mycobacteria gives a positive reaction with this test

Solution:

- **2nd generation Quantiferon-TB (gold) used ESAT-6 and CFP-10.**
- Interferon-gamma release assays -IGRA (**Quantiferon TB):**
  - **Cell-mediated immune (CMI)** responses to peptide antigens that simulate mycobacterial proteins.
  - Available assays are:

  → **Quantiferon TB**(1st generation):measured cell mediated immunity to the same tuberculin purified protein derivative.

  → **Quantiferon TB Gold** (QFT, 2nd generation): Whole blood enzyme linked immunosorbant assay.
- In vitro assay for **diagnosis of latent tuberculosis, measures INFγ released by T cells** in response to **highly specific TB antigens ESAT-6 and CFP-10.**
- These proteins, **ESAT-6, CFP-10,** and **TB7.7(p4)** are absent from **all BCG strains** and from most nontuberculous mycobacteria with the **exception** of M. kansasii, M. szulgai, and M. marinum.

Note – Serological tests are banned in India for MTB diagnosis.

Q.2 A 3 week old child presented to the pediatrician with meningitis. A presumptive diagnosis of late onset perinatal meningitis was made. The CSF culture was positive for gram positive bacilli. Which of the following characteristic of this bacteria would be helpful in differentiating it from other bacterial agents:-

A. Ability to grow on blood agar
B. Ability to produce catalase
C. Fermentative attack on sugars
D. Motility at 25 degree Celsius.

Solution:

- L. monocytogenes is responsible for late onset perinatal meningitis.
- Neonatal meningitis is caused by:
  - Escherichia coli : Gram negative bacilli

- Group B Streptococcus : Gram positive cocci
- **Listeria monocytogenes: Gram positive bacilli**

LISTERIA MONOCYTOGENES:

- Tumbling motility (Peritrichous Flagella)

Culture:

- Blood Agar
- Chocolate Agar
- PALCAM Agar
- Cold ENRICHMENT METHOD: at 4°C.
- Anton test Positive: Putting the Listeria culture into eyes of Rabbit → Keratoconjunctivitis.
- Catalase test Positive.

Diseases:

1. Granulomatosis Infantiseptica [In neonates].
2. Meningitis : children (mainly), also in Adults.
3. Chorioamnionitis [In pregnancy] → PROM, Preterm Births Abortions.
4. Sepsis : in immunocompromised.

- Drug of Choice → **Ampicillin**
- **Differential motility**: Tumbling motility at 20-25 degree Celsius but not at 37 degree Celsius.

5 GRAM NEGATIVE BACILLI PART-1

ENTEROBACTERIACEAE FAMILY

00:00:22

Common Features

- Gram Negative Bacteria
- Catalase Positive, except: Shigella dysenteriae- type I
- Oxidase Negative
- Ferment sugar & Produce Acid with gas
- Nitrate to Nitrite Reduction Test Positive
- Motile, except: Klebsiella & Shigella, Salmonella gallinarum, Salmonella pullorum

LACTOSE FERMENTERS | NON LACTOSE FERMENTERS

1. E.COLI

00:03:48

Refer Table 5.1

- Color change in citrate Test: Green to Blue

Diseases

- UTI
- Diarrhea
- Neonatal Meningitis
- Emphysematous Pyelonephritis
- Peritonitis
- Fournier's Gangrene
- Scombroid Fish Poisoning
  - M/c: Proteus > Morganella > E. coli or Morganella > Proteus > E. coli (overall)
- Histamine Production increased
- Pneumonia
- Liver abscess

UTI

- Caused by 'P' Fimbriae possessing E. coli
  - It is Mannose Resistant
  - They attach to uroepithelial cells and caused UTI
- Criteria for diagnosis of UTI
  - Significant Bacteriuria: $> 10^5$ E. coli/ ml of urine
  - $>10^4$ CFU/ ml + Symptoms / signs of UTI
  - Even 1 bacteria/ml in supra Pubic Aspirate
- Mid stream urine sample taken and plated on Mac Conkey/ CLED (Preferred)
- Next Day: > 100 colonies x 10^3 (dilution factor) and it come out to be $>10^5$ its diagnostic of UTI

Diarrhoea Causing E. coli

00:08:48

a. EPEC: Entero Pathogenic E. coli
b. EIEC: Entero Invasive E. coli
c. ETEC: Entero Toxigenic E. coli
d. EHEC: Entero Hemorrhagic E. coli
e. EAEC: Entero Aggregative E. coli

a. EPEC

- Causes Pediatric age group Diarrhea
- Attach to Intestinal villi & Damage them Causing attaching & effacing lesions (A/E Lesions)

Previous Year's Questions

Q. Identify the organism which shows effacement & attachment of intestinal cells as shown below?
(AIIMS - May - 2018

A. EPEC
B. EIEC
C. Enterohemorrhagic E. coli
D. Diffusely enteroadherent E. coli

b. EIEC
- Resembles Shigellosis
- Invasiveness d/t VMA (Virulence Marker Ag)
- Sereny Test Positive

c. ETEC
- M/c Causes of Travelers Diarrhea
- Toxins
  - Heat Stable Toxin: acts by ↑ CGMP
  - Heat Labile Toxin: acts by ↑ CAMP
- CFA (Colony Factor Ag)

d. EHEC
- O157 : H7
  - O: somatic antigen
  - H: Flagellar antigen
- Causes Hemolytic uremic Syndrome & hemorrhagic colitis in children
- Toxins
  - Verotoxin 1 & 2 so termed as Vero Toxigenic E. coli / Shiga Like Toxin
- Cuture Media
  - SMAC [Sorbitol Mac - Conkey Agar] Does not Ferment Sorbitol
  - Rainbow agar

Previous Year's Questions

Q. Hemolyticuremia syndrome associate with?
(FMGE - June - 2019)

A. E. Coli 0157
B. Malaria
C. Parvovirus B19
D. Bartonella henselae

e. EAEC
- Causes Persistent type of Diarrhea
- Toxin: EAST 1
- Stacked Brick Appearance on Hep 2 cell lines
- Also Causes Traveler's Diarrhea

Neonatal Meningitis
- Common causes of Neonatal Meningitis
  - In India: E. coli [possessing K_1 antigen]
  - Worldwide: GBS [Streptococcus agalactiae]

Important Information

- E. Coli having KI antigen causes neonatal meningitis

Lab Diagnosis (E. coli) 00:26:21
- In case of UTI
  - In Wet mount examination: Pus cell along with Bacteria can be seen
  - Leukocyte esterase test: L. esterase enzyme in urine pus cells seen
  - Nitrate Reduction Test(Griess Nitrite Test)
  - Gram staining
  - Confirmatory: Culture on CLED/ Mac Conkey agar
    → It Differentiate Lactose Fermenting colonies
    → Sample: Urine/ Stool

00:29:27

2. KLEBSIELLA
- Gram Negative Bacillus
- Capsulated
- Non - motile
- Lactose Fermenter
- Urease Positive [Yellow → Pink]

UREASE POSTIVE ORGANISMS	
Staph	Staph. aureus
Klub	Klebsiella
Main	Morganella
Urease	Ureaplasma Urealyticum
Programme	Proteus
Hai	H. Pylori [max. Urease producer]

Urease Test -ve | Urease Test +ve

Klebsiella pneumoniae [Fried Lander's Bacilli]
- Causes typical pneumonia (Lobar pneumonia)
- A/w RED Currant Jelly Sputum
- A/w Bulging Fissure Sign on CXR

Klebsiella Ozaenae

- A/w Ozaena [atrophic rhinitis]
  - Roomy nose
  - Immense foul smell
  - Anosmia
  - Rx by Na HCO_3 douches
  - Modified Young's Operation

Klebsiella rhinoscleromatis

- Causes Rhinoscleroma [Woody Nose]
- On Biopsy
  - Mikulicz Cells
  - Russel Bodies

Klebsiella granulomatis

- Donovanosis (Granuloma inguinale)

Previous Year's Questions

Q. Which of the following is the cause of donovanosis?
(NEET - Jan - 2020)

A. H. Ducreyi
B. K. Granulomatis
C. Leishmania Donovani
D. Treponema Pallidum

Lab Diagnosis 00:35:26

• Disease	UTI	Pneumonia	Meningitis
• Sample	Urine	Sputum	CSF
• Culture	CLED/ Mac Conkey Agar/ BA	CLED/ Mac Conkey Agar/ BA	CLED/ Mac Conkey Agar/ BA

- LF Colonies
  - On CLED: Yellow colonies
  - On Mac Conkey agar: Pinkish colonies
- Very mucoid colonies
- PCR for K. Species
- NDM-1 (New Delhi Metallo -Lactamase)
  - Resistance to all types of Antibiotics
- E. coli/ Klebsiella tend to have ESBLs enzymes

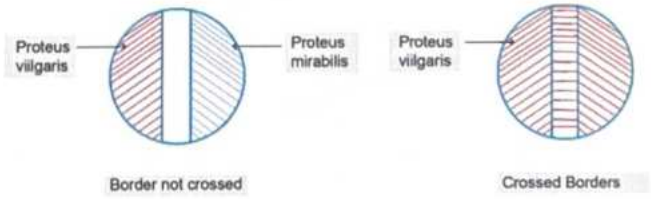
3. PROTEUS 00:38:53

- GNB, NLF, Non capsulated
- Swarms
  - Inhibit swarming by
    - → ↑ % Agar from 2% to 6%
    - → Instill some chemicals [Boric acid & chloral Hydrate]

Swarming

- Can be cultured on
  - BA
  - Mac - Conkey Agar: Produce Non Lactose Fermenting [NLF] colonies
  - Phenyl Pyruvic Acid Agar
    - → Fishy smell/Seminal smell produced on both media

Diene's Phenomenon



- If species are different: boundary between the 2 species are maintained
- If species are same: they cross boundary

Weil – Felix reaction

- It is a type of HAT (Heterophile agglutination test)
- Agglutination reaction Between Non- motile strains of proteus with Rickettsial family members
- Proteus Antigen Interactions
  - OX 19, 2 & K → cross reacts with Rickettsial family members
  - Gives Weil - Felix Reactions [Heterophile agglutination test]
  - OX K is strongly positive For scrub typhus
- Proteus a/w chronic UTI: Leads to Struvite/Triple Phosphate stones/Stag horn calculus

4. SALMONELLA 00:46:10

- GNB
- Motile mostly
- Non motile are
  - S. gallinarum

- S. pullorum
- Antigens: O, H, Vi
  - O: Somatic
  - H: Flagellar

Kaufmann & White Scheme Classification

Refer Table 5.2

'O' Antigen	'H' Antigen
• Part of 'LPS' cell wall	• Made of Flagellin
• Heat stable	• Heat labile
• Used for widal test	• Used for widal test
• Less immunogenic	• More immunogenic
• 'O' Ab appears early & goes early	• 'H' Ab appears late & goes late
• Granular chalky clumps are produced	• Loose fluffy clumps are produced

Variation in 'O' Ag
- S – R variation (Smooth-Rough)
- In Salmonella
  - Lysogenic conversion occurs
  - Bacteriophage mediated
  - Loss or gain of an 'O' Ag

Variation in 'H' AG

- OH-O' variation: Loss of Flagella
  - Because of culture on Phenol Agar
  - D/t mutation
- Phase variation
  - Phase 1: Serotype specific
  - Phase 2: Non specific

a. Salmonella Typhi
- Causes Typhoid Fever
- Enteric FEVER caused by
  - S. Typhi
  - S. Para typhi A, B & C
- Transmitted by Feco oral contamination
- Infective dose $10^3 - 10^6$ bacilli
- Entry through epithelial cells 'M' cells (Present in intestinal mucosa)
- Bacteria Mediated Endocytosis
  - Process of uptake of bacteria
  - Type III secretion system involved
  - Following entry bacilli remain in the vacuoles in cytoplasm

Enteric Fever
- Clinical features
  - Stepladder pattern fever
  - Rose Spots Seen
  - Early abdominal pain, Nausea, Vomiting
  - Hepatospleenomegaly, Relative Bradycardia aka Faget's sign

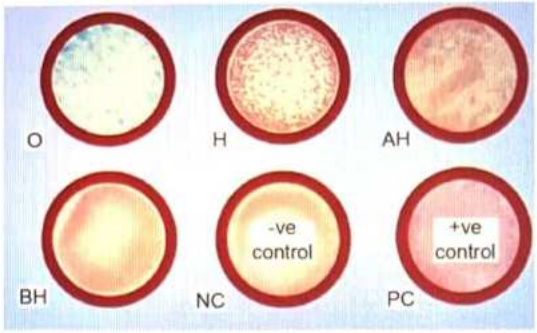
- Complications
  - In 3rd and 4th week of illness
    → GI bleed
    → Intestinal perforation
  - Pea soup diarrhea
  - Longitudinal ulcers
- Lab diagnosis 01:04:52
  - **B** - **B**lood culture/ Bone marrow culture in 1st week
  - **A** - **A**gglutination Test in 2nd Week
  - **S** - **S**tool culture in 3rd Week
  - **U** - **U**rine Culture in 4th Week

How to remember

- BASU

- Blood culture
  - Overall Best
  - Blood: culture Fluid ratio 1 : 10 [Eg. 10 ml : 100 ml]
    → Bile broth or Glucose broth can be used
  - Add SPS [Sodium Polyanethol Sulfonate] to remove antibiotic effect
  - Initial report: 3 days
  - Final report: 7 days

- WIDAL test



  - Highly sensitive but Poorly specific
  - Positive if Anti Body titer against
    - → O: > 1:100
    - → H: > 1:200
  - Paired testing should be done, after 2 – 3 weeks of widal test and there should be 4 Fold ↑ in titer should be present then Positive
  - IgM Typhi Dot Testing: better than WIDAL test

Previous Year's Questions

Q. When should you perform widal test in case of typhoid? (FMGE - June - 2019)

A. 1st week
B. 2nd week
C. 3rd week
D. 4th week

- Stool & Urine Culture
  - Enrichment media
    - → Selenite F Broth
    - → GN Broth
    - → Tetra thionate broth
  - Selective media
    - → Wilson & Blair Media
      - Best media
      - Has jet black colonies

S. typhi COLONIES IN WB-MEDIUM

    - → DCA [Deoxycholate Agar]
    - → XLD [Xylose Lysine Deoxycholate]
    - → SS [Salmonella Shigella Agar
  - Urine culture on
    - → Mac- Conkey agar
    - → CLED Agar
- Transport Media for Salmonella/ Shigella: Sach's Buffer Glycerol Saline
- Chronic Carrier: persons who harbor it for
  - > 1 yr
  - Mostly in Gall Bladder d/t abundant bile in Gall Bladder
- Vi Antigen
  - Vi mostly covering up 'O'
  - Vi Phage typing can be used For epidemiological studies

b. Non salmonella typhi

- S. typhi murium
  - Cause gastro enteritis
  - A/w contaminated Poultry Products
  - Can enter the intact egg shell
- S. enteridis: cause Gastro enteritis
- S. choleraesuis: cause Septicemia
- DOC: 3rd Generation

Important Information

- Reactive arthritis can be a/w Salmonella. Shigella
- Salmonella a/w Osteomyelitis (in Sickle Cell Anemic patients)

5. SHIGELLA

01:24:19

- GNB
- Catalase +ve, except
  - S. dysentriae type 1 (Catalase negative)
- Non motile
- Mannitol fermenters
  - Except: S. Dysentriae Type 1 (Non mannitol fermenter)
- Feco-oral route
- 10-100 bacilli: Infective dose
- 'M' cell play a role here
- VMA: Virulence Marker Ag
  - Responsible for Invasiveness
- Secretes
  - Shiga toxin
  - Made by: S. dysentriae type 1 (Secretes Exo & Endotoxin)

- MCC of Bacillary dysentery:
  - In India: S. flexneri
  - In world: S. Sonnei (Late Lactose Fermenter)
- Most Shigella species: Serotyping is done
- For S. Sonnei: Colicin typing is done (26 types)

Clinical Manifestations of shigellosis

- IP: 1-4 days
- Initial Phase: Watery diarrhea
- Dysentery phase: Mucopurulent stools (Bloody)
- Phase of Complications (<5 yrs of age)
  - Tonic mega colon
  - Perforation
  - Metabolic Complications:
    - → Hypoglycemia
    - → Hyponatremia
    - → Dehydration
- "Ekiri" Syndrome
  - Toxic Encephalopathy
  - Metabolic complications present
  - Neurological features
  - (Delirium, confusion)

- Post Infectious Phase: Those which posses HLA-B27 can develop Autoimmune reaction months after infection which is characterized by
  - Reactive arthritis
  - Urethritis
  - Conjunctivitis

Previous Year's Questions

Q. A child presents with Red swollen & tender knee joints 2 weeks following an episode of fever with bloody diarrhoea. What can be the most probable etiological agent? (AIIMS - June - 2020)

A. E. Coli
B. Shigella
C. Giardia
D. Rotavirus

Previous Year's Questions

Q. 25-year-old man presented with diarrhoea& blood, on investigation of stool sample, the organism was gram negative, non-lactose fermenter, non-motile, oxidase negative, H_2S negative diarrhea? (JIPMGE - Dec - 2020)

A. E. Coli
B. Shigella
C. Salmonella
D. Y. Pestis

- Culture Media
  - Transport Media: Sach's Buffered Glycerol Saline
  - Enriched media: SFB
  - Selective media:
    - → DCA
    - → XLD
    - → HE
    - → SS
- DOC: Ciprofloxacin

LACTOSE NON FERMENTERS

- Doesn't Ferment Sugars
- Includes
  - Pseudomonas
  - Burkholderia mallei
  - Burkholderia Pseudo mallei
  - Acinetobacter baumanii
  - STM

1. PSEUDOMONAS

01:40:04

- GNB
- Catalase +ve
- Oxidase +ve
- Oxidase reagent
  - Tetra methyl Para phenylene diamine di hydro chloride
  - Comes as white Disc
  - Change of color From white to blue within 10 sec: Positive
- Virulence factors
  - Capsular Polysaccharide
  - Pigments
    - → Pyocyanin: Blue
    - → Pyorubrin: Red
    - → Pyomelanin: Brown Black
    - → Pyoverdin: Green
      - Pseudomas Aeruginosa produce both Pyocyanin and Pyoverdin (mainly Pyocyanin)
  - Aliginic Acid
    - → Found in Mucoid strains of pseudomonas
    - → Helps in Biofilm formation

01:44:24

- Lab Diagnosis
  - Cetrimide Agar: Best selective media
  - Mac Conkey agar: Non-Lactose Fermenter on Mac Conkey agar
  - Nutrient Agar: Greenish pigmentation on NA
  - Can have Grape like odor or fruity smell on all 3 medias
- Diseases

a. Shanghai Fever
b. Malignant Otitis Externa (Fulminant)
c. Ventilator Associated Pneumonia (MCC)
d. Respiratory infections in cystic fibrosis (Burkholderia

cepacia > pseudomonas)

e. Bacteremia
f. Native Valve Endocarditis (IV drug abusers)
g. Ear infection: Swimmers ear (among children)
h. Eye infection: Corneal ulcer, in contact lens
i. Skin infections
   - Burn Wound Infections
   - Ecthyma Gangrenosum
   - Green Nail Syndrome
     → Inflammation of tissue adjacent to nail
     → Green pus/ Blue-Green pus
j. Meningitis
k. Bone & joint infection

- Treatment: Anti Pseudomonal Drugs
  - Vancomycin is not an Anti Pseudomonal
  - Polymyxin B & Colistin: No Drug resistance reported

2. BURKHOLDERIA MALLEI

- Causes Glanders Diseases: Animal disease [Horses]
- Non - motile

3. BURKHOLDERIA PSEUDO MALLEI

- Causes Meliodosis
- Aka Whitmore's Bacillus/ Vietnam Bomb
- Both Burkholderia are Bipolar stained/ Safety pin appearance

4. ACINETOBACTER BAUMANII

- MDR [multi Drug Resistant] isolate in the ICU
- Important species: A. Baumanii
  - OMPP
  - LPS
  - Siderophore
  - A/w formation of Biofilm formation
  - A/w UT, VAP
  - Lead to CLABSI (Central Line Associated Blood Stream Infection)
  - A/w Burn Wound Infection
- Gram negative Coco Bacillus
- Oxidase Negative
- As per WHO now-a-days Acinetobacter infections are increasing so they are in the critical list of WHO pathogens (A. Baumanii)

Previous Year's Questions

Q. Organism most likely associated with VAP? (AIIMS - May - 2018)

A. Acinetobacter
B. Klebsiella
C. Clostridium
D. Mycobacterium TB

5. STENOTROPHOMONAS MALTOPHILA

- Saprophytic in plant roots
- Immuno deficient individuals are more prone to infection
- Leads to pneumonia
- Oxidase negative
- Motile
- Oxidatively ferments: Glucose, Maltose & Lactose

02:03:11

VIBRIO

- GNB
- Curved: comma shaped
- It is COINS Positive
  - **C** - **C**atalase
  - **O** - **O**xidase
  - **I** - **I**ndole
  - **N** - **N**itrate reduction test
  - **S** - **S**ucrose Lysis, **S**tring test positive
- On the basis of O1 Vibrio is classified into
  - Classical Vibrio: further classified into
    → Ogawa
    → Inaba
    → Hikojima
  - Eltor Vibrio: classified into
    → Ogawa
    → Inaba
    → Hikojima

	Classical	Eltor
• Polymyxin B sensitivity	Sensitive	Resistant
• Mukherjee Phage IV assay	Sensitive	Resistant
• VP [Voges Proskauer]	Negative	Positive
• CAMP	Negative	Positive
• Chick Erythrocyte Agglutination	Negative	Positive

- O2 - O139 Vibros: termed as NAG
  - O139: Termed as Bengal strain. It resembles Eltor Vibrio

1. V. Cholerae

- Virulence factors
  - TCP (Toxin Co regulated Pilus):
    → Type - 4 Fimbriae, helps in attachment
    → Infective dose: 10^8-10^{10}
  - CT (Cholera Toxin)
  - Zona occludens toxin: disrupts tight junction
  - Cholera Enterotoxin
  - Siderophore
  - LPS
    → Does not play role in pathogenesis of cholera

→ Used to prepare killed vaccine
- Used to prepare a killed vaccine

- 1st 6 pandemics: by classical strain
- 7th pandemics: by Eltor
- O139: Bengal outbreak

Previous Year's Questions

Q. Diarrhoea in vibrio cholerae is due to disruption of?
(INICET - Nov - 2020)

A. Zonula occludens
B. Hemidesmosomes
C. Macula Densa
D. Intercalated disc

Cholera Toxin

- Made up of 2A (A_1, A_2) + 5B (B-helps in binding to GM1 ganglioside Receptor)
- A helps in ADP Ribosylation of GTP as a result there is an increase in adenylate cyclase activity causes an increase in CAMP leads to Intra cellular water in lumen causing Massive cholera (Rise watery stool)
- Rx: Rehydration with IV fluids
- It mimics arsenic metal poisoning

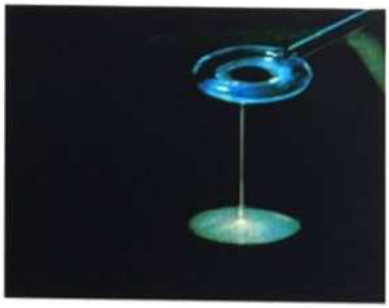
Previous Year's Questions

Q. What is the mechanism of action of cholera toxin?
(FMGE - Aug- 2020)

A. ADP Ribosylation of GTP
B. Decreases adenyl cyclase activity
C. Decreases CAMP
D. All of the above

Lab Diagnosis 02:14:21

- Stool sample
- Hanging drop: Darting motility seen
- String test: Stool + Na Deoxycholate: string forms



- Stool sample on slide: Fish in stream appearance
- Transport media for stool sample
  - VR
  - Carry Blair
- Enrichment media: Alkaline peptone water
- Best selective media: TCBS
  - Greenish colour media which changes to yellow due to sucrolysis
- Other selective medias:
  - Monsur's Taurocholate trypticase tellurite agar
  - Bile salt agar

2. Halophilic Vibrios

a. V. Parahemolyticus: 7% halophilic
b. V. Vulnificus: 8% halophilic
c. V. Alginolyticus: 10% halophilic

a. V. Para hemolyticus

- Capsulated, Bipolar stained
- Causes Sea Food Poisoning (Oyster & crabs)
- A/w Kanagawa Phenomenon on Wagatsuma Agar [enhanced beta Hemolysis on blood agar]
- IP: 8-24hrs

b. V. Vulnificus

- Coral cut wound infection

Important Information

Q. How to differentiate between Vibrio parahemolyticus and Vibrio cholera?

- V. Parahemolyticus is a Non sucrose fermenter so it does not turn TCBS yellow

TCBS

Table 5.1

	I	M	Vi	C
	Indole	Methyl Red	Voges Proskauer	Citrate
E. coli	Positive	Positive	Negative	Negative
Klebsiella	Negative	Negative	Positive	Positive

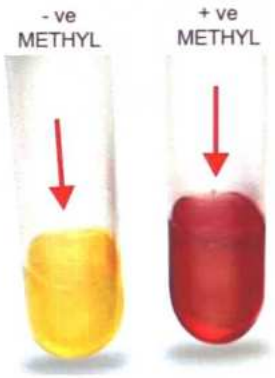


Table 5.2

	Serogroups	'O' Ag	Vi Ag	H	
				Phase 1	Phase 2
S. para typhi A	A	1, 2, 12	Absent	a	1, 5
S. para typhi B	B	1, 4, 5, 12	Absent	b	1, 2
S. typhi murium	B	1, 4, 5, 12	Absent	i	1, 2
S. typhi	D1	9, 12	Present	d	Absent

CLINICAL QUESTIONS

Q.1 A patient with repeated episodes of diarrhoea and fever visited the OPD. The doctor sent the patient for stool examination and culture in the microbiology laboratory. In the laboratory, what is the screening procedure used to differentiate Salmonella and Shigella from other Enterobacteriaceae?

A. Inability to ferment glucose
B. Inability to ferment lactose
C. Produce cytochrome oxidase
D. Reduce nitrates

Solution:

- **Inability to ferment lactose** is used to differentiate Salmonella and Shigella from other Enterobacteriaceae.

Option 1, 3, 4 **Enterobacteriaceae** properties:

1. Ferments glucose
2. Reduces nitrate to nitrite
3. Oxidase test negative
4. Catalase positive (**except** Shigella dysenteriae type-1)
5. Motile, **except** Shigella and Klebsiella.

	Lactose fermenters	**Non Lactose fermenters**
Option 2	Escherichia Klebsiella Enterobacter	Other members of Enterobacteriaceae family including Salmonella and Shigella.

Lactose fermenting colonies | Non-Lactose fermenting colonies

Q.2 A 65-year-old diabetic man presents to the emergency room with a severe productive cough producing thick bloody sputum resembling a "currant-jelly" like appearance. Culture using MacConkey agar reveals pink colonies, with large mucoid colonies on routine laboratory media. Which of the following organisms is most likely responsible for this patient's pneumonia?

A. Enterobacter cloacae
B. Escherichia coli
C. Klebsiella pneumoniae
D. Pseudomonas aeruginosa

Solution:

Klebsiella pneumoniae is most likely responsible for this patient's pneumonia.

- Patients with K. pneumoniae infections usually have predisposing conditions such as alcoholism, advanced age, chronic respiratory disease, and diabetes.
- Upper lobes of the lungs are predominantly affected.
- **The "currant-jelly" sputum** distinguishes *K. pneumoniae* from the other organisms.
- K. pneumoniae is Lactose fermenting (pink colonies on MacConkey agar) and due to its thick capsule the colonies have a **mucoid** appearance (Refer to the image below).

Q.3 A 31-year-old male presented to the ER with complaints of red and swollen joints after a period of about one week of bloody diarrhea caused by shigella that started approximately three weeks before. The species of shigella responsible for causing arthritis is:

A. Shigella dysenteriae-1
B. Shigella sonnei
C. Shigella flexneri
D. Shigella boydii

Solution:

- Reactive polyarthritis develops several weeks after ~1% of cases of nongonococcal urethritis and 2% of enteric infections, particularly those due to Yersinia enterocolitica, Shigella flexneri, Campylobacter jejuni, and Salmonella species.

- Shigella is highly communicable. The infective dose for shigella is less. It can be as low as 10-100 bacilli because they survive gastric acidity better than other enterobacteria.
- **Shigellae produce the following clinical features.**
  1. Intestinal: These are
     - Dysentery: Most common cause is Shigella dyenteriae type-I.
     - Diarrhoea: Usually by Shigella sonnie.
  2. Extraintestinal: These are hemolytic uremic syndrome (caused by Shigella dysenteriae-I), arthritis (Shigella flexneri), seizures (Shigella flexneri), pneumonia and Reiter syndrome (in HLA B27 association).

6 GRAM NEGATIVE BACILLI PART-2

HAEMOPHILUS

00:00:14

1. HAEMOPHILUS DUCREYI

- Causes chancroid (soft sore): Painful, soft genital ulcer

Important Information

- Painless, hard ulcer: Syphilis

- Lab Diagnosis: Tissue biopsy + Stain (gram stain) shows School of fish appearance / Tram track appearance
- Culture media: Chocolate agar with isovitale-X
- DOC: Azithromycin

Previous Year's Questions

Q. Urethritis in males is not caused by?

(NEET–Jan - 2020)

A. H. Ducreyi
B. Trichomonas
C. Chlamydia
D. Gonococcus

Previous Year's Questions

Q. Which are true about H. Ducreyi induced chancroid? Except?

(INICET–Nov - 2020)

A. Painful
B. Bleed on touch
C. Groove sign
D. School of fish

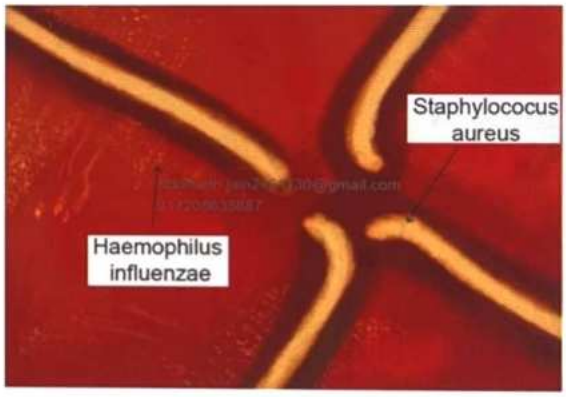
2. HAEMOPHILUS INFLUENZAE

- Termed as Pffeir's Bacillus
- It needs X and V factors for growth
- X is termed as Hemin
- V is termed as NAD
- Both Hemin and NAD are present in Chocolate Agar
- Modifications of Chocolate Agar: Levinthal & Filde's Agar
- H. influenzae is
  - Capsulated
  - Catalase +ve
  - Oxidase +ve
  - Pleomorphic
- Virulence Factors
  - Capsular polysaccharide
  - Ig A_1 protease
  - Endotoxin
  - OMP's
  - Pili

Clinical ManifestationsCapsulatedNon Capsulated

Capsulated	Non Capsulated
• Causes invasive disease o Pyogenic meningitis o Subdural effusion (high mortality rate) o Epiglottis o Pneumonia in infants	• Non-invasive disease o Otitis media o LRTI o Pneumonia in adults o Puerperal sepsis

- On the basis of Capsular Polysaccharides, 6 Serogroups are there: a, b, c, d, e, f
- Type 'b' PRP (Polyribitol Phosphate) led to formation of HIB Vaccine
- Satellitism: Shown by H. influenzae



Lab Diagnosis

- C.S.F should not be refrigerated because H. influenzae is cold labile
- Gm staining : Gm negative coccobacilli showing pleomorphism
- Capsule detection: Quellung reaction

- Ag detection kits: mostly available for Type 'b' capsular Ag
- Culture: BA, CA, Levinthal & Filde's Agar
- Hemophilus Selective Media

- H. Influenzae: Does not ferment sucrose
- H. parainfluenza: Ferments sucrose

3. HAEMOPHILUS AEGYPTI CUS 00:17:37

- Also termed as: Koch Week's Bacillus
- Causes
  - Pink Eyes
  - Brazilian Purpuric Fever

Normal Mouth Commensals

- They can cause culture negative Endocarditis in immunocompromised
- **H** - **H**. parainfluenza, H. aprophilus, H. para aprophilus
- **A** – **A**ggregatibacter species [Actinomycete comitans]
- **C** - **C**ardio bacterium hominis
- **E** - **E**ikenella coordens [a/w human bites]
- **K** - **K**ingella Kingae

BORDETELLA PERTUSIS 00:20:32

- Causes Pertussis / whooping cough / 100 day cough/ Barking dog cough
- Virulence Factors
  - Tracheal Cytotoxin
    - → Part of cell wall Peptidoglycan
    - → Damage to cilia of respiratory epithelium
  - Pertussis toxin
  - Adenylate toxin
  - Dermonecrotic toxin: Respiratory mucosal damage
  - Endotoxin (LPS)
  - Adhesins
    - → Filamentous haemagglutinin
    - → Pertactin (OMP)
    - → Fimbriae
    - → BrKA Protein: gives B. Pertussis resistance to killing & it's adhesion
- Acellular Pertussis Vaccine
  - Mc used Now
  - Equal in efficacy with cellular Pertussis vaccine but devoid of encephalitis like features
  - Made Up of
    - → Pertussis Toxoid
    - → Filamentous Haemagglutinin
    - → Pertactin
    - → Agglutinogens 1, 2 & 3
    - → Fimbrial part

Clinical Manifestations

- IP 1-2 wks / 7 to 14 days
- Stages
  - Catarrhal Phase
  - Paroxysmal phase
  - Convalescent phase
- Catarrhal Phase
  - 1-2 wks
  - Characterized by non specific features like Common cold, cough (mild)
  - Culture positive
- Paroxysmal phase: Whoops present
  - Whooping cough
  - Post tussive vomiting
  - Culture negative
- Convalescent / Recovery Phase
  - Abs may be present in the serum

Lab Diagnosis

- Nasopharyngeal swab (Dacron or calcium alginate swab) Gm staining or Culture
- 6 swabs are taken
- Culture Media
  - Bordet Gengou Media: Its having Bisected pearl appearance or Mercury drop appearance or Thumb print appearance on culture smear
  - Regan - Lowe media

DPT Vaccine

- Pertussis Acellular component present
- It plays a role of Adjuvant
- Increase immunogenicity of whole vaccine

Components of Acellular Pertussis vaccine

- Filamentous hemagglutinin
- Pertactin
- Agglutinogens 1, 2, and 3
- Fimbrial part

Treatment

- DOC: Macrolides

BRUCELLA 00:36:19

- Causes Brucellosis which is a Zoonotic Disease
- MOT
  - By eating & drinking contaminated food & milk products
  - Contact with animals (aerosols)
- Species of Brucella
  - B. abortus: infects cattles
  - B. melitensis: infects sheep, Goat, camel
  - B. suis: infects Pigs
  - B. canis: Infects Dogs

1. **B. Melitensis**

- Causes Acute Brucellosis/ Malta/ Mediterranean/ Undulant Fever
  - Typhoid Like illness
  - High grade Fever, Rash, Hepatospleenomegaly, features of Arthritis
  - Undulating Nature of fever: It has a Typical remittent course (Recovery phase between 2 febrile episodes)
  - Vertebral osteomyelitis involving lumbar region
  - Septic arthritis

Lab Diagnosis

- Specimens: Blood, Bone Marrow Culture
- Blood Culture: Castaneda method of isolation
  - Biphasic media [Solid & liquid culture media]
  - Ratio of blood & culture fluid is 1:10
  - Glucose Broth is taken as culture Fluid
  - Add Erythritol: Stimulate Brucella growth
- Gm staining: GNCB
- Catalase, Oxidase, Urease positive
- Tblisi Phage Typing
- Serological Test
  - SAT (Standard Agglutination Test): Detects IgM Ab
  - 2 ME (Mercapto Ethanol) Test: Done for IgG Ab
  - PCR for Brucella species

Test for Brucella Contamination

- Rose Bengal card test
- Milk ring test
- Whey Agglutination Test

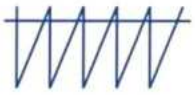
Treatment

- DOC:
  - Rifampicin + Doxycyclin (WHO)
  - Streptomycin + Doxycyclin (Harrison)

YERSINIA PESTIS

00:47:59

- Leads to Plague (Zoonotic infection)
- Culture Media: Ghee broth
  - Stalactite type of growth is seen



- Virulence Factors
  - F1 Ag
    - → Plasmid borne
    - → Heat labile capsular Ag
    - → Expressed at body temperature (37°C)
    - → Inhibit macrophage phagocytosis
  - Phospholipase
  - PH 6 Ag
    - → Fibrillar protein
    - → Binds to lipoprotein
  - LPS: Endotoxic activity
  - Siderophore
- Types of plague
  - **B** - **B**ubonic (IP: 2-7 Days)
  - **S** – **S**epticemic (IP: 2-7 Days)
  - **P** – **P**neumonic

How to remember

- B S P
  - → IP: 1-3 Days
  - → Most Dangerous

Bubonic Plague

- MC type
- MOT: Bite of Rat Flea → goes into Lymphatics → Multiplies
- IP: 2-7 days
- Formation of Buboes which are Regional LN (mostly Inguinal LN), they appear Tense & Tender
- Bubonic Plague can't spread from Person to Person

Pneumonic Plague

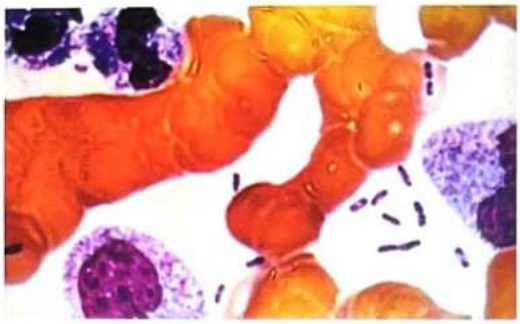
- IP: 1-3 days
- MOT: Inhalation of bacilli in droplets expelled from another person or an animal
- Rare: < 1%
- Agent of Bioterrorism
- Clinical Features: Fever, headache & pneumonia like features

Septicemic Plague

- Having LPS which contributes to Endotoxic shock
- IP: 2-7 days
- There is a Massive involvement of blood vessels leading to → Gangrene → Black death

Lab Diagnosis

- Sample taken
  - Bubonic: Buboes smear (Pus or Fluid)
  - Pneumonic: sputum & blood
  - Septicemic: Blood & splenic aspirate
- Transport Media: Carry Blair media
- On Direct Microscopy: Presence of pus cells & GNCB
- Wayson stain or Methylene blue stain
  - Bipolar stained appearance or Safety pin appearance



Bipolar Staining

- Culture
  - Ghee broth: stalactite type of growth
  - Shows Pleomorphism
- F1 Ag detection
- PCR for Y. Pestis

Treatment

- DOC: Streptomycin
- Prophylaxis: Doxycyclin

Important Information

- Cheopis Index is used for plague

Vaccine

- Haffkine vaccine
- Modification of Haffkine vaccine is Sokhey's modification (killed vaccine)
- Live attenuated is also there but not routinely given

YERSINIOSIS

- Zoonotic Infection
- Caused by Y. enterocolitica and Y. pseudoTB
- Y. enterocolitica
  - Virulence Factors
    - → Invasin protein
    - → Yersinia adhesin A (Yad A)
    - → PH6 Ag
    - → MyF Ag
- Y. pseudoTB
  - Virulence factors: Y. PseudoTB + enterocolitica together act as a Super Antigen leading to a massive cytokine release
- Clinical Features
  - Self limiting Gastroenteritis
  - Intestinal complications: Terminal ileitis which mimics appendicitis (Y. enterocolitica > Y. pseudoTB)
  - Septicemia
  - Post infective phenomenon
    - → Reactive arthritis (HLA B 27 positive)
    - → Erythema nodosum (HLA B 27 negative)
    - → Grave's disease: Y. enterocolitica mimics TSH binding site
- Specific Culture Media: Yersinia CIN agar (Cefsulodin Irgasan Novobiocin)
- Cold enrichment phenomena seen at 4°C
- They show, Differential motility
  - Motile at 22°C
  - Non motile at 37°C
- PCR for various Yersinia species

LEGIONELLA

01:14:05

Legionella pneumophila

- Most common serotype
- MOT
  - Aerosol route from centralized AC (best answer)
  - Drinking contaminated water with Legionella
- Diseases
  - Pontaic fever: self limiting illness
  - Legionnare's Pneumonia
  - Legionnare's Diseases: when Legionnare's pneumonia complicates with Encephalitis & Diarrhea
- Culture Media: Buffered Charcoal Yeast Extract [BCYE]
- PCR for Legionella Species
- In Urine: Legionella Ag can be detected
- Fluorescent microscopy
- Legionella micadedi is Acid Fast in nature
- DOC: Macrolides

Previous Year's Questions

Q. Legionnaire disease cause?

(FMGE - June - 2019)

A. Respiratory Disease
B. UTI
C. Retroperitoneal fibrosis
D. Acute Gastroenteritis

SPIROCHETES

01:19:55

- Spiral in shape
- It includes
  - Treponema
  - Borrelia
  - Leptospira
- Composition
  - Outer membrane
  - Periplasmic space containing flagella: Endo flagella
  - Peptidoglycan layer
  - Inner cytoplasmic membrane
- Observed on Dark Field Microscopy
  - Background is dark

- Reflected light is used

Dark field Microscopy

- Staining: Fontana & Levaditi Staining
  - Silver impregnation method
  - Makes them thick

	Treponema	Borrelia	Leptospira
• Spirals	6-12	3-10	Numerous Tightly coiled Hooked ends
• Endoflagella	3 - 4	7	1

1. TREPONEMA PALLIDUM

- Causes syphilis
- IP → 9-90 Days

Stages

- Primary
  - Hard, painless chancres
  - Painless Lymph adenopathy
- Secondary
  - Rashes over the palms & soles are present
  - Condylomata lata lesions are seen (Most infectious lesion of syphilis)
- Latency: for years
- Tertiary syphilis
  - Having Gummas [skin lesions]
  - Cardio vascular complication: Presents with Ascending Aorta Aneurysm
  - Neuro syphilis features
    - → GPI [General Paralysis of Insane]
    - → Tabes dorsalis

Lab Diagnosis

- Microscopy: Dark ground microscopy
- Direct Immuno Fluorescent staining
- Silver impregnated methods: L & F
- Mostly Non cultured but now can be cultured on Smith Noguchi media
- Serology:
  - Non Treponemal Tests
    - → VDRL [Veneral Disease Research Lab]
    - → RPR [Rapid Plasma Reagent]

VDRL	RPR
• Slide Flocculation Test • Microscopically read result • Ag to be used within 24 hrs • Preheating of serum • Test Blood, Serum, CSF • IOC for Neurosyphilis: VDRL - CSF • Cheaper	• Better Test o Card test o No Fluid is prepared • Test Blood, Serum but not CSF • In CSF sample, RPR Fails • Costly

  - Treponemal Tests (More Specific Test)
    - → TPHA [Treponema pallidum Haemagglutination Assay]
    - → TPPA [Treponema pallidum Particulate agglutination Assay]
    - → FTA – ABS [Fluorescent Treponemal Antibody Assay]
    - → TPI [Treponema pallidum Immobilization Assay]

Important Information

- **Most sensitive: FTA - ABS**
- **Most specific: TPI > TPPA**
- **Mc done in labs: TPHA**
- **IOC for Neurosyphilis: VDRL CSF > FTA - ABS CSF**
- **IOC for Congenital Syphilis: IgM FTA ABS assay**

Treatment

- DOC for Primary, Secondary & Latent stages of Syphilis: Benzathine Penicillin
- DOC for Neurosyphilis: Penicillin > Procaine Penicillin

Non Treponemal Pallidum

- T. Pertunae: causes YAWS
- T. endemicum: causes endemic syphilis aka Bejel
- T. carateum: causes PINTA

	[illegible]'aws	Endemic Syphilis	PINTA
MOT	Skin to Skin	Kissing, Sharing utensils	Skin to Skin
	Early childhood	Early childhood	Late childhood
	Ulcerative Papilloma Seen	Ulcerative Papilloma Rarely seen	Non-ulcerating papules are seen
Sites	Extremities	Oral	Extremities, Face
Relapses	Common	Not common	No relapse

2. BORRELIA

- Epidemic Louse Born Relapsing Fever: caused by B. recurrentis
- Endemic Tick Born Relapsing Fever: caused by B. duttoni and B. hermesii
- Relapsing Fever
  - Relapses are d/t Antigenic Variations
  - B. recurrentis undergo Antigenic variation
  - Last for 3-5 days
- Diagnosis: PCR for Borrelia species

B. Burgdorferi

- Transmitted by Ticks
- Causes LYME'S Disease
  - Characteristic lesion: Erythema Migrans aka Annular Bulls Eye Pattern Rash
- Lyme's Disease stages

Stage 1	Stage 2	Stage 3
• Early localized infection • IP: 3 to 30 days • Annular maculo papular lesion Develops at tick bite site • Erythema Migrans over thigh, groin	• Early dissemination • Blood Borne • Secondary annular skin lesions • Arthralgia • Profound fatigue • Neurological features: Bannwarth's syndrome o Meningitis o Typical Lymphocytic meningoradiculitis • Cardiac Involvement	• Late persistent infection k/a Lyme's arthritis • Large joints involved • Acrodermatitis Chronica Atrophicans (Late skin manifestation) • Post Lyme Syndrome: aka Chronic Fatigue Syndrome

Lab Diagnosis

- Culture
  - Blood & CSF
  - BSK media or Kelley's media
- PCR for Borrelia Burgdorferi
- Dark field microscopy

Treatment

- DOC: Doxycyclin (for all stages)
- Except in CNS/ CVS involvement:
  - DOC: Ceftriaxone

B. Vincentii & fusiformis

- Causes Vincent Angina / Trench Mouth
- Inflamed pharyngeal mucosa covered by greyish membrane
- DOC: Penicillin, Metronidazole

CLINICAL QUESTIONS

Q.1 A microbiology intern is studying growth of different bacteria using different culture media. Match the following bacteria with their appropriate culture medium?

1. Salmonella typhi enrichment medium	A. Hela cells
2. Bordetella pertusis	B. LJ medium
3. Chlamydia	C. Selenite F broth
4. Mycobacterium tuberculosis	D. Bordet gengou media

A. 1-c,2-d, 3-a, 4-b
B. 1-b, 2-c, 3- d, 4-a
C. 1-a, 2-d, 3-c, 4-b
D. 1-d, 2-A 3- b, 4-a

Solution:

Bacteria	Culture
S. typhi enrichment medium	Selenite F broth, Tetrathionate broth
Bordetella pertusis	Bordet gengou agar, Regan lowe medium
Chlamydia	Cell line culture in - McCoy , Hela cell, Hep2 cell
	LJ media (solid)
M. TB	Kirchner media, Middle brook 7H9 } Liquid Media

7 GRAM NEGATIVE BACILLI PART-3

LEPTOSPIRA

00:00:15

- L. interrogans (Most pathogenic for humans)
- L. biflexa
- MOT: Zoonotic Transmission

00:03:00

  - R - Rat urine
  - R - Rice [Paddy Field workers]
  - R - Rainy water
- No Human to Human transmission
- L. interrogans: causes WEIL's Disease aka Icterohemorrhagic fever

Clinical Stages

1.Mild illness

	First Stage	Second Stage
	• Septicemic phase	• Immune phase
	• 3-10 days	• 10-30 days
Features	• Fever, Myalgia, Conjunctival suffusion, Abdominal pain, Vomiting	• Meningitis, Uveitis, Chorioretinitis, Rash, Fever, Peripheral neuropathy
Isolation	• Blood & CSF	• From urine
Serum IgM	• Absent	• Present
Antibiotics	• Doxycyclin	• Refractory to Rx

2. Severe illness: Weil's Disease

00:07:45

	First Stage	Second Stage
	• Septicemic phase	• Immune phase
	• 3-10 days	• 10-30 days

Features	• High grade fever
	• Liver: Jaundice & increased Liver enzymes
	• Hemorrhages
	  ○ Pulmonary
	  ○ Skin
	  ○ Conjunctival
	• Kidney: Raised creatinine level and impending RF
		• All features of septicaemic phase can be present

Isolation	• Blood & CSF • No IgM • Sensitive to antibiotics	• From Urine • IgM present • Refractory to antibiotics

Lab Diagnosis

- Specimens: CSF, Blood, urine
- Microscopy: Dark ground Microscopy
- Isolation from culture media's: EMJH, Fletcher, Korthof
- Animal inoculation: Not done now
- Serological test: MAT (Microscopic Agglutination Test)
- IOC: PCR for Leptospira species
- Treatment
  - DOC: Doxycycline
  - For severe cases: Penicillin
- Dinger's ring: Leptospira produces a ring of organism, just under surface of medium

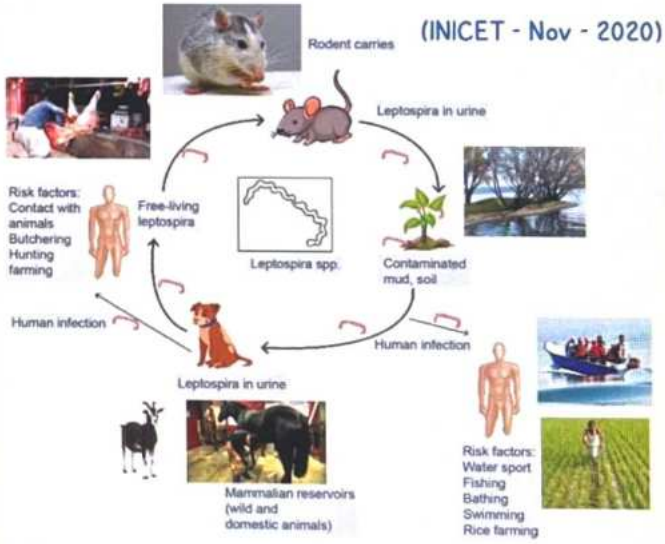
Faine's criteria (WHO approved guidelines)

- For diagnosis of leptospirosis
- It is a mixture of Epidemiological and Lab findings
- Non specific findings
  - Altered renal functions
  - Altered liver functions
  - Urinary sediments

Previous Year's Questions

Q. Transmission cycle of rat urine contaminated water, dog's urine, coiled bacterium?

(INICET - Nov - 2020)



A. Leptospira
B. Yersine
C. Hanta

RICKETTSIAE

00:22:19

- Obligate intracellular organisms
- Arthropod borne Transmission
- Mostly non cultured
- Fever, Rash, Lymph adenopathy

	Caused by	Arthropods involved
Epidemic Typhus	R. prowazekii	Louse
Endemic Typhus	R. typhi	Flea
RMSF	R. rickettsi	Tick
Indian Tick Typhus	R. conori	Tick
R. pox	R. akari	Mite
Scrub Typhus	Orientia tsutsugamushi	Mite

- Presents with fever, rash & lymphadenopathy
- DOC: Doxycycline
- Target Site
  - Endothelial Cells
  - R. akari, O. tsutsugamushi: Can attack monocytes also

Previous Year's Questions

Q. Trombiculid mite?

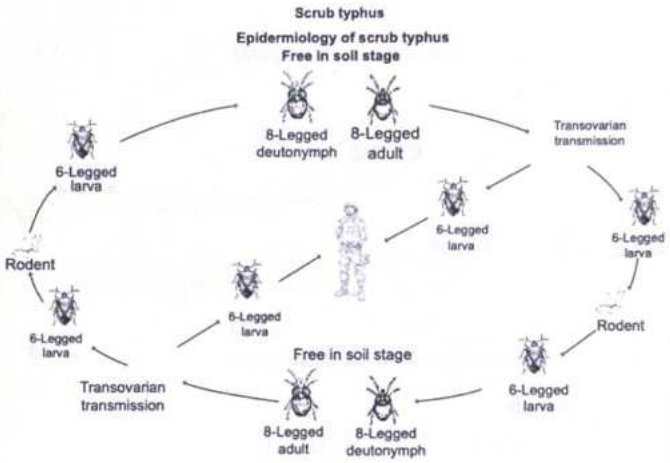
(AIIMS - Nov - 2019)

A. KFD virus
B. Chandipura virus
C. Orientia Tsutsugamushi
D. AnaplasmaPhagocytophilia

Previous Year's Questions

Q. Man was accidentally infected by 6 legged larvae. Life cycle of organism is given below. Identify organism shown?

(AIIMS - June - 2020)



A. Hard Tick
B. Soft Tick
C. Louse
D. Trombiculid mite

Tunica Reaction aka Neil Mooser's Reaction

00:29:25

- It is the intraperitoneal inoculation of the infected blood into the nail of a guinea pig leads to scrotal necrosis and inflammation, then the reaction is said to be +ve and its done by Endemic Typhus
- Differentiate between Epidemic typhus and Endemic Typhus
  - Scrotal necrosis & inflammation of guinea Pig: Endemic typhus
  - No such Reaction & inflammation in guinea pig: Epidemic typhus
- OX19 & OX2 +ve for both Tunica and Neil Mooser's reaction
  - These are non motile strains of proteus, cross reacts with Rickettsial Ab's and this is termed as Weil-Felix reaction (HAT)
- OXK is strongly positive in scrub typhus

O. TSUTSUGAMUSHI

00:32:33

- Causes scrub typhus
- Transmitted by Chiggerosis larvae

Scrub typhus

- Clinical Manifestations
  - Triad of Black Escher, Regional LAP, Maculopapular rash
  - Non-Specific Features: Fever, Headache, Myalgia
- In India its confined to Hilly region
- Zoonotic Tetrad
  - Trombiculid mite
  - Mice, rats
  - Scrub vegetations
  - Wet season
- Strongly +ve for OXK
- DOC: Doxycycline
- Diagnosis: PCR for Scrub typhus

COXIELLA BURNETTI 00:38:16

- Causes Q fever
- MOT: Aerosol transmission [no arthropod transmission]
- No Rash occurs
- Non cultured
- A/w culture negative endocarditis
- Pathogenesis
  - Escapes intracellular killing by
    - → Inhibiting final step of phagosome maturation
    - → Resist acidic PH of Phagolysosomes
- Clinical Manifestations
  - Acute 'Q' fever
    - → IP: 3-30 days
    - → Hepatitis, pneumonia, fever
    - → CNS & CVS involvement may be seen
    - → Post 'Q' Fever Fatigue Syndrome
    - → Latency: can be for 2-3 days
  - Chronic 'Q' Fever
    - → A/w Endocarditis
    - →Fever mostly absent
- Phase Variation
  - Phase 1: Fresh isolates (LPS surface)
  - Phase 2: On repeated passage in Yolk sac Surface Protein get Exposed and change to phase 2
- GNCB, Pleomorphic
- Isolation: in BSL – 3 lab
- Ab detection
  - Indirect IF testing
  - IgM: appears in 7-10 days
  - IgG: appears in 14-20 days of infection
- In Chronic Infection
  - IgG to Phase 1: Shows elevated levels (> 1: 6400)
- Diagnosis
  - CFT
  - PCR for C. burnetti
- DOC: Doxycycline

Previous Year's Questions

Q. Giemsa-stained smear cannot defect?

(AIIMS – Nov - 2019)

A. Coxiella burnetti
B. Bartonella
C. Toxoplasmosis
D. E. Chaffnessis

EHRLICHIAE.

Refer Table 7.1

BARTONELLA 00:54:50

- Can be cultured on normal culture media
- B. henselae
  - Caused CAT Scratch Disease
    - → Determined by Regional LAP and Painless Erythmatous papule
  - Caused Bacillary Angiomatosis [common in HIV +]: Neovascular lesion in skin and other organs
  - Skin lesions are caused by B. henselae and B. quintana
  - Hepatosplenic lesion caused by: B. henselae
- Bacillary peliosis
  - Angioproliferative disorder involving liver
  - Bacteriaemia& Endocarditis: Rare association
- B. Quintana
  - Causes Bacillary Angiomatosis [Fever cases]
  - Causes Trench Fever/ 5 day fever
  - By inoculation of louse feces
  - Seen in WW-I in 1919
- B. bacilliformis
  - Transmitted by sand fly
  - Also termed as Lutzomyia

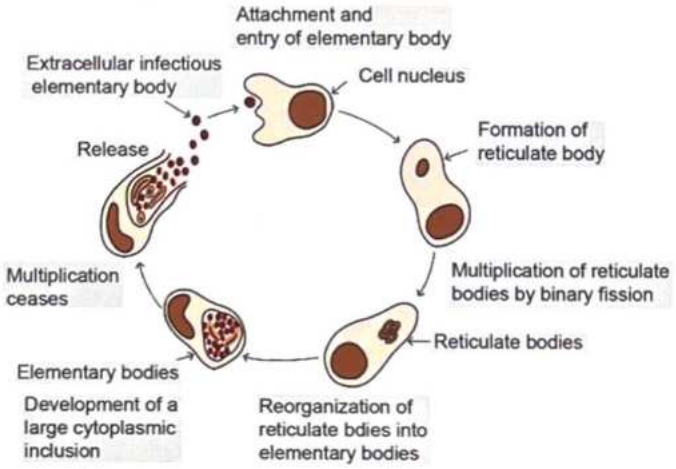
- Leads to oraya fever or Carrion's disease
  - → Initially: Bacteremia and Systemic illness can happen
  - → Lately: verruga peruara (Cutaneous vascular lesion)
- Treatment
  - Bacillary Angiomatosis: Erythromycin or Doxycyclin
  - Cat scratch disease: Azithromycin
  - Trench fever: Gentamycin

CHLAMYDIA

01:03:00

- Obligate intracellular organism
  Exists in

- Elementary body
  - Extracellular from
  - Infectious
- Reticulate body
  - Intracellular form
  - Replicating



Elementary and Reticulate Bodies

- Termed as: Basophilic viruses
- Pass through bacterial filter
- Non cultured
- Obligate intracellular
- Also termed as ATP parasite
- Responds to the Antibiotics so classified under bacteria
- Multiply by binary fission
- Contains prokaryotic 70s ribosome

1. C. TRACHOMATIS

- Serogroups A, B, Ba, C: causes TRACHOMA
- They are K/a TRIC agents
- Mx Strategy
  - **S** - Surgery
  - **A** - Antibiotics
  - **F** - Face clean
  - **E** - Environmental Clean
- Serogroups D-K: causes
  - Inclusion Conjunctivitis
  - Infantile Pneumonia
  - Genital Chlamydiasis
- Serotypes L_1, L_2, L_3: causes Lympho Granuloma Venerum [L2>L1]

Previous Year's Questions

Q. Incubation period of LGV's is?

(AIIMS - May - 2018)

A. 3-7 days
B. 7-10 days
C. 10-30 days
D. 30-90 days

2. C. PSITTACI

- Causes Bird Pneumonia
- Mainly associated with parrots
- Inclusion bodies: LCL (Levinthal Cole Lilee Bodies)

3. C. PNEUMONIAE

- Causes Adult Pneumonia
  - TWAR strain leads to adult pneumonia
  - It is an Atypical Community acquired pneumonia

Diseases

a. LGV
- $L_2 > L_1$
- Features
  - **A** - Asymptomatic
  - **B** - Buboes
  - **C** - C. trachomatis
  - **D** - DOC: Doxycyclin
  - **E** - Esthiomene (Rectal & Vulval Strictures)
  - **F** - Frie's test
  - **G** - Groove sign

How to remember

- A B C D E F G

b. Genital Chlamydiasis
- Done by D & K serotype
- Causes NGV: 30-50% MCC
- IP: 7-10 days
- Features
  - Mucopurulent discharge
  - Dysuria & urethral irritation

c. Reactive arthritis
- A/w Reiter's syndrome
- Occurs in 1-2% NGU cases
- M > F
- HLA-B27 strong association

d. Inclusion Conjunctivitis
- Looks like Ophthalmia neonatorum
- C. trachomatis > Gonococci
- IP: 6-21 days
- Mucopurulent discharge

e. Adult Inclusion Conjunctivitis
- Aka Follicular conjunctivitis / Swimming pool conjunctivitis

f. Infant Pneumonia
- 3 wks to 3 months of birth
- Infection develops in conjunctiva and goes into Pharynx
- Infection through Eustachian tube it can cause Otitismedia

Gram Staining
- Gm –ve (Poorly stained)
- Sterile Pyuria (No organism)
- Inclusion bodies
  - Trachoma: HP Bodies [Halberdt Prowazek]
  - Psittacosis: Levinthal Cole Lilee Bodies
  - LGV: Miyagawa corpusdes

Diagnosis
- IOC: NAAT/PCR for Chlamydia species

Treatment
- Adult conjunctivitis: Azithromycin (DOC)
- Uncomplicated genital infection: Azithromycin
- Complicated genital infection: Doxycyclin or Erythromycin
- Ophthalmia Neonatorum & Infant Pneumonia:
  - Macrolides: Erythromycin
  - Can also use Topical ointment
- Psittacosis: Tetracycline

Important Information

- Mcoy cell lines: preferred cell lines on which Chlamydia prefers to grow

CAMPYLOBACTER

01:26:26

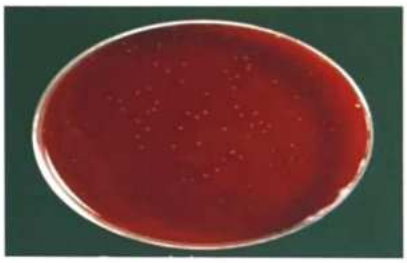
- GNB
- Gull wing shape
- Micro aerophile
- Motile: Darting motility

1. C. JEJUNI

- A/w causing the contamination of Poultry products
- Leads to Diarrheal episodes
- In Children
  - Diarrheal episodes followed by GBS (Gullian barre syndrome)
- Lab Diagnosis: Stool sample → Wet mount → Fecal leucocytes present → Inflammatory form of diarrhea

Gm staining
- Gull wing shaped GNB along with Leucocytes

Cultured on
- Campy BAP
- Butzler
- Skirrow
- All 3 are charcoal based media



- Incubation temp: 42°C

HELICOBACTER

01:31:38

- GNB, curved
- Normal Commensal of stomach

1. H. PYLORI

- Present in 50% of world human population
- Motile
- Virulence factor: flagella
- Acid resistance d/t
  - Urease enzyme

    → Urea $\xrightarrow{\text{Urease}}$ NH_3 → Neutralizes the gastric acid

  - Amidase & Arginase enzymes also contribute to acid resistance by producing NH_3
- Adhesins (adhere to mucus layer)
- Have 2 particular genes
  - VacA gene which code for Vacoulating cytotoxin
  - CagA gene is a Pathogenicity islands codes for type-IV secretion system
- These genes make H. pylori more pathogenic

Disease by H. pylori
- Antral gastritis
- Peptic ulcer disease
  - 80% leads to Duodenal ulcer
  - 60% leads to Gastric ulcer
- Chronic atrophic gastritis
- Autoimmune gastritis
- Pernicious anemia
- Adenocarcinoma of stomach
- Non-Hodgkin's gastric lymphoma

Protective role of H. pylori
- **A** - **A**denocarcinoma of Esophagus
- **B** - **B**arrett's esophagus
- **G** - **G**ERD
- **Analysis** - **A**sthma

How to remember

- ABG Analysis

Lab Diagnosis
- Non invasive test
  - Urea breath test: Sensitive, Quick and simple test
- Invasive procedures
  - Endoscopic biopsy
    - → Site: Antrum
    - → Biopsy tissue + Warthin Starry Silver Stain
    - → Culture on Skirrow media
    - → From gastric biopsy tissue: Rapid urease test can be done (highly sensitive)
- Biochemical test: Catalase, Oxidase and Urease positive (strongest urease positive organism)
- PCR for H. pylori can also be done

Rx Regimes
- OCA regime: Omiperazole / Clarithomycin / Amoxicillin
- OBM regime: Omiperazole/ Bismuth salicylate / Metronidazole
- OBTM regime: Omiperazole/ Bismuth salicylate / Tetracyclin / Metronidazole
- These are given for weeks together
- Helibact / Pylokits are available

Rat Bite Fever 01:44:48
- Characterized by High grad fever
- Rashes
- Painful Polyarthritis
- Done by
  - Streptobacillus Moniliformis
    - → Normal flora in throat of rats
    - → Leads to Haverhill Fever
    - → IP: 7-10 days
    - → DOC: Penicillin
  - Spirillum minus
    - → Sodoku Fever
    - → IP: 1-3 weeks
    - → DOC: Penicillin

FRANCISELLA TULARENSIS
- Causes Tularemia
- Plague like disease of rodents
- Zoonotic infections
- MOT
  - Contact with wild / domestic animals
  - Aerosols inhalation
- Clinical manifestations
  - UlceroglandularTularemia (MC)
  - Pulmonary Tularemia
  - Oropharyngeal Tularemia
- Also used as Agent of Bioterrorism → belongs to Category A bioterrorist weapon
- Highly infectious

Lab Diagnosis
- Difficult to Culture
- PCR for F. tularensis (Best)
- GNCB, Catalase +ve but Oxidase –ve
- Bipolar stained appearance can be seen

Treatment
- DOC: Gentamycin

MYCOPLASMA 01:51:38
- Smallest bacteria
- No cell wall
- Cell membrane is made of sterol
- Size

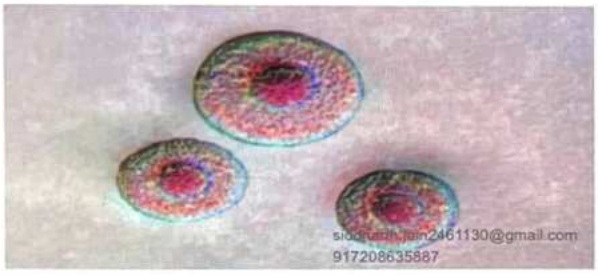
- Very small (150 to 350 nm)
- Can pass through bacterial filters

1. MYCOPLASMA PNEUMONIAE AKA EATON'S AGENT

- Causes Atypical pneumonia (MCC)

Culture media

- PPLO Agar
  - It is a complex agar
  - On PPLO agar they form Fried Egg colonies



Fried Egg colonies

- Standard Liquid media: PPLO broth
- SP- 4 Media
- Hay flick modified media

Staining

- The fried egg colonies on PPLO agar are not easy to see by naked eyes, so a unique type of staining is done K/a Diene's methord of staining
- Diene's methord of staining is done for Mycoplasma
- Diene's Phenomenon is seen in Proteus

Other tests

- CAT [cold Agglutination Test]: type of Heterophile Agglutination test'
- Streptococcus MG Agglutination Test: Type of Heterophile Agglutination Test
- Bothe the tests are obsolete now
- Sample: sputum, Throat swabs, NP swabs
- Antigenic detection
  - Direct IF testing
  - PCR for different Mycoplasma species

Extra Pulmonary Manifestation of M. pneumonia

- Aka Walking Pneumonia
- Extrapulmonary features:
  - Neurologic features
    - → Meningo encephalitis
    - → GBS
    - → Aseptic Meningitis
  - Dermatological features
    - → Skin rashes: Erythema multiforme (major) aka SJS
  - Cardiac features
    - → Myocarditis
    - → Pericarditis
  - Rheumatologic features
    - → Reactive arthritis
  - Hematological features
    - → Anemia &Hypercoagulopathy

Previous Year's Questions

Q. Which of the following statement is not correct regarding mycoplasma pneumoniae?

(AIIMS - June - 2020)

A. Cannot be cultured easily
B. Serology is useful for diagnosis
C. Respond well to amoxycillin + clavulanic acid
D. Cause bilateral chest infiltrates on CXR

2. M. GENITALIUM & M. HOMINIS

- A/w Non-Gonococcal Urethritis (3rd MCC)
- 2nd MCC: Ureaplasma Urealyticum
- MCC: C. trachomatis
- Also a/w Reiter's syndrome
- DOC: Macrolides

How to remember

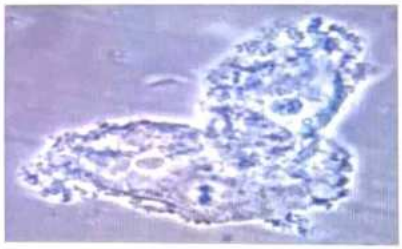
CAT SAT on a MAT

- CAT (cold agglutination test): Done for Mycoplasma
- SAT (Standard agglutination test) : Done for Brucellosis
- MAT (microscopic agglutination test): Done for Leptospira

BACTERIAL VAGINOSIS

02:06:42

- Caused by
  - Gardanerella vaginalis
  - Mobiluncus
  - Prevotella
- Amsel's Criteria
  - PH> 4.5
  - Greyish white Discharge + 10%KOH gives a fishy smell and this test is K/a Whiff Test
  - Clue cells: epithelial cells with bacteria



...ue cells

- Nugent's Criteria
  - Replacement grading of normal Vaginal Flora by Pathogenic Organisms like Gardanerella, Mobiluncus, Prevotella
- DOC: Metronidazole

BACTEROIDES

02:10:03

- GNB
- Anaerobe obligate
- B. fragilis
  - Normal commensal of gut
  - May lead to infection in those who undergo abdominal / gut Surgery
- A/w Brain abscess
- Foul smelling pus → anerobic growth
- Bacteroids have LPS because of which it has an Endo toxic action
  - Mild potent Endotoxin
  - May lead to 'Shock & DIC'

MISCELLANEOUS

- Pasturella Multocida: Associated with Cat and Dog bites
- Eikenella Coordens And Anerobic Streptococci: Associated with Human bites
- Streptobacillus Moniliformis: Causes HAVERHILL fever
- Spirillum Minus: causes Rat Bite Feve

Non Sporing Anaerobes

- Gram positive
  - Engineers: Eubacterium
  - Loe: Lactobacillus
  - M: Mobiluncus
  - B: Bifiodobacterium
  - A: Actinomyces
  - Program: Propionibacterium acnes

How to remember

Engineers Loe MBA Programme

- Gram negative
  - B - Bacteroids
  - P - Prevotella, Prophyromonas
  - L - Leptotrichia
  - Fuse – Fusobacteriums

How to remember

BPL Fuse

Table 7.1

E. chaffenensis	E. ewingii	Anaplasma phagocytophyllum	Neorichettsiae Sennetsu
• Causes HME (Human Monocytic Ehrilichiosis) • Features o Leucopenia o Thrombocytopenia o Elevated Liver Enzymes • Transmitted by Tick	• Causes HGE (Human Granulocytic Ehrilichiosis) • Features same as E. chaffenensis, but less severe • Transmitted by tick	• Causes HGA (Human Granulocytic Anaplasmosis) • Features o Leucopenia o Thrombocytopenia • Transmitted by tick	• Causes HLE (Human Lymphocytic Ehrilichiosis) • Mononucleosis Like Syndrome • Transmitted by Fish Ingestion (infected flukes

CLINICAL QUESTIONS

Q.1 A 35 years old patient complains of abdominal cramps along with dysentery. The treating physician wants to process the stool culture for isolation of Campylobacter jejuni. Which of the following is a correct growth requirement for Campylobacter jejuni?

A. Incubate at 37°C, microaerophilic
B. Incubate at 42°C, microaerophilic
C. 5% CO_2 is required
D. Incubate at 37°C and 10% capnophilic

Solution:

- Campylobacter jejuni is a **microaerophile i.e it needs only 5% O_2. It grows at 42°C.**
- **Causative agents of Predominantly dysentery:**
  - Shigella species
  - Campylobacter jejuni
  - Enteroinvasive E.coli
  - Vibrio parahaemolyticus

Causative agents of Predominantly inflammatory diarrhea:

- Salmonella species
- Yersinia enterocolitica
- Listeria monocytogenes
- Clostridium difficile
- Aeromonas hydrophila
- Plesiomonas shigelloides

Characteristics of Campylobacter jejuni:

- Motile Gram negative bacteria.
- It is associated with poultry bird product contamination.
- **Gull wing shaped** bacteria.
- Motile
- causes Inflammatory Diarrhoea - Fecal leucocytes are present
- In children, C. jejuni leads to Gullian Barre syndrome.

Culture done on **Charcoal based medias**:

- CAMPY BAP
- BUTZLER
- SKIRROW → incubate at **42°C**

Other Examples of microaerophilic bacteria :-

- Helicobacter pylori
- Mycobacterium bovis

LEARNING OBJECTIVES

UNIT 3: MYCOLOGY

- Mycology
  - Introduction
    - → Classification
    - → Reproduction
    - → Fungal infections
  - Superficial mycosis
  - Subcutaneous mycosis
  - Systemic mycosis
  - Occupational fungal disease

8 MYCOLOGY

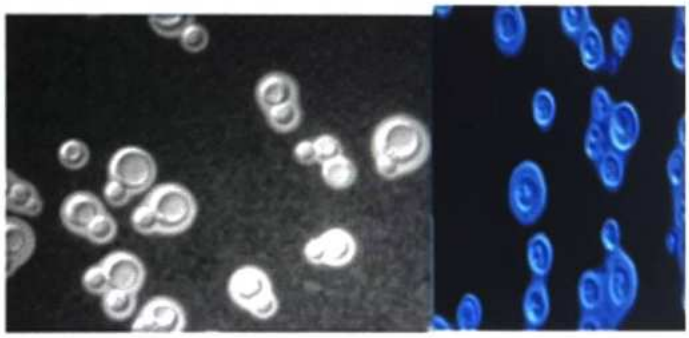
INTRODUCTION TO MYCOLOGY

- Fungi are eukaryotic protista
- Cell walls contain chitin (rigidity and support), mannan & polysaccharides
- Cytoplasmic membrane contain ergosterols
- Possess true nuclei, nuclear membrane & paired chromosomes

Classification

1. Depending on Cell Morphology ⏱ 00:02:25

- Yeast / True Yeast
  - Unicellular fungi which reproduce by budding
  - On culture produce smooth, creamy colonies
  - Spherical or Ellipsoidal cells
  - Eg: Cryptococcus neoformans (Capsulated yeast)
    - → India ink staining: specifically done for Cryptococcus
    - → MCC of meningitis in an HIV +ve individual

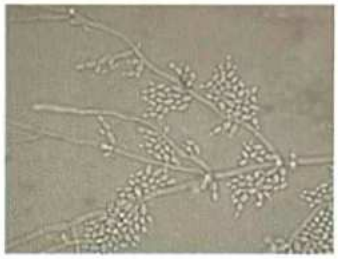


Previous Year's Questions

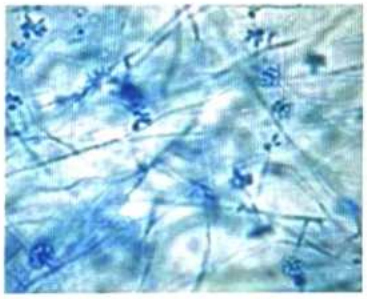
Q. Identify organism stained by India ink dye & shown below? (FMGE – Dec - 2020)

A. Cryptococcus
B. Chlamydia
C. Histoplasma
D. Aspergillus

- Yeast Like
  - Partly as yeast and partly as elongated cells resembling hyphae which are called pseudohyphae
  - Eg: Candida Albicans
    - → Can show true hyphae / pseudohyphae
    - → Can show yeast/yeast like [main form]



- Molds
  - Form True mycelia and reproduced by Spores
  - Eg: Aspergillus, Zygomycetes (Mucormycetes)



- Dimorphic Fungi: Dimorphism is based on temperature
  - Yeast at 37°C
  - Mold at 25°C
  - Most fungi causing systemic infection are dimorphic Except: Sporotrichosis
  - E.g. of Dimorphic fungal diseases
    - → **H** - Histoplasmosis
    - → **S** - Sporotrichosis
    - → **B** - Blastomycosis
    - → **C** - Coccididomycosis
    - → **P²** - Para coccididomycosis, Penicilliosis

How to remember

- HSBCP²

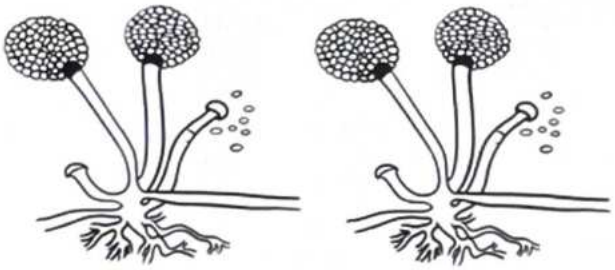
Previous Year's Questions

Q. Which is not a dimorphic fungus? (AIIMS - Nov - 2017)

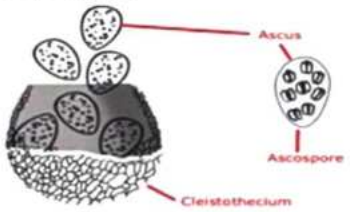
A. P. Marneffei
B. Histoplasma Capsulatum
C. Blastomyces Dermatitidis
D. Pneumocystis Jirovecii

2. Based on Sexual Spore formation 00:07:21

- Z – Zygospores
  - Found is Zygomycetes family



  - → Lower fungi
  - → Broad, nonseptate hyphae
  - → Sexual spore: Zygospore
  - → Asexual spores: Sporangiospores, present within a swollen sac – like structure called as sporangium
  - Eg. Rhizopus, Mucor
- A – Ascospores
  - Found is Aspergillus



Ascospores in an ascus

- B – Basidiospores
  - Sexual fusion of Basidiomycetes results in formation of a club shaped organ called base or basidium which bear spores called as Basidiospores
  - Found in Cryptococcus

Important Information

- Deuteromycetes
  - Aka fungi imperfecti
  - No sexual phases
  - Grows as molds as well as yeast
  - Asexual stage: Conidia
  - Eg. Candida, Cryptococcus

3. Based on location

- Superficial Mycosis
  - Involves the superficial layer of skin
  - Eg. Tenia Versicolor, Dermatophytes
- Subcutaneous Mycosis
  - Skin and subcutaneous tissue is involved
  - Eg.
    - → M - Mycetoma
    - → R - Rhinosporidiosis
    - → C - Chromoblastomycosis
    - → S - Sporotrichosis

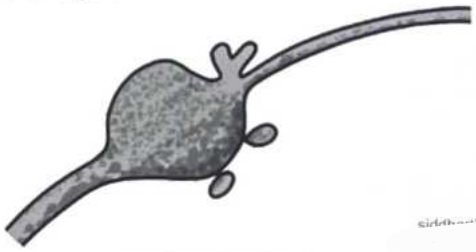
How to remember

- MRCS

- Deep / Systemic mycosis
  - All dimorphic fungal disease comes under deep / systemic mycosis
  - Zygomycetes
  - Aspergillus
  - Candida and Cryptococcus

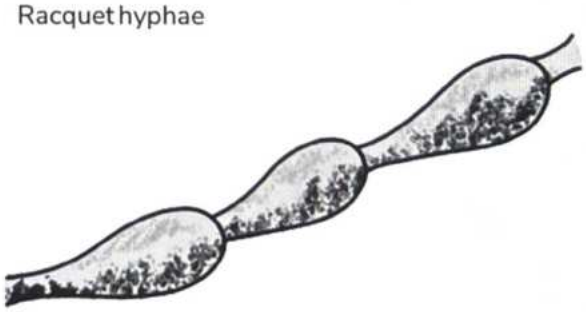
Vegetative structures of fungus

- Formed by the vegetative Mycelia (modification of single vegetative cells or hyphae)
- Have no reproductive value
- Important in the differentiation/ identification of clinically significant fungi
- They include
  - Nodular organ



A. Nodular organ

  - Racquet hyphae

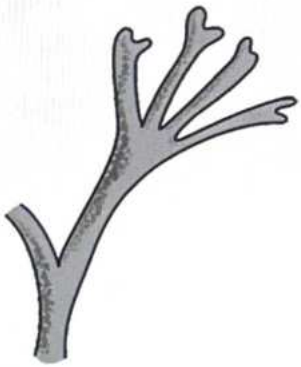


B. Racquet hypha

- Pectinate body

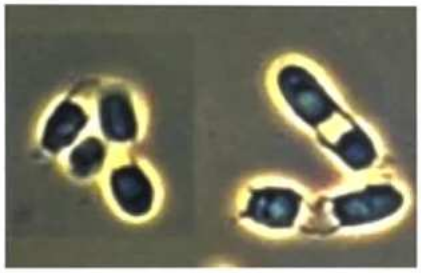
C. Pectinate body

- Favic chandelier



D. Favic chandelier

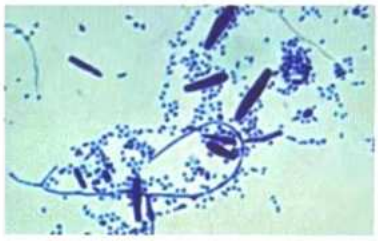
- Spiral Hyphae seen in T. Mentagrophytes
- Arthrospores: Formed by segmentation and condensation of hyphae



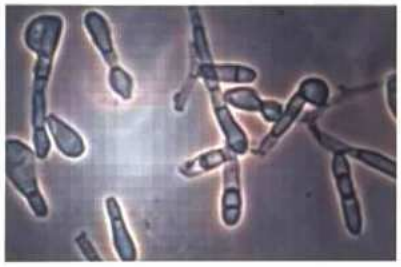
- Chlamydospores: thick walled resting spores → Eg. C. albicans

Reproduction in fungi

- Sexual: by formation of Zygospores, Ascospores or Basidiospores
- Asexual
  - By budding or fission
  - Spores (conidia) are formed on or in specialized structures
  - Spores vary in size, shape and colour but these characteristics are constant for a particular species
- Micro conidia: Small, single celled



- Macro conidia: large, single or many celled



Fungal infections / Mycoses

- Superficial mycoses
  - 2 types: Surface and Cutaneous mycosis
  - Skin, hair and nails
  - Mild but chronic disease
- Deep mycoses
  - 2 types: Subcutaneous and systemic mycoses
  - Caused by soil saprophytes
  - Infection is accidental
  - Range from a symptomatic infection to fatal disease

Opportunistic infections

- Pts with debilitating disease, altered physiological state
- Mainly caused by fungi which are common lab contaminant on culture media
  - Aspergillus
  - Pencillium
  - Mucor
  - Rhizopus

Useful properties of fungi

- Source of food Eg. Mushrooms
- Fermentation Eg. Sacchromyces species
- Antibiotic production Eg. Penicillin from penicillium notatum

- Ergot from Claviceps purpurea, used to induce uterine contraction
- Vaccines for hepatitis B: Sacchromyces cervisiae

Lab Diagnosis

- Done to confirm clinical suspicion to establish fungal cause of disease
- To help in
  - Choose a therapeutic agent
  - Monitoring the course of disease
  - Confirming mycological cure

Sites and Types of specimens

- Specimen collection depends on the corresponding disease
- Very important to proceed for a final diagnosis
- For superficial mycosis
  - Clean the part with 70% alcohol
  - Collect the material in a sterile paper or a sterile petridish to
    - → Allow drying of the specimen
    - → Reduce bacterial contamination
    - → Maintain viability
  - Dermatophytic lesion: spreads outwards in a concentric fashion with healing in the centre, scrape outwards from the edge of the lesion with a scalpel blade or use Cellophane tape
  - Scalp lesion
    - → Scraping with a blunt scalpel, including hair stubs, scales and contents of plugged follicles
    - → Wood lamp's examination of infected hair: Fluorescence
    - → Hairbrush sampling technique
  - Onchomycosis: stop antifungal for one week prior to collection
  - Mucosal infection: Mucosal scrapings
- For Subcutaneous Mycosis
  - Scrapings or crusts from the superficial parts of lesions
  - Pus aspirates
  - Biopsy
- For systemic mycosis
  - Pus, Tissue Biopsy, feces, urine, sputum, CSF, Blood
  - Scrapings or swabs from the edge of lesions.

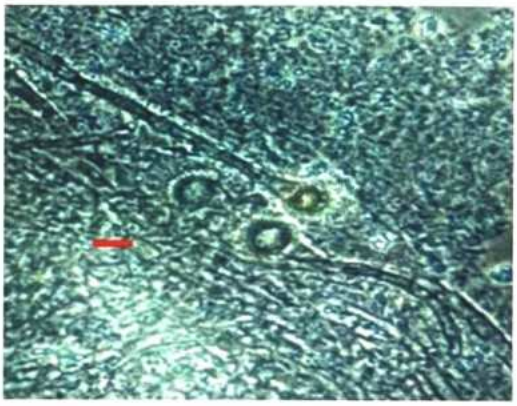
Collection and transport of specimen

- Proper collection of specimen and in adequate quantity
- Early transport to the lab to avoid overgrowth of contaminant
- Respiratory specimens
  - Sputum: Early morning sample, after mouth wash, flakes to be used for cutting
  - Bronchoscopy: If non productive cough
  - Bronchial brushing or lung biopsy: to rule out invasion or colonisation
- Blood
  - In biphasic Brain Heart infusion agar
  - Inoculated in 2 bottles – for dimorphic fungi
- Cerebrospinal fluid
  - Should be immediately processed else store at Rt or at 30^{0} C in an incubator
  - Centrifuge and use sediment for culture
- Skin, Hair and Nail
  - Taken for Dermatophytic infections
  - Hair – plucked with forceps
- Tissue, BM and Body fluids
  - Tissues: grind or mince before culturing
  - Body fluids: centrifuge and use sediments for culture
- Urine: centrifuge and use sediment for culture

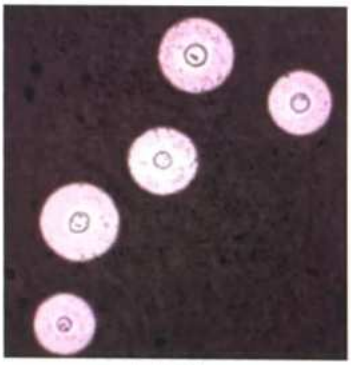
Laboratory diagnosis

1. Direct examination

- Very decisive in the diagnosis of fungal infections
- Wet mounts
  - Slide and tube KOH mounts: 10 to 20%KOH – digests protein debris, dissolves keratin. DMSO can be added to KOH to hasten clearing in skin scrapings and nail clippings



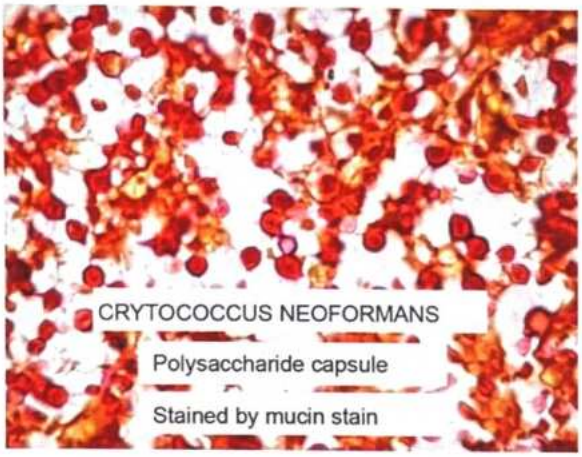
  - Calcofluor white: fluorescent stain – excellent morphology of pathogenic fungi
  - India ink: capsulated fungi



India ink-
Cryptococcus

- Gram stain – Fungi are Gram +ve

- Other Stains used
  - H and E stain
  - GMS: It is more advantageous since it stains old and nonviable fungal elements more efficiently than the others
  - Mayer's Mucicarmine stain: It stains the mucopolysaccharide capsule of Cryptococcus Neoformans



  - PAS (Periodic Acid - Schiff)

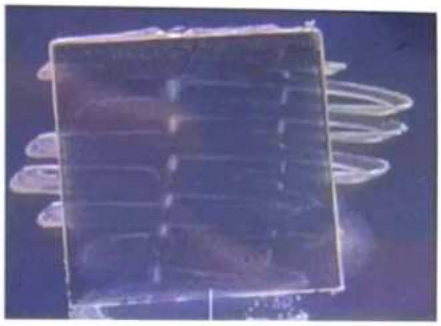
  - Fluorescent – antibody staining
    → To detect fungal Ag in clinical specimen such as pus, blood, CSF, tissue sections
    → Advantage – can detect fungus even when few organisms are present
    → E.g. Calcofluor white stain

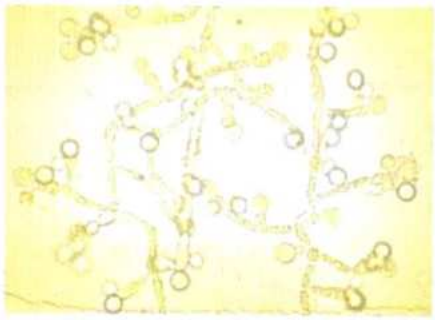
  - Alcian blue
  - LPCB – lactophenol cotton blue
  - India ink stain

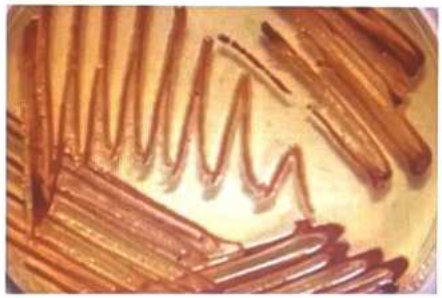
2. Fungal Culture

- Sabouraud dextrose Agar (SDA)
  - Contain 2% dextrose, antibiotics (Gentamycin, Chloramphenicol) and cycloheximide
  - PH around 5.6

- Selective media
  - Corn meal agar(CMA): Sporulation, Chlamydospores formation
  - Bird seed agar: Cryptococcus, forms brown colonies because of release of enzyme Phenol oxidase
  - Brain Heart infusion (BHI) agar: Dimorphic and other fastidious Fungi



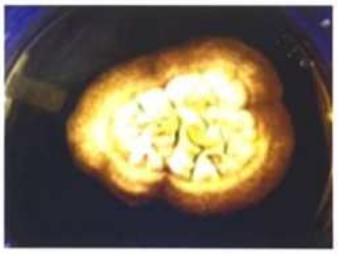


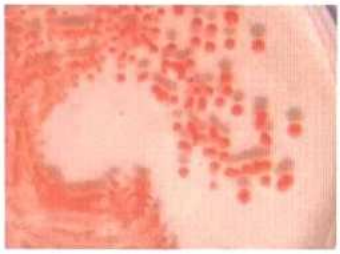


- Temperature requirement
  - Majority of Fungi: 37°C
  - Superficial mycosis: 30°C
  - Dimorphic Fungi: 25°C and 37°C

- Incubation time
  - At least 4 weeks
  - Usually positive culture are obtained in 7 – 10 days
  - Candida and Aspergillus: 24 to 72 hrs

- Identification of fungal cultures

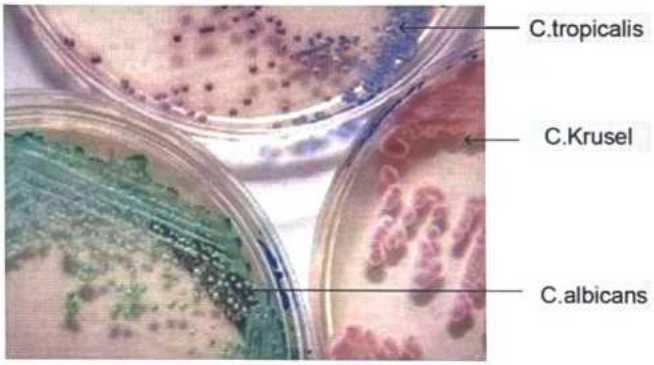




  - Colony morphology: Colour, texture, pigment
  - Fungal morphology: Under microscope using Lactophenol Cotton Blue (LPCB) stain
  - Composition of LPCB
    → Lactic acid: preserves fungal structure
    → Phenol: kills any living organism

→ Glycerol: prevents drying
→ Cotton blue: imparts blue colour to structures

- Special culture techniques: Slide culture to see sporing structure and spore arrangement, CHROM agar for Candida species



CHROM Agar

- Biochemicals: Ability to assimilate carbon and nitrogen, sugar fermentation

3. Serology and immunology

- Detection of Ag or Ab in serum or body fluids
  - Ab detection
    → Diagnosis of systemic and subcutaneous mycosis
    → Assess prognosis of the disease
    → Assess response to treatment
  - Ab detection
    → Early stages of infection
    → In patients with impaired immunity

- Delayed hypersensitivity test – with Ags like candidin, histoplasmin, etc.

- Agglutination
  - Whole cell agglutination
  - Latex particle agglutination
  - Passive Haemagglutination
- Immunodiffusion – most widely used
- Counter immunoelectrophoresis (CIEP)
- Indirect fluorescent Ab detection
- ELISA, RIA

4. Other methods

- PCR: Polymerase chain reaction
- RELP: Restriction fragment length polymorphism
- Protein electrophoresis
- Nucleic acid probes
- Serotyping
- karyotyping

SUPERFICIAL MYCOSIS

1. TINEA VERSICOLOR

- Caused by Malasezzia furfur
- Present as hypo to hyper pigmented scales over the skin
- Lab diagnosis
  - Scales are mixed with 10% KOH → Spaghetti and Meat ball appearance

  - Scales + SDA and Olive oil overlay: Fried egg colonies
  - Scales in wood lamp examination: yellowish fluorescence

- Treatment
  - Topical: 10 % sulphur ointment, 1-2% imidazole creams, selenium sulphide shampoo
  - Systemic: Itraconazole, Fluconazole
  - Ketoconazole with zinc pyrithione in a shampoo base is very effective for dandruff

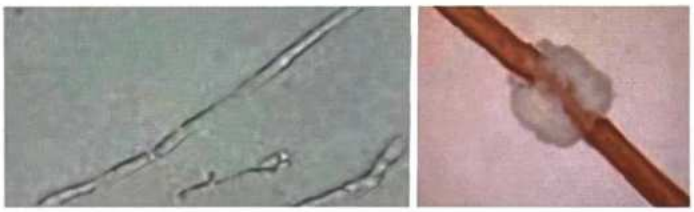
- Seborrheic dermatitis
  - Whitish, dry, loose flakes on scalp. Generally called as Dandruff
  - Caused by Malasezzia Furfur

2. TENIA NIGRA

- Blackish colour regions over the palms and sole
- Done by fungus Hortae Werneckii

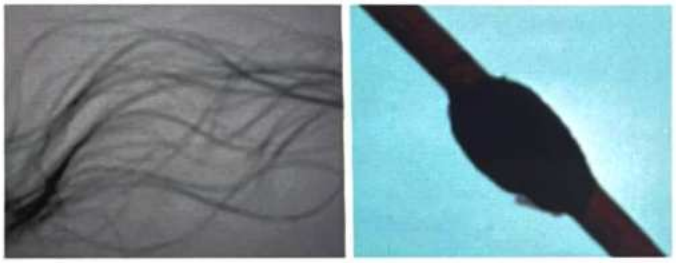
3. PIEDRA

- Two types
  - White piedra



→ Caused by: Trichosporon Beigelii
→ On KOH mount of hair: Hyphae and rectangular Arthrospores within and around hair

  - Black piedra



→ Caused by: Piedrae hortae

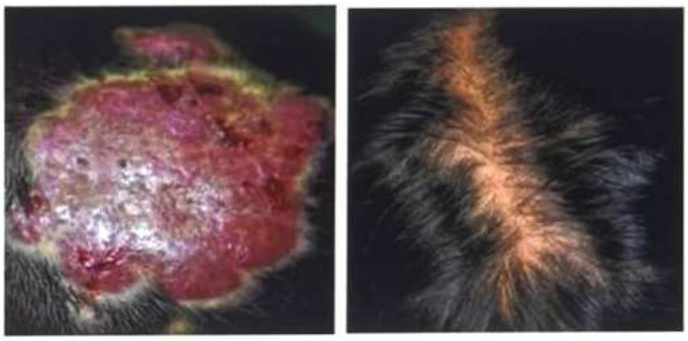
4. DERMATOPHYTOSES

- Popularly called as Tinea or Ring worm infection
- Involves: Skin, Hair, and nails
- 3 genera
  - Trichophyton: Skin, Hair, and Nails
    → Eg. Trichophyton rubrum, Trichophyton Mentagrophytes
  - Microsporum: Skin and Hair
  - Epidermophyton: Skin and Nails

- Various Dermatophytic infections
  - T. capitis: Scalp involvement
  - T. barbae: Beard involvement
  - T. Ungum: Nails
  - T. mannum: Hands
  - T. cruris: Groin.
  - T. pedis: Athlete's foot
    → Caused by: T. rubrum, E. floccosum

a. T. capitis

- Infection of the shaft of hairs of scalp & may lead to Alopecia & scarring
- Inflammatory → Kerion, Favus
  - Kerion (Honey Comb)



→ Painful boggy swelling over the scalp
→ Painful severe form of inflammatory reaction
→ Caused by Trichophyton verrucosum, T. Mentagrophytes

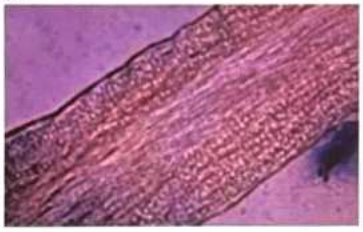
  - Favus
    → Dense crust formation over the scalp
    → Caused by Trichophyton Schoenleinii

- T. capitis has Ectothrix and Endothrix
  - Ectothrix

ECTORTHRIX OF HAIR - arthrospores form a sheath or appear as chains on the surface of hair shaft

→ Sheaths of spores around the shaft
→ Caused by Microsporum canis, M. audounii

- Endothrix

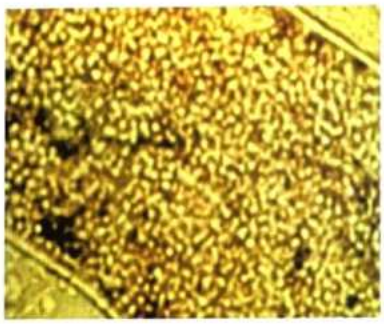


ENDOTHRIX OF HAIR- arthrospores present within the hair shaft.

→ Spores within the hair shaft
→ Caused by
  - T - Trichophyton Tonsurans
  - V - Trichophyton Violacaeum
  - S - Trichophyton Schoenlenii

How to remember

- TVS



b. **Tinea Cruris/Dhobi Itch / Jock Itch**

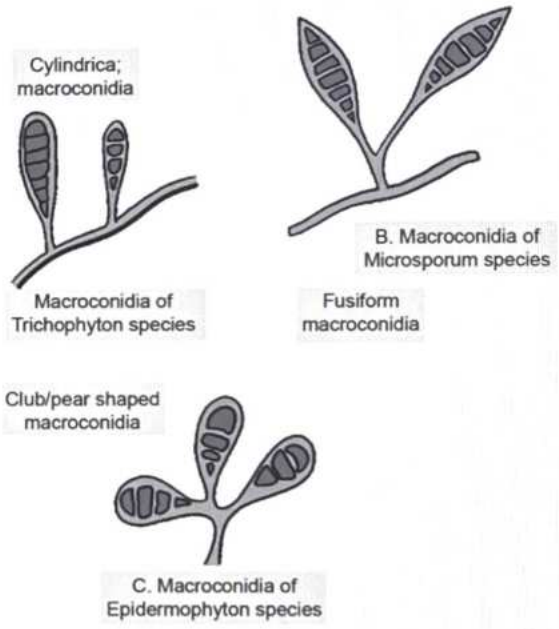
- Caused by: E. floccosum, T. rubrum
- Infection of groin → seen in men
- Involves Perineum, Scrotum, Perianal regions

c. **Tinea Pedis [Athlete's Foot]**

- Toe web scaling, fissuring, maceration
- Also Known as sandal ringworm

Lab Diagnosis of Dermatophytes

- Skin Scrapings, Nail Scrapings, Infected Hair +10% KOH→ Fungal Elements are Observed
- To confirm the species
  - Sample + SDA or Sample can be put on DIM or Sample can be put on DTM → Growth → LPCD mount of this growth → under microscope can observe whether its Trichophyton, Microsporum or Epidermophyton



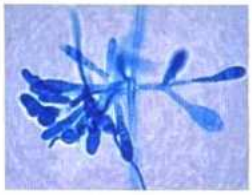
→ Trichophyton: Cylindrical Macroconidia and Numerous Microconidia
→ Microsporum: Spindle shaped Macroconidia and Less Microconidia
→ Epidermophyton: Club / Boat shaped Macroconidia and Absent Microconidia

- Id reaction – hypersensitivity to Dermatophytic fungal antigens and its mostly away from the site

Previous Year's Questions

Q. The LPCB mount shown below is of?

(FMGE - Aug - 2020)



A. Aspergillus Niger
B. Epidermophyton
C. Rhizopus
D. None of the above

d. **Tinea Corporis**

- Infection of non–hairy skin of the body
- Erythematous scaly lesions, sharp margins and raised borders

e. **Tinea Imbricata**

- Unusual form of tinea Corporis
- Concentric rings of scrapings which spreads out peripherally over years

f. **Tinea Gladitorum**
- Infection of wrestlers and athletes
- Direct skin to skin contact
- Lesions on arm, trunk or head and neck

g. **Tinea incognito**
- Steroid modified tinea
- Misuse of corticosteroid in combination with topical antifungals

h. **Tinea Facici**

- Infection of skin of face excluding infection of beard area

i. **Tenia Barbae**

- Infection of beard and moustache area of the face including the hair

j. **Tenia Mannum**

- Infection of skin of hands
- Diffuse hyperkeratosis

k. **Tenia Unguium**

- Infection of the nail plates
- Commonly seen in adults

Important Information

Onychomycosis
- Infection of nail
- Caused by
  - M/c by Dermatophytes
  - Other organism - Candida

Treatment and Prophylaxis of Dermatophytes
- Topical antifungals: Azole derivatives, Whitfield's ointment
- Oral Griesofulvin is the DOC
- Itraconazole is preferred for Onychomycosis

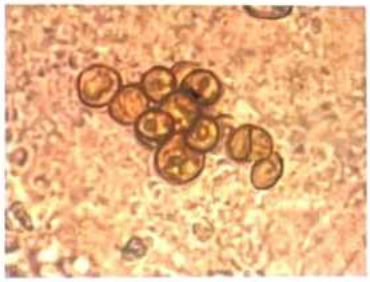
SUBCUTANEOUS MYCOSIS
- Involves skin and subcutaneous tissue

1. MYCETOMA
- Aka Madura foot
- They are 2 types

Actinomycetoma	Eumycetoma

• Multiple sinus and swelling
• Purulent discharge containing granules
  ○ Granules are crushed to observe <1 micron bacilli
• Red coloured Granules
  ○ Actinomycetes Madurai palletri
• Black coloured Granules and White coloured Granules seen in
  ○ Actinomadura madurai
  ○ Nocardia
• Yellow coloured granules can also be seen
• Treatment: Antibiotics
	• Single sinus and swelling
	• Serous discharge containing granules
	  ○ Granules are crushed to observe >1micron fungal hyphae
	• Granules are mainly black seen in
	  ○ Madurella griesia
	• White granules are also present in some
	  ○ Acremonium Pseudoallescheria boydii
	• Treatment: Amputation

2. CHROMOBLASTOMYCOSIS
- Aka Dematiaceous fungus or Pigmented fungal disease
- H/O wood worker or Thorn prick causing a Verrucous lesion (warty lesion on the skin)
- On tissue biopsy and staining of lesion with H and E stain there is presence of Medlar bodies / sclerotic bodies having a Muriform cell appearance or copper penny appearance



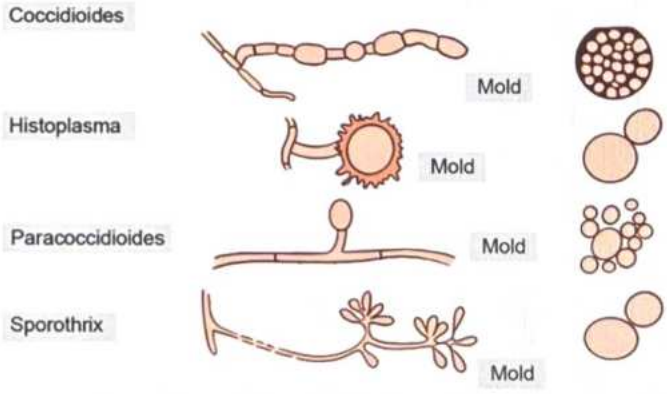
Previous Year's Questions

Q. A patient presented with some unknown fungal infection. Microscopic examination revealed brown coloured spherical fungi with septate hyphae. Possible conditions? (FMGE - June - 2019)

A. Histoplasmosis
B. Chromoblastomycosis
C. Coccidioidomycosis
D. Candida albicans

3. SPOROTRICHOSIS
- Caused by sporothrix schenki

- H/o
  - Rose gardener getting a thorn prick
  - Traveller gone to Himalayas walking bare foot
- It's a Dimorphic fungus having Both Yeast and mold forms
  - Mold form have – flower like Sporulation

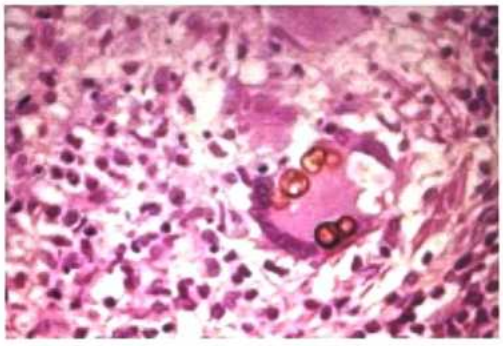


- Yeast form have narrow based budding yeast cell or cigar shaped yeast cell

- M/c Type - Lymphocutaneous type
- Asteroid bodies are seen which are characteristic of sporotrichosis

Previous Year's Questions

Q. 40-year-old farmer was struck with a wooden stick while working in the forest of Himachal Pradesh. The wound later became deep infection & involved the surrounding lymphatics too? (AIIMS – Nov - 2017)



A. Chromoblastomycosis
B. Histoplasmosis
C. Sporotrichosis
D. Penicillium Marneffei

Previous Year's Questions

Q. Which of the following is most likely to be acquired by traumatic inoculation? (NEET - Jan - 2020)

A. Blastomyces Dermatitidis
B. Sporothrixschenkii
C. Coccidioides immitis
D. Paracoccidioides Brasiliensis

Previous Year's Questions

Q. Gardner noticed multiple vesicles on his right leg along the lymphatics. What is the probable cause for this? (AIIMS - June - 2020)

A. Sporothrixschenkii
B. Histoplasma capsulatum
C. Candida albicans
D. Rhinosporidiumseeberi

RHINOSPORIDIOSIS

- Causative organism: Rhinosporidium seeberi which is an aquatic protist
- Mostly present with U/L or B/L nasal polyp
- On polyp biopsy spherules (sporangium) can be observed
- Inside the spherules there are endospores (typical of fungus)
- Treatment:
  - Initially: Dapsone
  - Later on: surgical Polypectomy using FESS
- It is not a fungus because it is Non cultured on SDA

SYSTEMIC MYCOSIS

1. CANDIDIASIS

- Candida affects: mucosa, skin, nails and internal organs and leads to superficial and deep infections
- Normal flora of skin, GIT and female genital tract
- HIV +ve individuals
- Predisposing factors
  - Natural receptive states like infancy, old age, pregnancy.
  - Changes in local bacterial flora secondary to antibiotics.
  - Endocrine disease like DM
  - Severe chronic underlying debilitated conditions
  - Malignancy
  - Drugs: Steroids, immunosuppressant and chemotherapeutic agents.
  - Neutropenic patients and any T cell deficient disorders
  - Trauma, burns or injury
- Pathogenesis and Pathology
  - Adhesion: entry into host as yeast cell

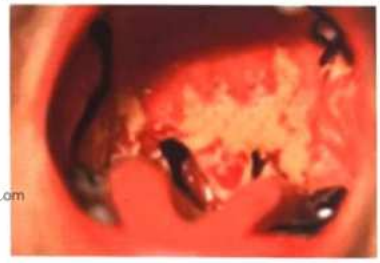
- Local colonization and invasion into deeper tissue
- Hyphal form: Phospholipase at tip and they help in local colonization and invasion
- Large size: resistant to phagocytosis
- Biofilm formation around cells: facilitates survival of organism

Diseases

- Oral thrush
- Oesophageal Candidiasis
- Vaginal Candidiasis: Thick curdy white discharge
- Candidaemia: Candida yeast cells in blood
- Candiduria: Candida yeast cells in urine
- Onychomycosis: Nail Folds are involved
- UTI/RTI
- Meningitis: In HIV +ve
- Endocarditis
- Osteomyelitis

Previous Year's Questions

Q. What could be most probable diagnosis for the image given below? (FMGE - Aug - 2020)



A. Leucoplakia
B. Oral cavity CA
C. Oral thrush by candida
D. Oral lichen planus

a. Mucocutaneous manifestations

- They include
  - Oral thrush
  - Esophageal Candidiasis
- Both are very common in HIV +ve individuals

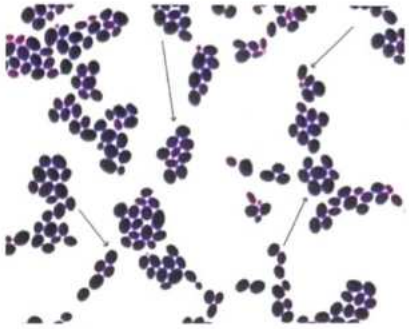
b. Cutaneous manifestations

- Intertriginous: involvement of skin folds
- Paronychia: involvement of nail folds
- Diaper dermatitis: In babies

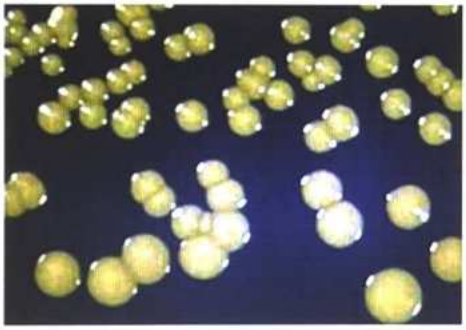
c. Systemic Candidiasis

- Gastrointestinal Candidiasis: Mostly secondary to antibiotic treatment and also in some malignancy

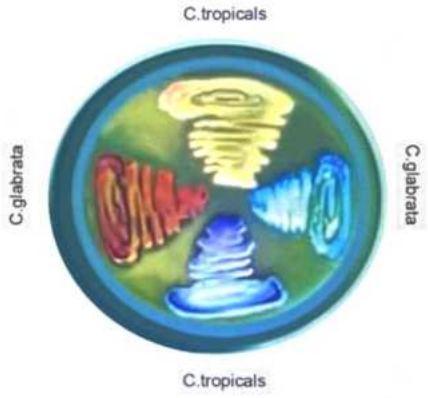
Lab diagnosis



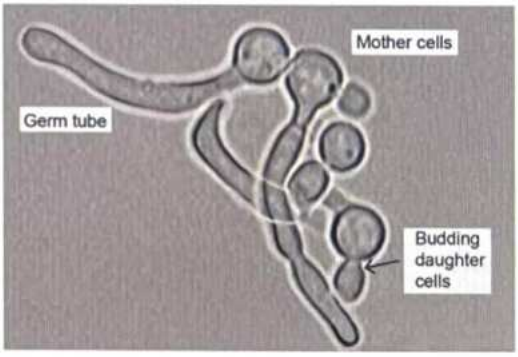
- Tissue biopsy sample + 10% KOH → can observe yeast cells under microscope
- Only fungus which can be in the form of yeast, Yeast like, can show pseudohyphae and True Hyphae → seen with C. Albicans
- Culture Growth on SDA: In 2 to 3 days they form creamy, pasty colonies



- Chrome agar – Give species of Candida

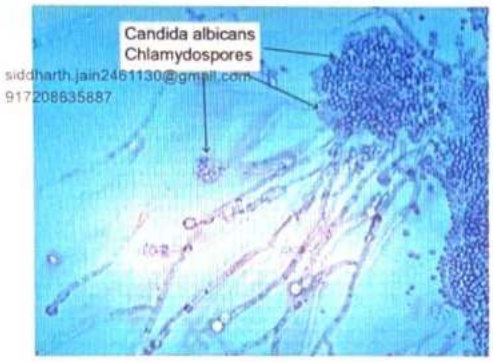


- Germ tube test (Reynold braude phenomenon)



- Culture growth + human Serum and after 30 minutes germ tube can be seen
- Mainly seen with Candida albicans

- On corn meal agar
  - It is a nutrient deficient media
  - Chlamydia spore formation seen
  - Chlamydia spores are Asexual spores and are typical for Candida albicans



Treatment and prophylaxis
- Oral and Mucocutaneous: 1% Gentian Violet
- Resistant Mucosal lesion: Nystatin
- Vaginal Candidiasis: Oral Fluconazole (single dose), suppositories and creams
- Systemic lesions: Amphotericin B
- DOC: Azoles

Miscellaneous points about Candida
- Beta 1,3 glucan test is done for invasive
  - **C** - Candidiasis
  - **A** - Aspergillosis
  - **P** – Pneumocystis carnii

How to remember
- CAP

- Galactomannan test: for invasive Aspergillosis
- Sugar fermentation
- Sugar assimilation test
- PCR for Candida species

Previous Year's Questions
Q. I-3 Beta d-glucan assay is done for which infection?
(AIIMS – Nov - 2017)

A. Invasive candidiasis
B. Cryptococcus
C. Invasive Rhinosporidiosis
D. Rhinocerebralmucormycosis

Previous Year's Questions
Q. An HIV positive patient with CD4 count of 300 / Wm presents with mucosal lesions in the mouth. On microscopy budding yeast & pseudo-hyphae are seen. What is the likely diagnosis?
(NEET – Jan - 2020)

A. Candidiasis
B. Oral hairy leucoplakia
C. Lichen planus
D. Oral cavity CA

NCA (Non-Candida albicans)
- They include
  - C. Krusei - Naturally resistant to Azoles
  - C. glabrata
  - C. stellatoidea
  - C. guillermondii
  - C. aureus - Causes worldwide infections mostly in Immunodeficient
- They are becoming more resistant to antifungal drugs

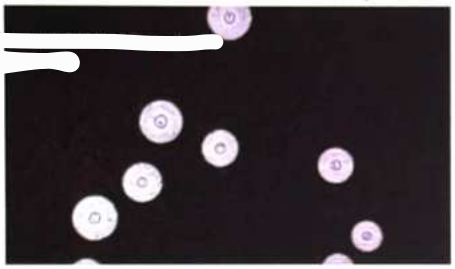
2. CRYPTOCOCCOSIS
- Caused by an encapsulated yeast Cryptococcus neoformans, pathogenic to man and animals
- 2nd most common fungal infection after Candidiasis in HIV infected individuals
- Isolated from pigeons nests, droppings, old buildings and nitrogenous soil – Creatine favours the growth
- Virulence factors
  - Capsule: inhibits phagocytosis.
  - Melanin production by the enzyme phenol oxidase. This enzyme convert L – DOPA to melanin
  - Melanin: protect the organism from intracellular killing by phagocytes

- Infection occurs by inhalation, but sometimes through skin or mucosa
- Weakness of immune system leads to reactivation and dissemination to CNS and other sites
- Visceral forms stimulate tuberculosis and cancer clinically
- Cutaneous form varies from small ulcers to granuloma
- Seen in HIV +ve patients when CD4+ count falls below 200 cells / mm^3
- Extrapulmonary Cryptococcus is one of the AIDS – defining disease
- Clinical types
  a. Pulmonary
  b. Extrapulmonary: CNS, Viscera, bone and skin are involved

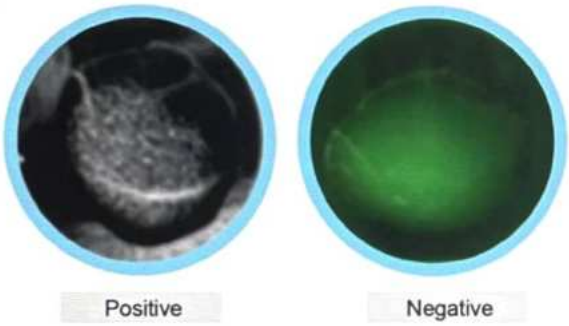
- Cryptococcal meningitis is the most serious type of infection, resembles TB and is often seen in AIDS patients

Laboratory Diagnosis

- Can do CSF culture and Blood culture
- Specimen: Serum, CSF, Body fluids
- Indian ink staining:
  - It is a negative staining
  - Done in a suspected case of HIV +ve individual
  - You can see yeast cell surrounded by thick capsule



- Fungal culture and serology
  - Grows on SDA: smooth, mucoid, creamy colonies
  - On Niger seed agar: brownish - black colonies, Phenol oxidase enzyme is responsible for formation of brownish-black colonies
  - Latex card agglutination test:



→ Done for Cryptococcal antigen
→ > 1:8 titters significant

- PCR for Cryptococcus neoformans

Treatment:

- DOC: LAMB (liposomal Amphotericin B) + Flucytosine

Previous Year's Questions

Q. HIV positive patient develop Cryptococcal meningitis which rapid test will be used?
(FMGE – Aug - 2020)

A. Indian Ink Test
B. Latex card agglutination test (Antigen based test)
C. Culture
D. PCR

3. MUCORMYCOSIS

- Invasive disease caused by lower fungi – Zygomycetes
- Found in food items, soil and air
- Zygomycetes family include
  - Rhizopus
  - Mucor
  - Absidia
- Usually occurs in diabetic patients with ketoacidosis
- High glucose and acidotic condition favours their growth
- M/c infects terminally ill patients and Covid-19 patients
- Rhinocerebral zygomycosis: it is the M/c and fulminant infection caused by theses family members
  - It involves: nasal mucosa, turbinate, bones, Paranasal sinus, orbit, palate and brain
  - There is formation of Blackish crus (because it is angio-invasive)
- Pulmonary
- Cutaneous
- Disseminated: goes to Lungs, kidney, brain, heart and GIT

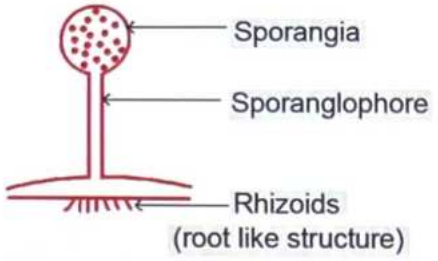
- Culture growth on SDA shows
  - Cotton wooly growth

  - Salt and pepper appearance

Diagnosis

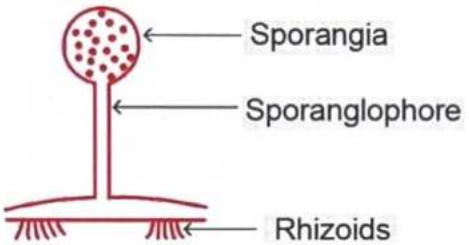
- Tissue biopsy + H and E stain shows 90° or 120° aseptate hyphae
- Culture Tissue biopsy on SDA and stain the culture with LPCB, you can observe

- Sporangia opposite to rhizoids: Rhizopus



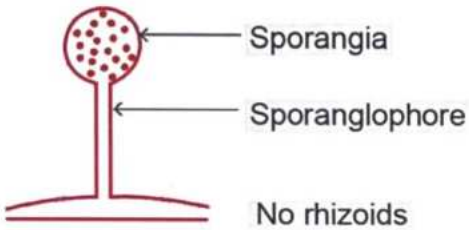
RHIZOPUS

- Sporangia at the sides: Absidia



ABSIDIA

- No rhizoids seen: Mucor



MUCOR

Treatment and prophylaxis

- Rapid correction of underlying predisposing condition like diabetic ketoacidosis
- Surgical debridement of necrotizing tissue
- Antifungals (not azoles)
- DOC: LAMB
- Adjunctive therapy with hyperbaric oxygen

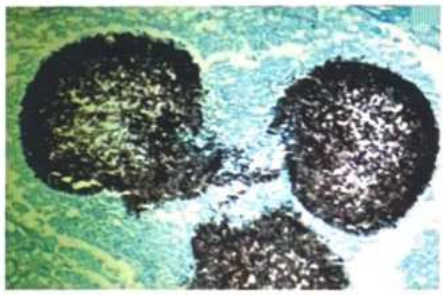
Previous Year's Questions

Q. Beta 3 glucan assay testing done for?
(AIIMS - Nov - 2019)

A. Invasive candidiasis
B. Aspergillosis
C. Pneumocystis Carinii
D. Mucormycosis

4. ASPERGILLOSIS

- It can cause systemic infections in immuno-compromised and immuno-competent individuals
- Infection occurs by
  - Inhalation of conidia
  - Direct entry through wounds during surgery
- Imp species:
  - A. fumigatus
  - A. Flavus
  - A. niger
  - A. clavatus
- Pulmonary Aspergillosis: Subdivided into 3 categories depending upon whether the host is atopic or immunocompromised
  - Allergic Aspergillosis
    - → Aspergillus asthma
      - Atopic individuals
      - Inhalation of spores
    - → Allergic bronchopulmonary Aspergillosis (ABPA)
      - Repeated and heavy exposure to spores
      - Breathlessness, fever and malaise
    - → Obstructive Aspergillosis
      - Plugs of entangled Mycelia and mucus block segments of lung tissue and even entire lobe
      - Productive cough: contains Aspergillus hyphae
    - → Aspergilloma
      - Non invasive
      - Colonization in a pre-existing cavity (tubercular)
      - Compact mass fungal mycelia surrounded by dense fibrous walls: Fungus Ball
      - Usually solitary 8-10 cm



  - Invasive Aspergillosis
    - → Important cause of morbidity and mortality
    - → May disseminate to kidney and brain
    - → DOC: Voriconazole
- Extrapulmonary Aspergillosis: through blood it can go to CNS, Paranasal sinus, Skin, Endocardium (in prior cardiac surgery)
- Miscellaneous forms
  - Occulomycosis: infection of eye
  - Otomycosis: infection of ear
  - Onychomycosis: infection of Nails

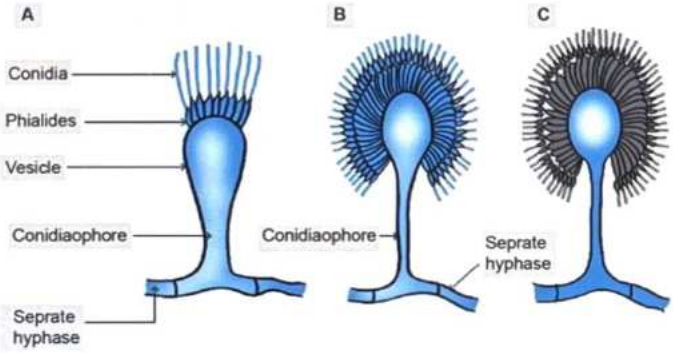
- A. fumigatus mainly associates with ABPA and Aspergilloma
- A. Flavus leads to Aflatoxin secretion which is associated with HCC
- A. Niger causes Blackish crust formation
- Common lab contaminant, hence repeated isolation from specimen is mandatory
- Specimen: sputum, BAL, Tracheobronchial biopsy

Diagnosis

- Initially direct examination by KOH mounting
- Tissue Biopsy + H and E stain or KOH shows Acute angle septate hyphae dichotomous branching → Aspergillus
- Tissue biopsy and culture on SDA or Czapek Dox agar produce colonies.

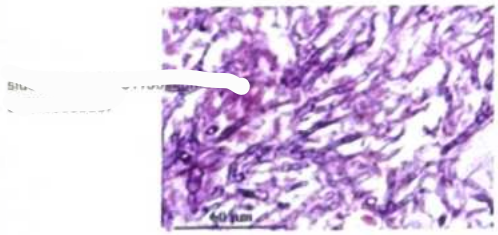
A.niger A.fumigatus A.flavus

- These colonies on LPCB mount shows the following appearances



Previous Year's Questions

Q. Fungal Hyphae with acute angle branching & septate hyphae. Identify? (FMGE - Dec - 2019)



A. Aspergillosis
B. Mucor
C. Histoplasma
D. Sporothrix

Previous Year's Questions

Q. Following figure showing slide of organism stained with Gommorimethenamine silver. What features & which organism is it? (AIIMS - Nov - 2017)

A. Fungi is showing acute angle branching with septate filament & is aspergillus
B. Fungi is showing non septate acute angle branching &is candida
C. Fungi is showing rightacute angle branching with septa and is mucor
D. Fungi is showing non septate acute angle branching & is aspergillus

5. PENICILLIOSIS

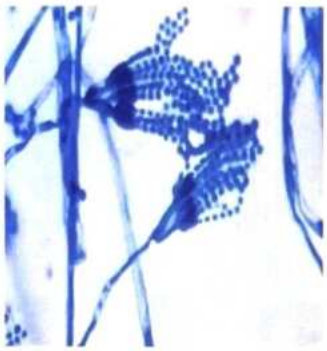
- Caused by penicillium marneffi in HIV +ve
- Dimorphic, facultative intracellular fungi
- Route of transmission:
  - Inhalation of conidia
  - Ingestion (eating rats, china)
  - Direct inoculation of skin

- Bamboo rat harbours P. marneffei in their internal organs
- Isolated from feces and soil sample from burrows
- RES is the 1° site of infection
- 3 histological patterns of disease
  - Granulomatous: granuloma
  - Suppurative: Multiple abscess in lung, skin and subcutaneous tissue of immunocompromised individuals
  - Necrotizing: immunocompromised

Lab diagnosis

- Wright's, Giemsa stain of skin, biopsies shows – septate yeast like cells
- Isolated from blood, skin, BM, sputum, LN, pleural fluid, urine and BAL
- On SDA and BA at 25°C shows Wooly pigmented colonies, reverse is bright rose colour

- Microscopy
  - Short, septate hyphae with branching
  - Brush borderd conidiophores bearing conidia



6. PNEUMOCYSTIC CARNI / PNEUMOCYSTIS JIROVECII

- Protozoa: Cyst/Trophozoite stage present which favours it to be Protozoa
- Fungus: Chromosomal studies favours it to be Fungus

Pneumocystis carnii pneumonia

- Occurs in HIV + ve individuals
- Occurs in persons with CD_4 counts < 200
- DOC: Cotrimoxazole
- Lab Diagnosis
  - BAL [Broncho Alveolar Lavage] Fluid
    → OT procedure
    → Bronchoscopy is done for BAL
  - Gastric washings: Taken in children as they tend to swallow their sputum
    → BAL / Gastric Washings + GMS staining [Gomorii Methanamine Silver] shows
    - Cysts which are Black (d/t Glycogen content in cysts)
    - Trophozoites which stains Green (GMS is naturally green colored)

ERGOTOXICOSIS

- It is termed as Ergot poisoning
- Ergot poisoning occurs because Claviceps purpurea seeds contaminate edibles
- Ergotoxicosis manifests in the form of
  - St. vitus dance
  - St. Anthony's fire

OCCUPATIONAL FUNGAL DISEASE

1. FARMER'S LUNG

- D/t Hay dust (stored Hay)
- Caused by Micro monospora faenii, Thermo Actinomyces vulgaris

2. BAGGASSOSIS

- D/t Sugar Cane Dust
- Caused By Thermo Actinomyces Sacchari

3. MALTSTER'S LUNG

- Seen in people working in Barley & Malt factories
- Caused by Aspergillus clavatus

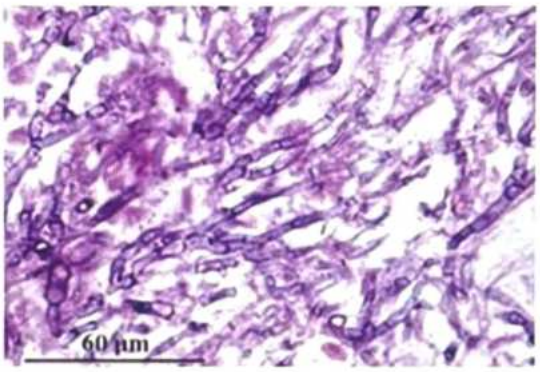
CLINICAL QUESTIONS

Q.1 A patient with asthma shows growth of molds upon processing his sample. LCB mount shows septate hyphae with acute angle branching. Blood tests shows eosinophilia. Which of the following condition cannot be associated in this case?

A. Bronchopulmonary allergy
B. Otomycosis
C. Dermatophytosis
D. Keratitis

Solution:

- **Dermatophytosis cannot** be associated in this case.
- The clinical scenario given in the question is suggestive of Allergic **Broncho Pulmonary Aspergillosis caused by Aspergillus.**
- **Points in favor of Aspergillus (Image below):**
  - Septate hyphae
  - dichotomous branching
  - acute angled (mostly 45°C)



Clinical manifestations of Aspergillus:

- **A. fumigatus → causes:**
  - ABPA (Allergic **Broncho Pulmonary Aspergillosis)**
  - Aspergilloma (Fungal ball)
  - Oculomycosis (**Keratitis**, endophthalmitis)
  - **Otomycosis**
- **A. niger** → causes Otomycosis. **Blackish** crust is seen.
- **A. clavatus** → causes Maltster's Lung, common in Barley/malt factor workers.
- **A. flavus** → produces Aflatoxin associated with HCC (Hepatocellular carcinoma).
- **Dermatophytosis (or tinea or ringworm)** is the commonest superficial mycoses infecting keratinized tissues caused by

- Trichophyton species: Infect skin, hair, and nail
- Microsporum species: Infect skin and hair
- Epidermophyton species: Infect skin and nail

Q.2 A 25 years old female complains of recurrent rhinitis, nasal discharge, and b/l nasal blockage for one year. She has a history of asthma and allergy. On examination, multiple ethmoidal polyps are noted with mucosal thickening and impacted secretions in both the nasal cavities. A biopsy is taken and the material is cultured which shows the growth of many hyphae with dichotomous branching typically at 45 degrees. Which of the following is the most likely responsible organism?

A. Aspergillus fumigatus
B. Rhizopus
C. Mucor
D. Candida

Solution:

- The most likely responsible organism is **Aspergillus fumigatus.**

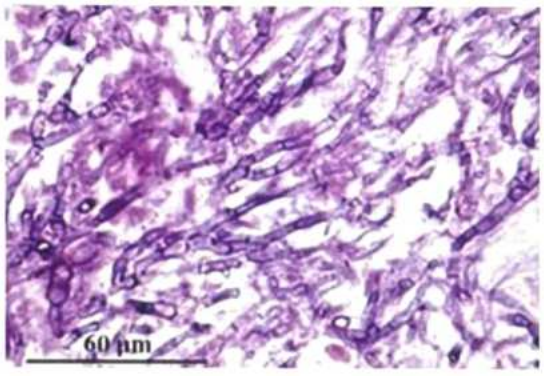
1. Key points towards diagnosis:

- Recurrent rhinitis, nasal discharge, bilateral nasal blockade.
- History of asthma and allergy
- Hyphae with dichotomous branching at **45 degree**

2. All points go towards Aspergillus infection.

- Most common type of fungal infection of **nose and paranasal sinuses** are due to Aspergillus.
- Aspergillus has **septate hyphae that branch typically at 45 degrees (V-shaped).**

3. Hyphae with dichotomous branching at 45 degrees:



Answers to other options:

- Broad **aseptate** hyaline hyphae with **wide-angle branching** seen in members of Zygomycetes. i.e Rhizopus and Mucor
- Candida: Gram-positive oval budding yeast cell with **pseudohyphae.**

LEARNING OBJECTIVES

UNIT 4: PARASITOLOGY

- Protozoology
  - Classification of protozoa
  - Amoeba: Naegleria fowleri, Acanthamoeba, Balamuthia, Entamoeba histolytica and Entamoeba coli
  - Ciliates: Balantidium coli
  - Flagellates: Giardia lamblia, T. vaginalis, Leishmania and Trypanosoma
  - Sporozoa
- Helminthology
  - Cestodes
  - Trematodes
  - Nematodes: Intestinal Nematodes and Tissue Nematodes

9 PARASITOLOGY PART-1 (PROTOZOOLOGY)

CLASSIFICATION OF PROTOZOA

00:02:43

1. **Amoebae**
- Free living
  - Naegleria Fowleri
  - Acanthmoeba
  - Balamuthia
- Intestinal
  - Entamoeba histolytica
  - Entamoeba coli
2. **Ciliates**
- Eg. Balantidium coli
  - Largest intestinal protozoa
  - It is having 2 types of nucleus (micro and macro)
3. **Flagellates**
- Intestinal
  - Eg. Giardia lamblia
- Vaginal
  - Eg. Trichomonas vaginalis
- Blood & Tissue
  - Eg. Leishmania and Trypanosoma
4. **Sporozoa**
- Blood species
  - Plasmodium
  - Babesia microti
- Tissue species
  - Toxoplasma Gondii
- Intestinal species
  - Cryptosporidium parvum
  - Isospora
  - Cyclospora

Clinical scenario I

Q. A 52 year old patient with a history of recent travel to India presented to interventional radiology from an outside hospital for aspiration of a liver abscess, and was subsequently returned to the outside hospital. The patient has spent 2 months in India before returning to US, and about 1 month later developed right upper quadrant pain. Abdominal CT showed 2 cystic masses measuring 2 to 4cm. Aspiration of cysts yielded 0.5ml of bloody fluid, which was sent for bacterial culture and smear

Diagnosis: Amoebic Liver abscess

- The above gives case is typical of Amoebic liver abscess

AMOEBA

ENTAMOEBA HISTOLYTICA

00:17:10

- Infective Form: Quadrinucleate Cyst
  - Transmission occurs through feco oral
  - It resists gastric acidity and cause Excystation in small intestine or large intestines
  - Excystation: It means, from the cysts Trophozoites (active feeding & growing stage) are being produced
  - Trophozoite can act in two ways according to conditions
    - → Unfavorable condition: If the person's immune system is very strong, immediately they try to Encyst and uninucleate cyst followed by Binucleate cyst and then Quadrinucleate cyst are produced
    - → Favourable conditions: 85% remains asymptomatic but 5-15% can invade intestinal layers due to certain virulence factors like Cysteine proteinase and lectin (help in adherence) and cause a Flask shaped ulcer
      - Through these flask shaped ulcers they get access to the portal circulation and reach liver
      - Liver is the M/c site of EIA (Extra Intenstinal Amoebiasis)

EIA (Extra Intenstinal Amoebiasis)
- M/c site For EIA: Liver > Lung > Brain
- Chocolate pus/ Anchovy sauce pus formed due to damage to hepatocytes
- Right lobe, posterio superior portion is involved
- IOC For EIA: ELISA

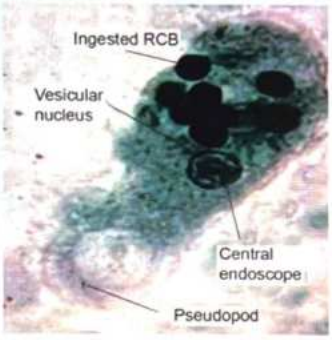
Intestinal Amoebiasis
- Its having features of Amoebic dysentery

Lab diagnosis
- Saline mount: for Trophozoites
  - Stool sample + Normal saline → Trophozoites seen under microscope which have
    - → Eccentric nucleus with central Karyosome is present

→ Erythrophagocytosis seen which is Engulfed RBC's (confirmatory feature)

- Iodine mount: for cysts
  - Stool sample + Iodine mount → cyst is seen under microscope which are
    → Uninucleate or Binucleate cysts with chromatoidal bars & Glycogen vacuoles
    → In Quadrinucleate cyst: only 4 nuclei seen

- Culture: Media used are
  - P - Philips Media
  - C - Craige's Media
  - R - Robinson Media
  - Balamuth Media
- PCR for E. histolytica

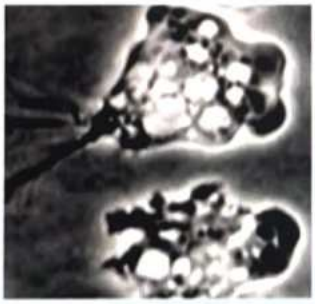


- Trichrome stain of Entamoeba histolytica Trophozoites in amoebiasis

- Mature cyst of Entamoeba coli in an unstained wet mount preparation with saline

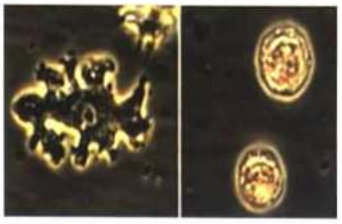
- Histopathology of a typical flask shaped ulcer of intestinal amoebiasis

- Charcot-Leyden crystals-fecal smear (Trichrome stain)



- Acanthamoeba: Trophozoite with thorn like pseudopodia in CSF saline mount

- Naegleria fowleri: Amoeboid trophozoite and flagellated trophozoite in saline mount



- Balamuthia mandrillaris: Trophozoite and cyst in saline mount

Previous Year's Questions

Q. Patient presented with colicky pain &diarrhoea. No history of fever & Stool sample is given for investigation & showed in the image diagnosis?

(FMGE - Dec - 2020)

A. E. Coli
B. E. Histolytica
C. Giardia Lamblia
D. Shigella

FLAGELLATES

00:35:23

1. INTESTINAL FLAGELLATES

Giardia Lamblia

- Associated with traveler's diarrhea
- Motility: falling leaf like motility
- Infective form: Binucleate cyst > Quadrinucleate cyst
- Binucleate cyst gets ingested with contaminated food & water →Bypass gastric acidity → Excystation in duodenum →Trophozoites attach to intestinal layers and damage intestinal villi which will lead to
  - Malabsorption: manifest as watery diarrhea or Bulky greasy stools (Steatorrhea)

- Lab diagnosis
  - Saline mount: For Trophozoite
    → Trophozoite is pear shaped / monkey face / Tear drop appearance

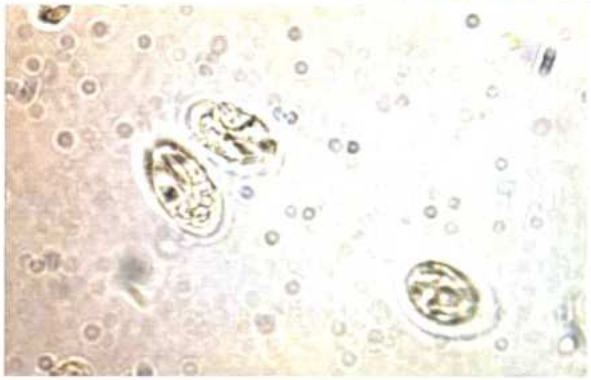
    - → It has 4 pairs of flagella
    - → Having Axostyle and ventral sucking disk
  - Iodine mount: For cyst
    - → Cysts have 2 nuclei > 4 nuclei
    - → Having Axostyle
  - String test / Enterotest
  - ELISA
  - PCR for Giardia lamblia

- Immunodeficiency associated with Giardiasis
  - Selective IgA deficiency
  - CVID (common variable immunodeficiency)
- Treatment
  - DOC: Metronidazole

Previous Year's Questions

Q. Cyst are parasite seen in stool microscopy. What is the organism?

(AIIMS - Nov - 2017)



A. Entamoeba Dispar
B. Balantidium Coli
C. Giardia Lamblia
D. Taenia Solium

- Giardia lamblia Trophozoite

- Giardia lamblia cyst

Case scenario 2

A 19- year-old woman presents to the physician's office for the evaluation of an itchy vaginal discharge that she has had for about a week. She has had no fever, abdominal pain, or dysuria. She became involved with a new sexual partner approximately 3 weeks ago.

She takes birth control pills but does not regularly use condoms during intercourse. Her partner is asymptomatic. On examination, her vital signs are normal, and a general physical examination is unremarkable. On pelvic examination, her external genitalia are normal.

After inserting a speculum, you see a bubbly, thin, yellow vaginal discharge. Her cervix is Erythematous but without discharge. She has no cervical motion or uterine or adnexal tenderness.

A wet mount of the vaginal discharge examined microscopically reveals numerous motile, flagellated, pear-shaped organisms along with numerous white blood cells.

Q. What is the most likely infectious cause of her vaiginal discharge?
A. Trichomonas Vaginalis

Q. What is the most likely source of her infection?
A. Sexual contact with infected but asymptomatic partner

2. VAGINAL FLAGELLATES

00:53:01

Trichomonas Vaginalis

- Does not have any cystic stage
- Only having pear shaped and Flagellated (2 - 10) Trophozoite stage
- Clinical manifestation:
  - PH > 4.5
  - Yellowish, Greenish, or Greyish white discharge
  - Strawberry cervix / Colipitis macularis / strawberry vagina
- Lab diagnosis:
  - Urine wet mount: Motile Trophozoite seen
  - PAP smear
  - Culture on:
    - → Bushley Johnson and Trussel media
    - → CPLM
  - NAAT for Trichomonas vaginalis

- Treatment:
  - DOC: Metronidazole
  - Sexual partner should also be treated with Metronidazole to prevent recurrences

Previous Year's Questions

Q.Identify image below?

(FMGE - Aug - 2020)

A. Trichomonas
B. Plasmodium falciparum
C. Entamoeba histolytica
D. Giardia lamblia

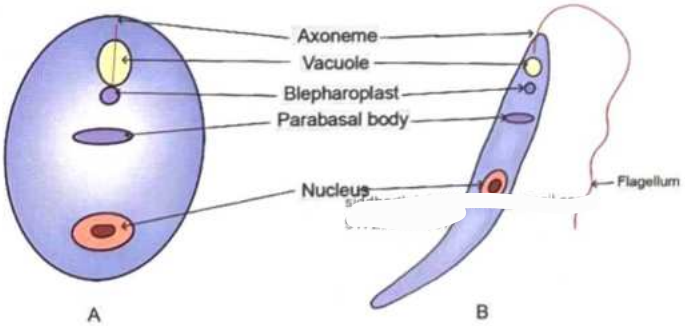
3. HAEMOFLAGELLATES

01:00:27

a. Leishmania: infective form is Promastigote
b. Trypanosoma: infective form is Metacyclic Trypomastigot

a. Leishmania

- L. tropica: leads to oriental sore or Delhi boil or Baghdad boil or Aleppo button
- L. brasilensis: causes Espundi [Mucocutaneous leishmaniasis]
- L. Donovani: causes Kala Azar
  - Infective form: present in Sand fly
    - → Promastigote: Flagellar form
    - → Amastigote: Non Flagellar form

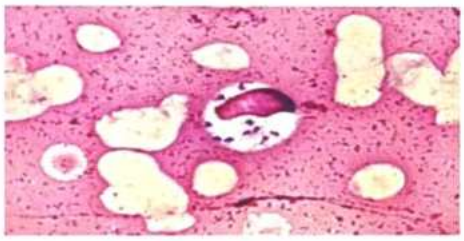


Refer Flow Chart 9.1

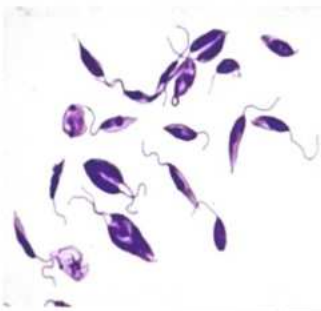
Kala azar

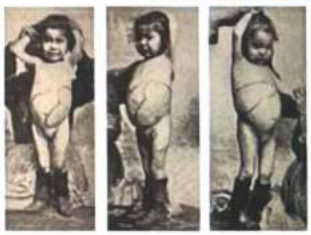
01:09:40

- Hyper pigmentation + Fever + Hepatosplenomegaly
- Kala Azar along with HIV +ve: Hepatosplenomegaly absent (atypical presentation)
- Other clinical features
  - Pancytopenia: Anemia + Thrombocytopenia + Leucopenia
  - Hypergamma globulinemia
- Lab Diagnosis
  - Blood Smear: shows Anemia, Leucopenia, Thrombocytopenia and Hypergamma globulinemia
    - → Hypergamma globulinemia demonstrated by
      - Napier's Aldehyde Test
      - Chopra's Antimony Test
  - Bone marrow or splenic biopsy
    - → Shows Amastigot form aka LD bodies (Leishman Donovan bodies)



  - Culture of Promastigote form on: NNN media (Novy-MacNeal - Nicolle medium)
  - RK39 antigen card test: Rapid card test
  - ELISA
  - Compliment fixation test (obsolete)
  - PCR for L. donovani
- Treatment
  - DOC: LAMB (Liposomal Amphotericin B)





PKDL [Post Kala Azar Dermal Leishmaniasis]

- Viscerotropic organism now become dermotrophic
- After 2 yes of successful treatment, an individual suddenly develops Hypopigmented nodular Lesions d/t dermotrophic conversion
- DOC: Oral Miltefosine

b. Trypanosoma

Sleeping sickness

- Caused by Trypanosoma brucei
- Vector: Glossina / Tse-Tse fly
- Trypanosoma brucei is of two types
  - Gambiense: Causes West African sleeping sickness
    - → West African sleeping sickness has winter bottom sign
    - → Winter bottom sign: prominent B/L cervical LAP
    - → Chronic CNS
  - Rhodesiense: Causes East African sleeping sickness
    - → Winter bottom sign absent
    - → Acute CNS
    - → More dangerous, associated with high mortality
- Treatment:
  - For Acute stage: Suramin
  - For chronic stage: Melarsoprol

How to remember

WWG Tomorrow
W – Western sleeping sickness
W – Winter bottom sign
G – Gambiense
Tomorrow – Tse-Tse fly. Trypanosoma

Chagas disease 01:31:42

- Aka American Trypanosomiasis
- Caused by: T. cruzi
- Infective form: Metacyclic trypomastigote
- Vector: Reduvid Bug
- Entry site edema: Chagoma

- Features:
  - Romana sign: eyelid swelling
  - Mega esophagus
  - Mega colon
  - Myocarditis
  - Meningo encephalitis

- DOC: Benznidazole

Refer Flow Chart 9.2

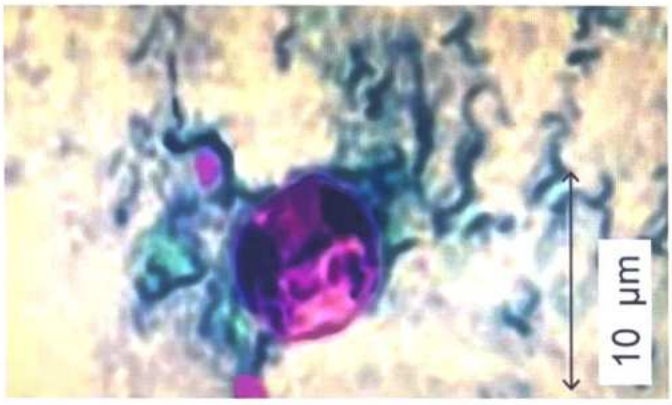
SPOROZOA

COCCODIAN PARASITES 01:43:12

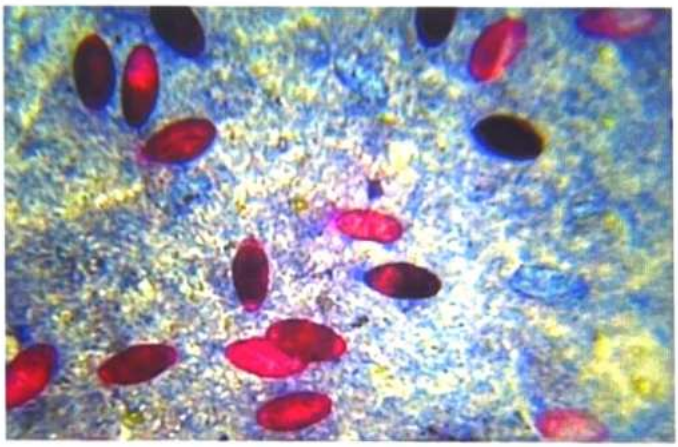
- Can cause diarrhea in HIV +ve (C. parvum is M/c)
- Includes

	Cryptosporidium parvum	Cyclospora	Isospora belli
• Oocyst size	• 4-6 µ	• 8-10 µ	• 23-36 µ
• Acid fastness	• Acid fast (0.2-1%)	• Acid fast (0.2-1%)	• Acid fast (0.2-1%)
• Autofluoresence	• Absent	• Present (++)	• (+ /-)
• Sporulation in	• Enterocytes	• Soil	• Soil
• Sporozoites	• 4	• 4	• 8
• Treatment	• IV fluids + Nitazoxonide	• IV fluids + Cotrimoxazole	• IV fluids + Cotrimoxazole

- **Cyclospora cayentenensis: 8-12 µm in size, round, variably acid fast (1%H_2SO_4), contains two sporocysts each bearing sporozoites.**



- **Isospora belli:23-36 µm in size, oval, uniformly acid fast (1% H_2SO_4), contains two sporocysts each bearing four sporozoites**



Previous Year's Questions

Q Post renal transplant patient on Immunosuppressants complains of chronic diarrhoea. Stool examination reveals cyst of 4-6 micron in size. Positive on Kinyoun staining which of the following statements is true regarding the causative agent of this clinical condition?

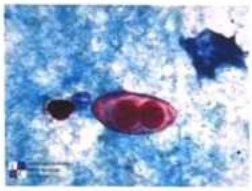
(AIIMS - Jun - 2020)

A. MC cause is Cryptosporidium
B. Oocyst becomes ineffective immediately after coming out in stools
C. These are obligate intracellular organisms
D. Autoinfection is seen with them

Previous Year's Questions

Q. A known HIV patient on anti-retroviral therapy presented with diarrhoea of six months duration. Stool microscopy was done in which 10-30 micrometre cyst were seen. Kinyoun Stain was positive. What is most likely. Diagnosis?

(AIIMS - May - 2018)



A. Cystoisospora
B. Cryptosporidium
C. Balantidium Coli
D. Strongyloides

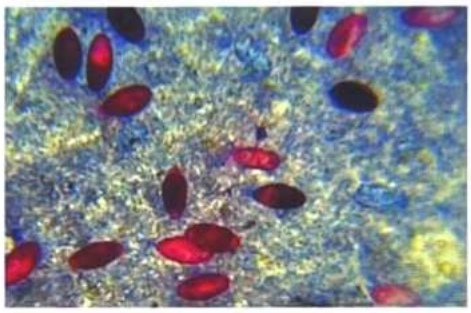
TOXOPLASMA GONDII

- Leads to Toxoplasmosis
- Define Host: Cat
  - Host in which the sexual cycle of organism happens
  - Cat → Enteric cyclic occurs (oocyst formed)

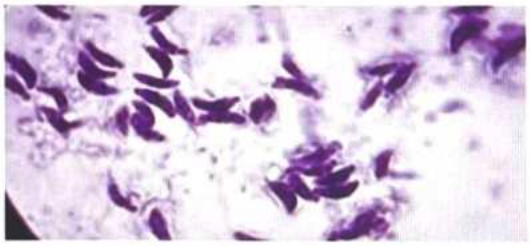
- Intermediate Host: Rat / man / Pig
  - Rat/Man → Exoenteric cyclic occurs (No oocyst formation)
- Infective forms
  - Bradyzoites:
    → Present in Tissue Cyst
    → Slowly dividing forms
    → Spreads by eating infected meat
  - Tachyzoites:
    → Present in blood
    → Rapidly dividing forms
    → Spreads by blood transfusion
  - Sporulated oocyst
    → Present in soil
    → Spreads by contaminated food and water

- Manifestations:
  - Toxoplasmosis in Normal individual: mild cervical lymphadenopathy
  - Toxoplasmosis in immunodeficiency (HIV +ve): Seizures
  - Toxoplasmosis in pregnancy:
    → At 1st Trimester: max damage to fetus
    → At 3rd Trimester: max chance of transmission
    → DOC: spiramycin

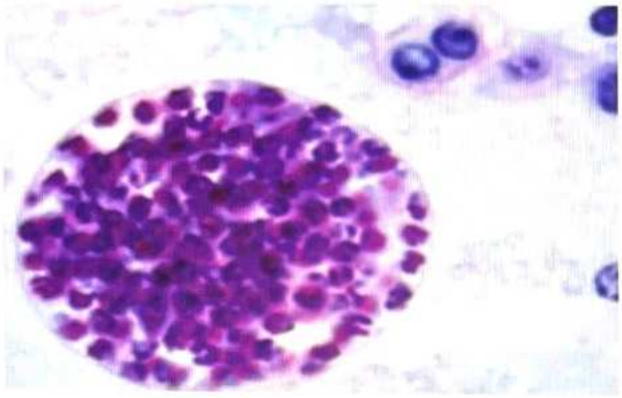
- Lab Diagnosis
  - Blood Smear + Giemsa stain: Shows Tachyzoites
    → Rapidly dividing forms
    → Sickle shaped
  - Tissue Smear + Giemsa stain: Shows Bradyzoites / Tissue cysts
    → Slowly dividing forms
  - Compliment Fixation Test: Sabin-Feldman Dye Test
  - IgA ELISA
  - PCR for Toxoplasma Gondii



- **Isospora belli: 23-36 µm in size, oval, uniformly acid fast (1% H_2SO_4), contains two sporocysts each bearing four sporozoites**



- **Toxoplasma gondii: Tachyzoites (comma-shaped) from the peritoneal exudates of infected mice stained with Giemsa**



- **Toxoplasma gondii: Tissue cyst (containing bradyzoites) in organ biopsy**

PLASMODIUM

02:14:41

- Species:
  - P. vivax
  - P. falciparum
  - P. ovale
  - P. malaria
  - P. knowlesii
- Definitive host: FAM
- Intermediate host: MAN
- Infective form: Sporozoites

Refer Flow Chart 9.3

- Conditioning Preventing P. Falciparum Infection
  - **G** - **G**6P Deficiency
  - **S** - **S**ickle cell anemia
  - **T** - **T**halassemic trait
  - Excess of Fetal Hb

How to remember

GST

- Condition Preventing P. Vivax infection: Duffy negative RBC

- P. falciparum:
  - Most dangerous
  - Cause Cerebral malaria
  - Durck's granuloma: histopathological finding in the brain capillaries of a patient died of P. falciparum
  - Formation of Ague spleen
  - Poor prognostic features: hypoglycemia and Lactic acidosis
  - Massive hemoglobinurea leads to Black water fever

Previous Year's Questions

Q. Malaria relapse common with which type plasmodium species?

(AIIMS - Nov - 2017)

A. Plasmodium malariae &vivax
B. Plasmodium falciparum&vivax
C. Plasmodium vivax &ovale
D. Plasmodium ovale &malariae

URINE

NORMAL PATIENT DILUTED

Lab diagnosis

- Blood smear Examination
  - Thick smear: to know prevalence of parasite
  - Thin smear: to know speciation
- Buffy coat examination
  - Technique used Cava motto technique
  - Fluorescent dye used: Acridine orange
  - Acridine orange stains parasite Green
- Immuno Chromatographic Test: card test
- HRP2 (Histidine Rich protein) based kits: for P. falciparum
- LDH and Aldolase kits: for other Plasmodium species
- Culture on RPMI-1640
- PCR for plasmodium species

P. vivax	P. Falciparum
• Young RBCs affected	• All age RBCs affected
• RBC Size increases	• Normal size RBCs
• Schuffner's Dots Seen	• Maurer's Dots Seen
• Single large ring within RBC	• Multiple rings are seen within RBC

• Schizont stage present	• Schizont stage is present in tissues but its absent from the blood smear
• Gametocyte no proper shape	• Banana shaped
• Accole forms absent	• Accole forms present
• Relapse present	• Relapse absent
• Recrudescence absent	• Recrudescence present
• Pigment: yellowish brown	• Dark brown pigment

- Important features of P. falciparum
  - A – Accole forms
  - M – Maurer's dot
  - B – Banana shaped gametocyte
  - A – All age RBCs affected
  - R – Renal failure, Recrudescence

How to remember

AMBAR

Previous Year's Questions

Q. A female from a tribal area of Jharkhand reports with fever for last 3 days peripheral blood is collected & stained with Giemsa. A diagnosis of malaria is made. The smear is shown in the figure. What is the likely cause?

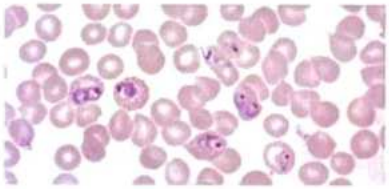
(NEET – Jan - 2020)

A. P. Falciparum
B. P. Vivax
C. P. Malariae
D. P. Ovale

Previous Year's Questions

Q. Identify organism based on peripheral smear finding given below?

(FMGE - Dec - 2020)

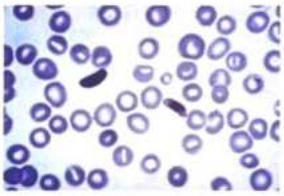


A. Plasmodium vivax
B. Plasmodium falciparum
C. Babesia
D. Plasmodium ovale

Previous Year's Questions

Q. Banana shaped gametocytes are typically formed by:

(FMGE - Aug - 2020)



A. Plasmodium vivax
B. Plasmodium ovale
C. Plasmodium malariae
D. Plasmodium falciparum

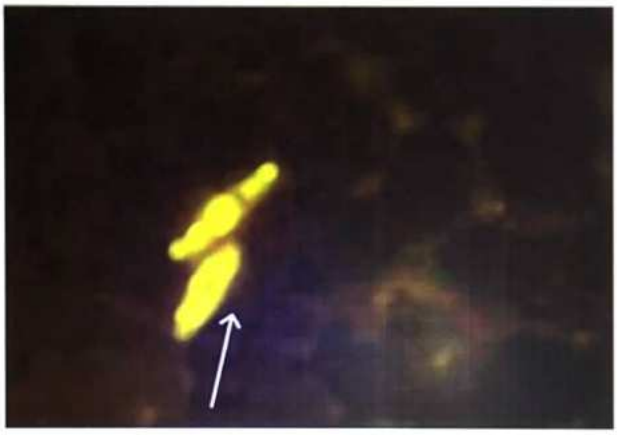
Plasmodium malaria 02:41:23

- Important features of P. malaria
  - Z – Zeimann's dot
  - O – Old age RBCs
  - O
  - M – Max incubation period
  - I
  - N – Nephrotic syndrome
  - BAND form

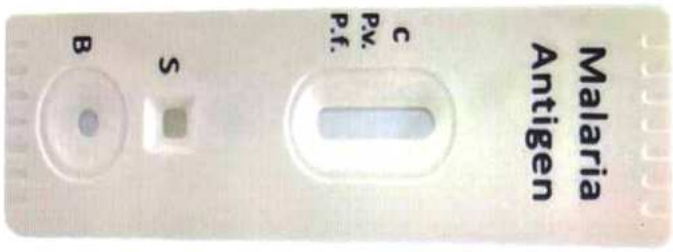
How to remember

ZOOM IN

Refer Flow Chart 9.4



QBC gametocytes of plasmodium falciparum



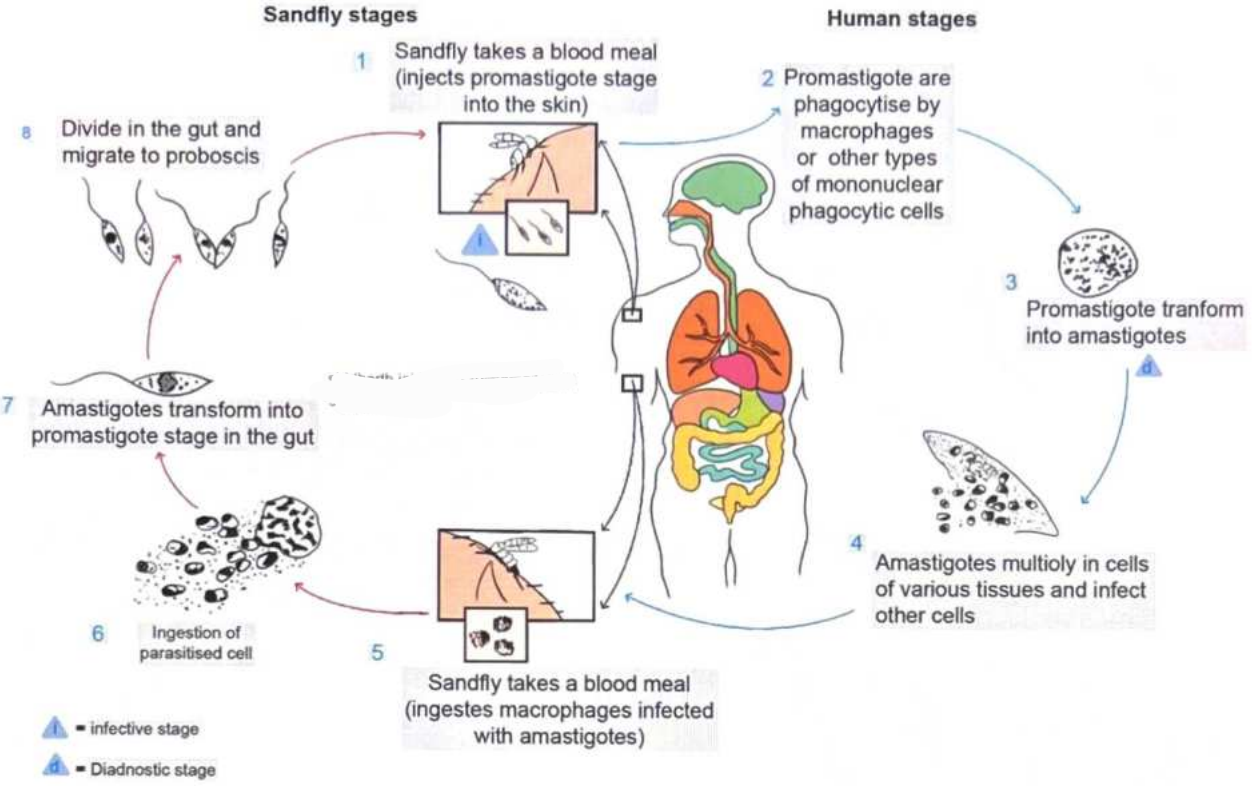
BABESIA MICROTI 02:51:23

- Leads to Babesiosis
- Tick borne illness
- Maltase cross seen in RBCs in case of Babesiosis infection
- Maltase cross of Babesia Microti mimics P. falciparum gametocyte stage
- Treatment: Clindamycin

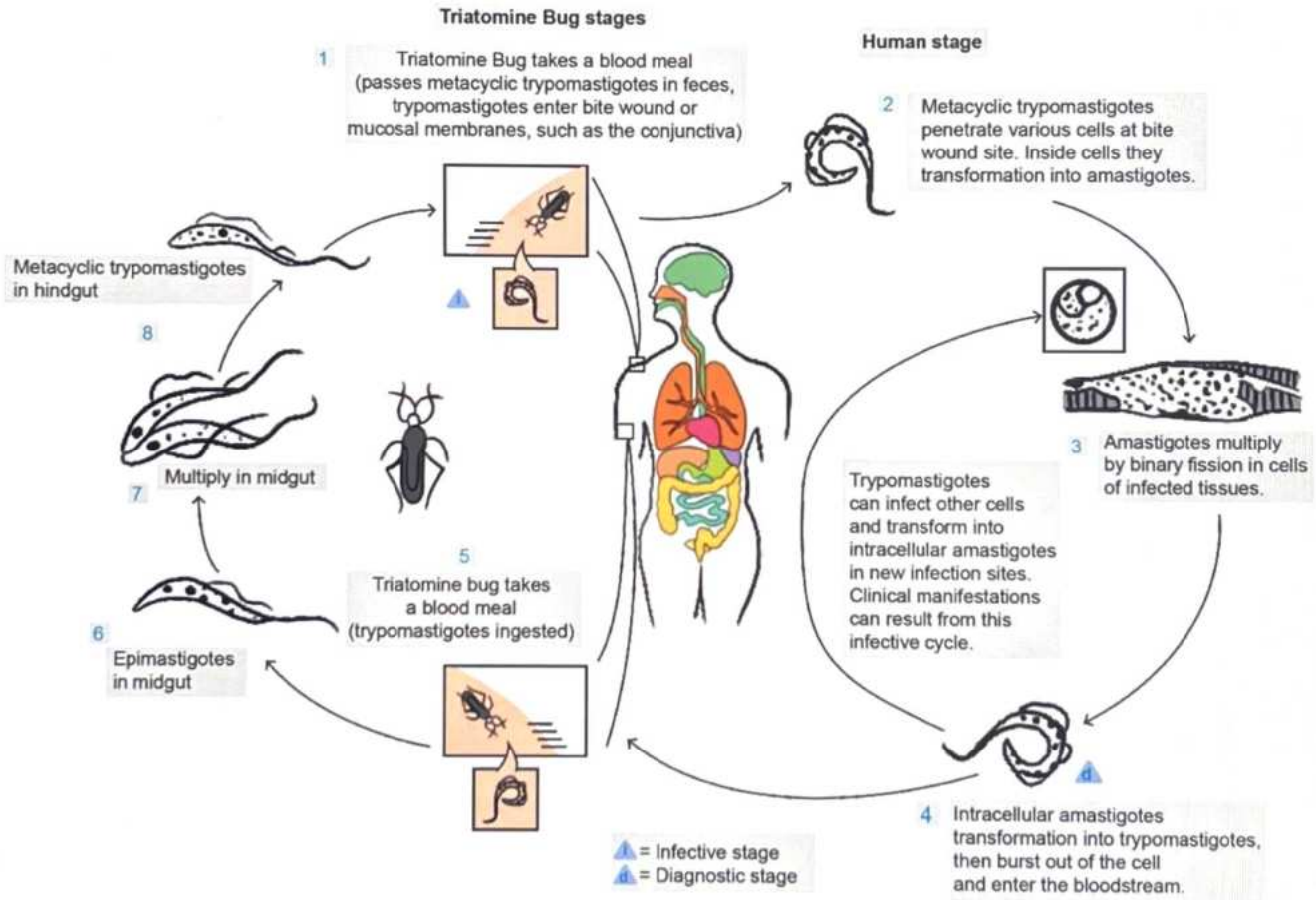
BALENTIDIUM COLI 02:53:58

- It is a ciliate
- Largest intestinal protozoa
- Have 2 type of nucleus: micro and macro
- Rotatory motility is a feature
- Diarrhea is a feature
- Transmission: Pig to man
- DOC: Doxycycline

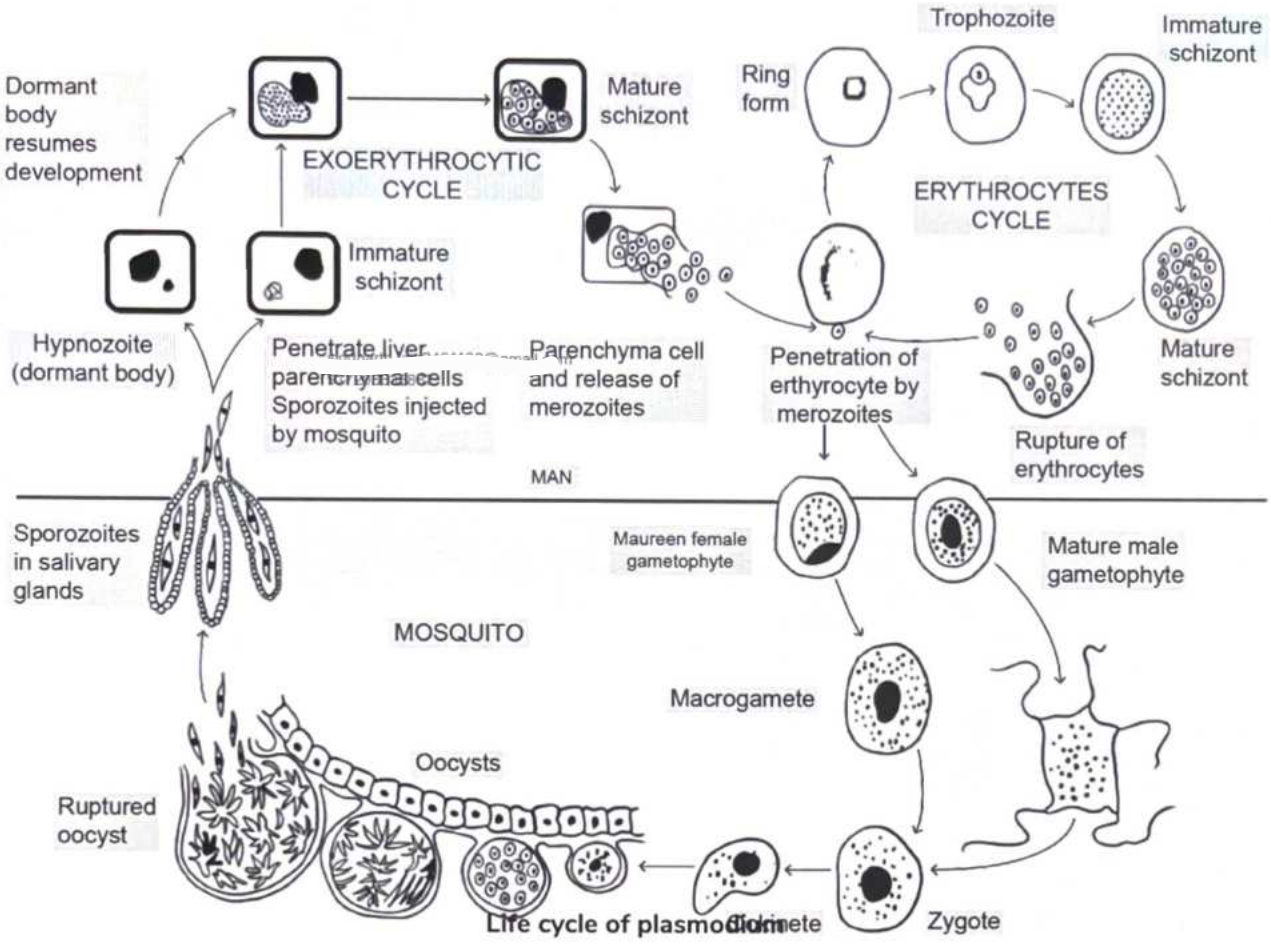
Flow Chart 9.1



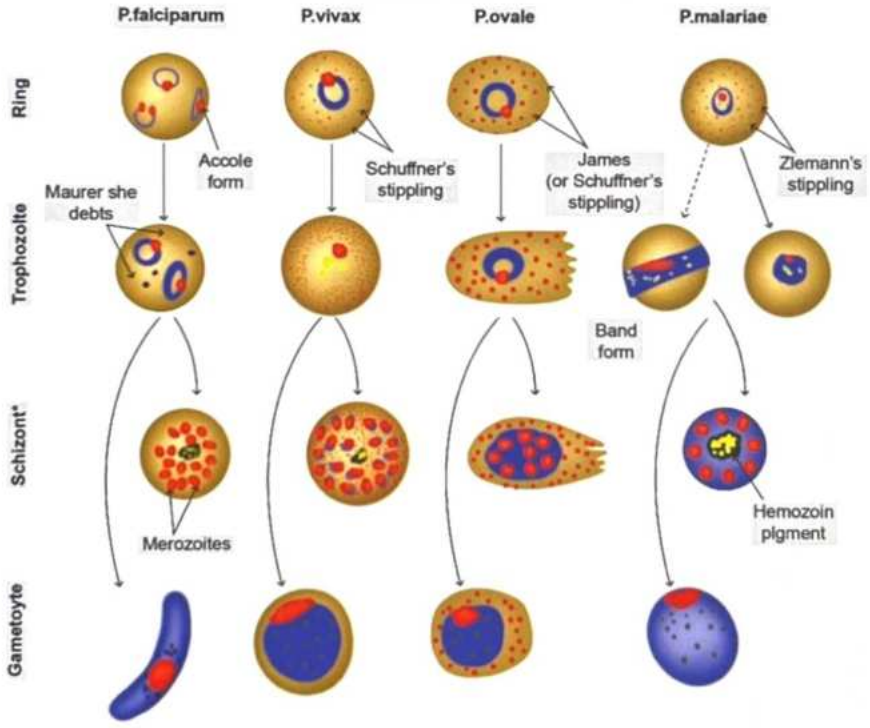
Flow Chart 9.2



Flow Chart 9.3



Flow Chart 9.4



CLINICAL QUESTIONS

Q.1 A 30-year-old patient presented with features of acute meningoencephalitis in the casualty. He had recently gone for diving during his vacations. His CSF on wet mount microscopy revealed a motile unicellular microorganism. What is the most likely organism responsible for this condition?

A. Naegleria fowleri
B. Acanthamoeba castellani
C. E. histolytica
D. Ent. Coli

Solution:

- The most likely organism is **Naegleria fowleri:**

1. **Acute and fulminating primary amoebic meningoencephalitis (PAM)** is caused by Naegleria fowleri.
2. **Clinical features of N. fowleri:** typical features of meningitis with rhinitis are seen. The key to diagnosis is clinical suspicion based on the recent history of swimming or diving in warm waters.

NAEGLERIA FOWLERI	ACANTHAMOEBA	BALAMUTHIA
• Causes PAM(primary amoebic meningoencephalitis) • Infective form -amoeboid form · • Resides in freshwater pond · • History of swimming in the freshwater pond	• Causes granulomatous encephalitis • contact lens keratitis	Causes granulomatous encephalitis

Flagellated forms-enter nares-cribriform plate-brain-PAM

·So-called "brain-eating amoeba"

- **Option-2-** Acanthamoeba species have been isolated as etiologic agents in **Acanthamoeba keratitis (image below)**: A. castellanii, A. polyphaga.
- **Option-3-** Disease: Granulomatous amebic encephalitis and Ulcerative keratitis
- **Option-4-**E. histolytica and Entamoeba coli do not cause meningoencephalitis.

Q.2 A 35 year old army person has been plagued by a painful, erosive lesion near his ear lobe since his return from his duty in desert area. Punch biopsy of the leading edge of the erosion reveals macrophages distended with oval amastigotes. Which of the following is the causative organism?

A. T. brucei

B. T. cruzi

C. Leishmania donovani

D. Plasmodium vivax

Solution:

- **Leishmania donovani** is the causative organism.
- The clinical scenario is suggestive of **Leshmaniasis.**
- **Sandfly (Genus Plebotomus)** transmits **Leishmaniasis (caused by Leshmania donovani).**

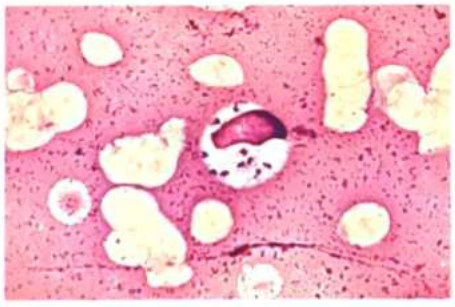
Option 1 **Trypanosoma brucei:** leads to sleeping sickness., transmitted by tse tse fly (Glossina species).

Option 2 Trypanosoma cruzi: reduviid bugs (Triatomine).

Option 3 **LEISHMANIA:**

- **L. donovani** → causes Kala Azar [Vector → Phlebotomous (Sand Fly)]
- **L. Tropica** → causes Oriental sore/ Delhi boil or Baghdad boil/ Aleppo button
- **L. brasilensis** → causes Espundia [Muco cutaneous leishmaniasis]

Punch biopsy of the leading edge of the erosion reveals macrophages distended with aflagellar amastigotes, also known as **Leishman Dononvan bodies**. Refer to the image below.



Option 4 Plasmodium vivax: transmitted by female anopheles.

10 PARASITOLOGY PART-2 (HELMINTHOLOGY)

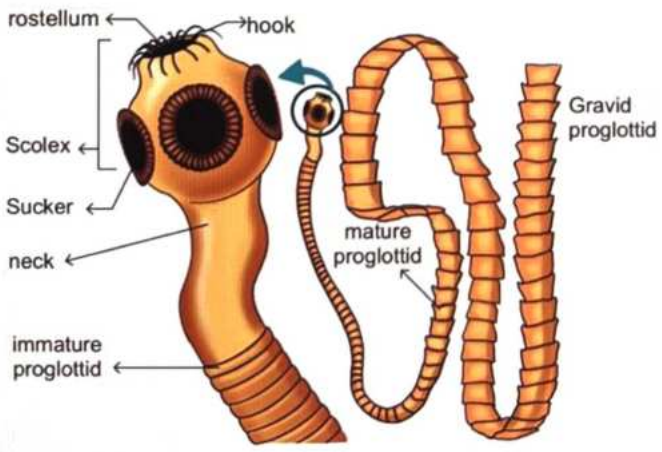
CESTODES	TREMATODE	NEMATODES
• Tape like • Monoecious	• Leaf like • Monoecious except Schistosomes (Dioecious)	• Cylindrical • Dioecious
• Suckers with hooks	• Suckers without hooks	• Have Buccal capsule
• Absent alimentary canal	• Poorly developed alimentary canal	• Well developed alimentary canal
• Body cavity absent	• Body cavity absent	• Body cavity present

CESTODES

00:05:46

Basic Structure

Scolex	Neck	Strobila
• Consists of o Suckers o Rostellum with or without Hooklets		• Contains Proglottids



Clinically important cestodes

Cestodes	Definite Host	Intermediate Host
• Taenia solium	• Man	• Pig
• Taenia Saginata	• Man	• Cattle
• Echinococcus granulosus	• Dog	• Sheep / Man
• H. nana	• Man	• Man
• Diphyllobothrium latum	• Man	• 1st – Cyclops • 2nd – Fish

1. DIPHYLLOBOTHRIUM LATUM/ FISH TAPE WORM

00:12:53

- Infective Form: Plerocercoid larva (infested in fish)
- Life cycle:
  - Infection is transmitted when man eats infected Fish → Larvae released into human body→ larvae changes to Adult Forms in small & large intestine and ↓Vit. B_{12} absorption → leading to Megaloblastic Anemia.
  - Adult Forms present in the Small intestine and Large intestine Produce eggs which go into water, Hatch into Coracidium larvae [L_1 Larvae]. Coracidium larvae reaches Cyclops & in Cyclops it changes to Procercoid [L_2 larvae]
  - This Cyclops containing Procercoid is eaten by Fish. Inside the Fish Procercoid changes into Plerocercoid larvae [L_3 larvae]

- Eggs of cestodes are non operculated (having lid)
- Only cestodes with operculated egg: Diphyllobothrium latum

Cyclops

D. latum egg

2. HYMENOLEPIS NANA / DWARF TAPE WORM

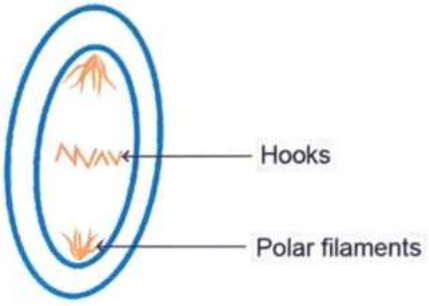
- Infective form: Eggs 00:20:03
- Life cycle:
  - Eggs are ingested with contaminated food & water →Eggs hatch in intestine → Cysticercoid larvae released → Attach to the intestinal layers → Changes to Adult form → Produce eggs → Eggs passed out In Feces
  - Sometimes Eggs hatch within intestine and Releases Hexacanth embryo which Penetrates the intestinal walls and gets back into circulation → leads to Auto infection

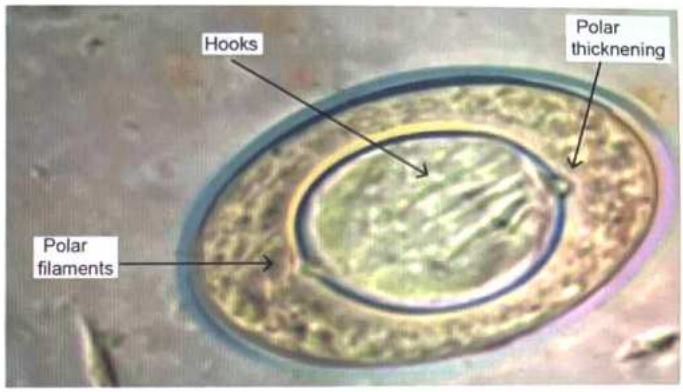
- EGG Contains Polar filaments and in between Central hooklets
- Treatment
  - DOC: Praziquantel
- Non bile stained eggs
- Non bile stained egg
  - **N** – **N**ecator Americanus
  - **E** – **E**ntrobius vermicularis
  - **H** – **H.** nana
  - **A** – **A**ncylostoma duodenale

How to remember

Non bile stained eggs: NEHA

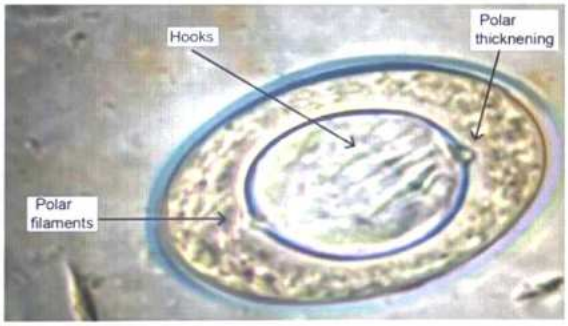
- Egg of H. Dimunita (rat tapeworm) does not have polar filaments and hooklets





Previous Year's Questions

Q. A child with 10 days abdominal pain presented to OPD. Stool microscopy was done which showed the given findings. What is the DOC for the disease caused by the given organism? (AIIMS - May- 2018)



A. Albendazole
B. Mebendazole
C. Praziquantel
D. Pyrantel Pamoate

3. ECHINOCOCCUS GRANULOSUS/ DOG TAPEWORM

00:31:23

- Infective form: Eggs
- Eggs of E. granulosus ingested with contaminated Food & water → Eggs hatch in duodenum → Releases Hexacanth embryo → Penetrates the intestinal layer →Through Portal circulation → Reaches Liver → Forms Hydatid cyst
- M/c site of Hydatid cyst: liver > lung > brain

- Hydatid cyst consists of 3 layers
  - Pericyst
  - Ectocyst
  - Endocyst (Germinal epithelium)
    - → Most active layer
    - → Secretes Hydatid fluid
    - → Brood capsule, Protoscolices are released from this layer

Hydatid cyst

- Tests done with Hydatid Fluid
  - Casoni Test
    - → Type 1 hypersensitivity: Immediate Wheal & Flare reaction demonstrate it to be a Type-1 HSN
    - → Obsolete now: because it may lead to anaphylaxis
- E. multilocularis: causes Malignant Hydatid Disease (not a malignancy)

Treatment of Hydatid Cyst

- In initial phase:
  - Albendazole
  - Albendazole + PAIR therapy (Percutaneous Aspiration injection Reaspiration therapy using Scolicidal agents)
- Definitive Treatment: Surgical Removal

Previous Year's Questions

Q.Slow growing alveolar like tumour in liver?

(AIIMS - Nov - 2019)

A. E. Granulosus
B. E. Multilocularis
C. Cysticercus cellulosae
D. Amoebic liver abscess

4. TAENIA SOLIUM AND SAGINATA

00:41:42

Taenia Solium	Taenia Saginata
• Pork tapeworm	• Beef tapeworm
• Armed tapeworm (Rostellum & hooklets present)	• Unarmed tapeworm (No Rostellum & hooklets)
• Proglottids number <1000	• Proglottids number >1000
• Larval form: Cysticercus cellulose	• Larval form: Cysticercus bovis
• Infection happens by eating contaminated pork (Cysticercus cellulosae) causes → Intestinal Taeniasis (Diarrheal episodes) and Neuro Cysticercosis (in Few cases) o NCC occurs M/c d/t ingestion of eggs of T. solium	• Infection happens by eating contaminated Beef (Cysticercus bovis) → Intestinal Taeniasis (Diarrheal episodes) o Nothing happen & if eggs are eaten

Previous Year's Questions

Q.Which is correct about larval stage of Taenia solium ?

(FMGE - June - 2019)

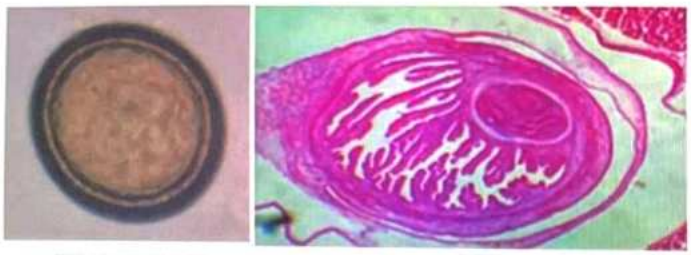
A. Larva currens
B. Cysticercosis Cellulose
C. Cutaneous Larvae Migrans
D. Visceral Larvae Migrans

Neurocysticercosis

- 2nd M/c ICSOL in India
- MC site: Sub arachnoid > Parenchymal
- Presents with Atypical Seizures
- IOC: CT/MRI
  - If calcified cyst: CT
  - Otherwise: MRI
- DOC: Albendazole (After giving steroids)

T. solium scolex

T. Saginata Scolex



Taenia egg

- Section through tissue infected with a bladder-worm or Cysticercus of taenia solium. skeletal muscle can be seen to the upper right and lower right

- Operculated eggs of Diphyllobothrium latum in stool wet mount

TREMATODES / FLIUKES

00:59:58

- Properties
  - Monoecious
  - Infective Form: Metacercariae (cyst like structure)
  - Have two Intermediate hosts
    - → 1st: Snail
    - → 2nd: Aquatic Vegetation, Fish, Crab / Cray Water Fish
  - All have Operculated Eggs (Lid)
- Exception to above properties: Schistosomes
  - Dioecious
  - Single intermediate host: Snail
  - Infective form: Cercariae
  - Have non operculated eggs

1. SCHISTOSOMES

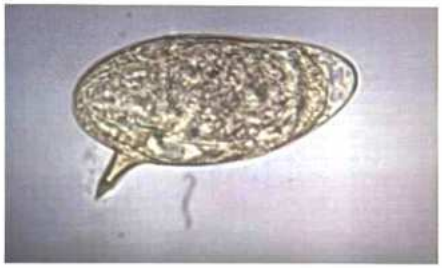
a. Schistosoma Haematobium

- Resides in Vesical plexus
- Causes SCC of Urinary bladder > Transitional cell Ca
- Causes Terminal Hematuria
- Egg has Terminal spine
- Eggs are excreted in urine

Schistosoma Haematobium

b. S. mansoni

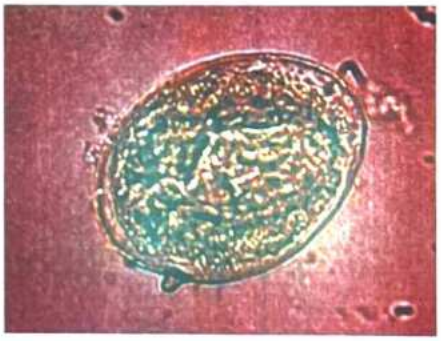
- Resides in Inferior Mesenteric plexus
- Causes Swimmer's itch & Katayama fever
- Eggs are having lateral spine
- Excreted in stool



Schistosoma mansoni

c. S. japonicum

- Resides in Superior Mesenteric plexus
- Causes Katayama Fever & Eosinophilic diarrhea
- Eggs are having a lateral Knob



Previous Year's Questions

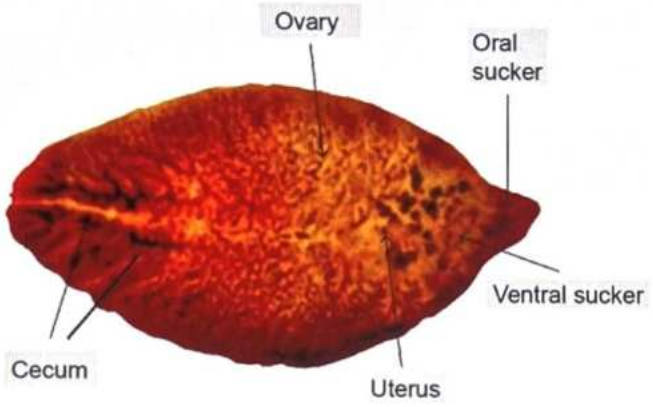
Q. Which parasite lives in bladder plexus?

(FMGE - Nov - 2017)

A. Schistosoma
B. Fasciola
C. Ascaris
D. Echinococcus

2. FASCIOLA HEPATICA / SHEEP LIVER FLUKE

- 1st Intermediate Host: Snail
- 2nd Intermediate Host: Aquatic vegetation (with Metacercariae)
- Man eats this aquatic vegetation → Metacercariae is ingested → In duodenum Excystation occurs → Larvae released → Penetrates intestinal layers → Penetrates Liver capsule →Penetrates bile ducts →Matures to Adult Forms → Produce Eggs → Eggs passed out in feces →Comes in contact with water → Eggs hatch up into → Miracidum → Sporocysts → Radiae (1st generation & 2nd generation) → Cercariae → Metacercariae (cyst like structure which forms in the aquatic vegetation)



Fasciola hepatica

F. hepatica egg (Operculated)

- Sheep liver infected with F. hepatica if eaten by man, will develop: Halzolin syndrome
  → Halzolin syndrome: Laryngeal edema

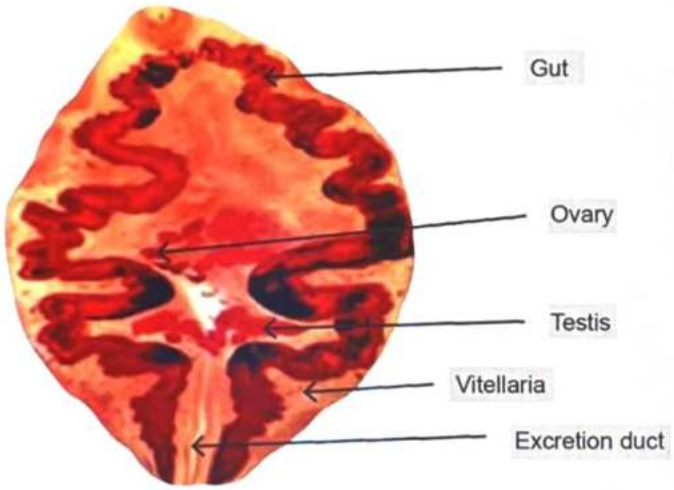
3. CLONORCHIS SINENSIS

- Aka Oriental liver fluke / Chinese liver fluke
- 1st Intermediate host: Snail
- 2nd Intermediate host: Fresh water Fish (with Metacercariae)
- Metacercariae ingested by man after eating fresh water fish → Excystation in duodenum → Larvae released → Penetrates intestinal layers → Penetrates Liver capsule → Penetrates bile ducts and matures to adult forms causing constant inflammation & fibrosis of bile ducts → leading to Cholangiocarcinoma aka Bile duct carcinoma

4. PARAGONIMUS WESTERMANII

01:24:42

- Aka Lung Fluke
- 1st Intermediate host: Snail
- 2nd Intermediate host: Crab/Cray water Fish (with Metacercariae)
- Eaten by man → Metacercariae ingested → Excystation in Duodenum → Larvae released → Penetrates intestinal layers → Penetrates diaphragm → Penetrates Rt. Lung capsule → Enters into Rt. Lung (mostly) → change into Cysts and cause Erosion → Red Brown sputum/ Golden brown sputum (Endemic Hemoptysis)



Previous Year's Questions

Q.A 35-year-old man presented with dry cough & rusty coloured sputum. He has history of eating in Chinese restaurant very often with consumption of crabs often: what is the causative agent in this condition?

(NEET - Jan - 2019)

A. Diphyllobothrium latum
B. Pneumocystis jirovecii
C. Paragonimus Westermanii
D. Strongyloides stercoralis

5. FASCIOLOPSIS BUSKII

- Largest intestinal fluke

NEMATODES

Classification

- Intestinal Nematodes
  - Small Intestinal Nematodes
    - → Ascaris
    - → Hook worm
    - → Necator americanus
    - → Strongyloides Stercolaris
  - Large Intestinal Nematodes
    - → Trichuris trichiura
    - → Enterobius vermicularis
  - Miscellaneous Nematodes
    - → Dracunculus medinensis
    - → Trichinella Spiralis (muscle worm)

- Tissue Nematodes
  - Resides in Lymphatics
    - → Wuchereria bancrofti
    - → Brugyia Malayi
    - → Brugyia timori
  - Resides in Subcutaneous tissue and eyes
    - → Loa-Loa
    - → Onchocerca volvolus
  - Resides in Serous cavities
    - → Mansonella Ozzardi
    - → Mansonella perstans

Transmission By

- Eating Eggs
  - **E** - **E**nterobius vermicularis
  - **A** - **A**scaris
  - **T** - **T**richuris Trichiura

How to remember

EAT

- Penetration of Sole of Foot (Filariform larvae)
  - **A** - **A**ncylostoma duodenale
  - **N** - **N**ecator americanus
  - **S** - **S**trongyloides Stercolaris

How to remember

ANS

Lungs for maturation

- **A**^{2} - **A**ncylostoma duodenale, **A**scaris
- **N** - **N**ecator americanus
- **S** - **S**trongyloides Stercolaris

How to remember

A^{2}NS

LARGE INTESTINAL NEMATODES

01:43:09

1. Trichuris trichiura

- Aka whip worm
- Eggs are ingested with contaminated Food & water → Hatch up in caecum → Larvae released which mature into Adult Male worms and Female worms → Fertilize and produce Eggs (dumbbell shaped / barrel shaped)

Male Female

- Egg
  - Barrel/dumb bell shaped egg
  - Have mucous plugs on both sides

Egg c̄
mucous plugs

- Causes Fe deficiency anemia
- Causes Frequent Rectal prolapse in children
  - Coconut Cake Like Rectum

Previous Year's Questions

Q.Identify the egg shown in image ?

(FMGE - Aug - 2020)

A. Ascaris Lumbricoidoes
B. Enterobius Vermicularis
C. Trichuris Trichiura
D. H. Nana

Previous Year's Questions

Q.A 5-year-old child presented to OPD with complaints of rectal prolapse: on examination stunting & growth retardation was documented. What is parasitological cause for this clinical feature?

(NEET-Jan - 2019)

A. Trichuris Trichiura
B. Trichinella Spiralis
C. Giardia Lamblia
D. Enterobius vermicularis

2. Enterobius Vermicularis

- Aka Pin Worm / Thread Worm / Seat Worm / Oxyuris
- Infective form: Eggs
- Presents with Perianal pruritis → Eggs deposit in nail beds → Child putting up fingers in mouth → Eggs are ingested → Autoinfection

- Autoinfection is seen in
  - **C^2 – C**apillaria Phillipensis, ryptosporidium parvum
  - **H – H.** nana
  - **E – E.** vermicularis
  - **S – S.** Stercolaris
  - **T – T**aenia Solium

- Egg
  - Plano convex egg
  - Having a Tadpole like larvae
  - Non bile stained egg

E. vermicularis

Enterobius egg

- NIH swab
  - National Institute Health Swab
  - Made up of glass rod with cellophane tape

NIH swab

- DOC: Albendazole

Previous Year's Questions

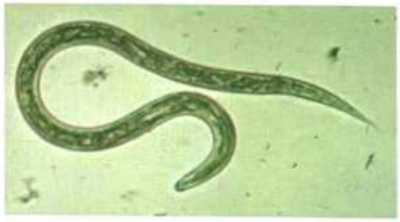
Q. Identify the egg shown in image? (FMGE - Dec - 2020)

A. Trichuris Trichiura
B. Enterobius Vermicularis
C. Ankylostoma
D. Ascaris

SMALL INTESTINAL NEMATODES

1. Ancylostoma Duodenale / Hook Worm

- Causes iron deficiency Anemia
- Life cycle:
  - Filariform larvae enters into the sole of foot → Causes Ground itch/ Maculopapular dermatitis → penetrates all skin layers → Goes into Cutaneous circulation →Goes into Lymphatics → enters the venous circulation → Portal circulation → Liver →from liver it goes to Rt. Side of heart → Lungs
  - After slight maturation it starts retrograde journey from lungs → Penetrates pulmonary capillaries → Alveoli → Bronchiole → Trachea → Larynx → Pharynx→ Swallowed with sputum
  - In the Esophagus, 3rd moulting happens → In duodenum, 4th moulting happens → Fully Mature larvae forms → Male and Female worms are formed
  - Male and Female worms Fertilize to form Eggs → Eggs passed out in Feces →goes into Soil → Eggs in soil undergo Moulting and change into Rhabditiform larvae (1st moulting) and undergo further moulting (2nd moulting) into Filariform larvae (Needle like, Infective Form)



Rhabditiform Larva

Ancylostoma egg

a. A. Brasilensis and A. Caninum 02:07:06

- Animal worm, they cannot penetrate all skin layers so they get trapped b/w skin layers which leads to Cutaneous larvae Migrans

Cutaneous larvae migrans

Previous Year's Questions

Q. Cutaneous larvae migrans caused by which organism?

(NEET-Jan - 2018)

A. Anisakiasis Simplex
B. Toxocara Species
C. Ancyclostoma braziliensis
D. Necator americanus

Previous Year's Questions

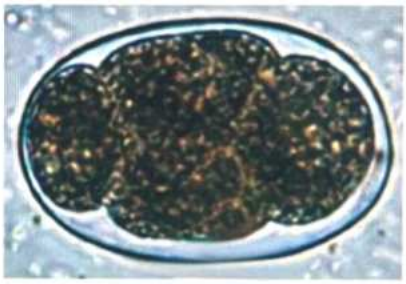
Q. A person work[illegible] which of the following profession can have the problem shown in the image?

(AIIMS-May - 2019)

A. Person is working in the butcher house
B. Life guard in swimming pool
C. Kennel worker
D. Poultry farm worker

b. A. Duodenale

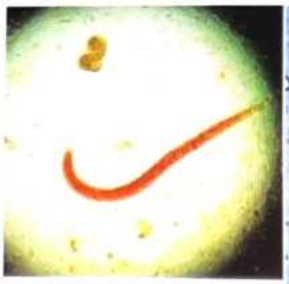
- Causes Fe deficiency anemia (Blood Sucked : 0.2 ml / day / worm)
- Egg
  - Segmentation is seen (Blastomeres)
  - Chandler's Index: >300 eggs/gm of Stool then its termed as a Social problem



Blastomeres

c. S. Stercolaris

- Strongly a/w HIV +ve
- Ovo – viviparous: that's is eggs immediately hatch into larvae
- Undergo Auto infection
  - This Autoinfection can turn into Hyper Infection Syndrome in HIV +ve
- Undergoes parthenogenetic development: Females directly lay eggs without male
- Associated with a form of Cutaneous larvae migran k/a Larvae Currens
  - Moves faster under skin
- DOC: Ivermectin

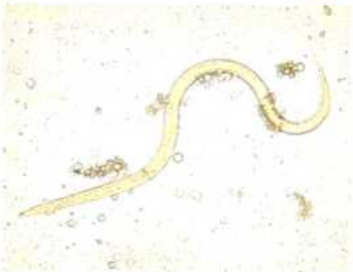


Strongyloides Stercolaris

Previous Year's Questions

Q. Patients with history of kidney transplantation presenting with diarrhoea. The motility of the worms is given. Correct statement is?

(AIIMS - Nov - 2018)



A. Monoecious & the organism related with pathogenesis
B. Transmitted by intake contaminated food & water
C. Loffler's pneumonia is not caused by same organism
D. Body gets this infection through filiform larvae

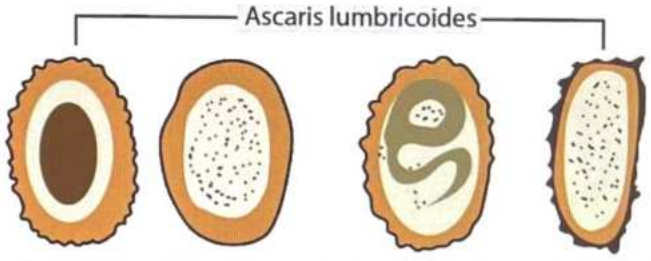
2. Ascaris

02:16:52

- Life cycle:
  - Eggs are ingested with contaminated food & water → Hatch up in duodenum → Larvae released → penetrates intestinal layers → gets into Portal circulation → Liver → Rt. Side of heart → Lungs (2nd & 3rd moulting happens)
  - From lungs Retrograde journey starts by penetrating pulmonary capillaries → Alveoli → Bronchiole → Trachea → Larynx → Pharynx → Swallowed with sputum →In duodenum, 4th moulting occurs → Matures into adult Male and Female worms → Fertilize → form Eggs →Passed out in Feces into Soil where 1st moulting happens and forms Rhabditiform larvae → This egg is eaten by man
- 2nd and 3rd moulting leads to Loffler's syndrome
- Ascaris causes Intussusception which can lead to Intestinal obstruction d/t massive Ascaris load
- Strongly a/w Loffler's syndrome having Massive Eosinophilia with Loffler's pneumonia
- Both fertilized and unfertilized eggs are Bile stained
- Eggs which float[illegible] salt solution
  - F - Fertilised egg of Ascaris
  - A - Ancylostoma duodenale
  - T - Trichuris trichiura
  - E - Enterobius vermicularis
  - H - H. nana

How to remember

FATEH



Previous Year's Questions

Q. Image shown here is? (INICET - Nov - 2020)

A. Ascaris
B. Ancylostoma duodenale
C. Enterobius vermicularis
D. Strongyloides Stercoralis

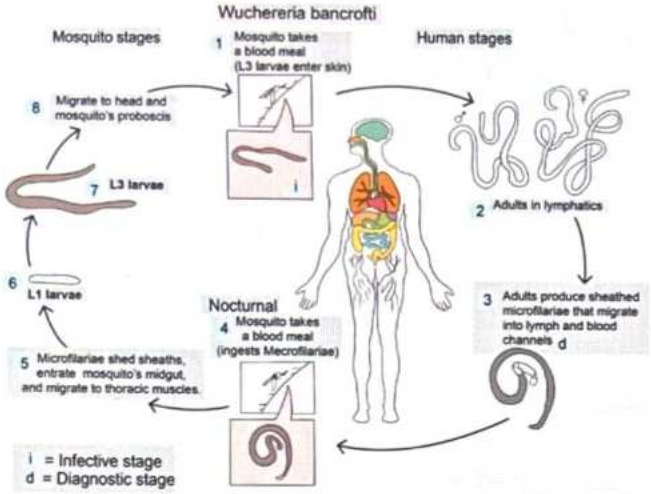
TISSUE NEMATODES

02:26:02

1. Wuchereria Bancrofti

- Infective Form: L_3 Filariform larvae
- Transmitted by
  - Culex (mainly)
  - Aedes
  - Anopheles
- Larvae goes into lymphatics where they change into adult forms (male and female) → fertilization occurs and new larvae are produced which are K/a microfilariae

- These microfilaria are sheathed and no nuclei seen on tail tip
- These microfilaria are having Nocturnal periodicity (10 pm – 2am)



- Clinical manifestation:
  - Acute Filariasis Features:
    - → Fever
    - → Lymphadenitis
    - → Lymphangitis
  - Chronic Filariasis Features
    - → Elephantiasis
    - → Hydrocele
    - → Granuloma breast
    - → Chyluria and Albuminuria: white urine

- Lab Diagnosis
  - Blood Smear Examination: Taken at night time d/t Nocturnal periodicity
  - DEC Provocative Test:
    - → DEC irritates Wuchereria & Wuchereria comes in blood → Do blood smear examination → Look for Microfilariae (Tail Tip having no nuclei)
    - → Tail tip with no Nuclei seen in W. bancrofti
    - → Tail Tip with 2 terminal Nuclei: Microfilariae of Brugyia malayi

Wuchereria bancrofti Brugia malayi

Loa loa

  - Radiological
    - → Chest X ray
    - → CT Scan
    - → USG of Scrotum: dancing worm sign
  - PCR For W. bancrofti
    - → Even picograms of DNA will be detected
    - → Best investigation

Elephantiasis

Hydrocele of Scrotum

Previous Year's Questions

Q. Pulmonary eosinophilia is seen d/t which of following infection?

(FMGE - Nov - 2017)

A. Ancyclostoma
B. Trichinella
C. Filaria
D. Roundworm

2. Onchocerca volvolus

02:40:19

Skin Disease	Eye Disease
• Onchocercoma: Subcutaneous nodules	• River Blindness: o By Simulium Fly / Black Fly o Microfilariae deposit in eyes of people residing near the river bed

- DOC: Ivermectin

Black Fly

Onchocercoma

3. Loa-Loa

Skin	Eye
• Leads to Calabar Swellings: Migratory swellings	• Leads to Loasis • Caused by: By chrysops fly/ Deer fly

4. Dracunculus medinensis

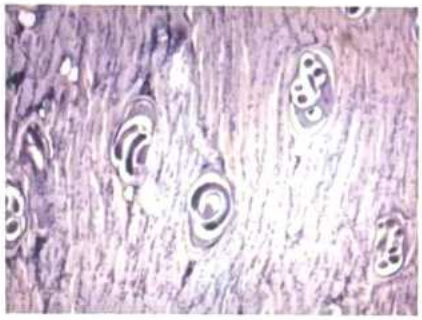
02:43:45

- Aka Guinea Worm
- Eradicated from India
- Last case was reported from Rajasthan
- Match stick method was used to remove larvae from lesion (Blister)

Matchstick method for guinea worm

5. Trichinella Spiralis / muscle worm

- Transmitted by eating larvae in uncooked pork
- Viviparous
- Finally settle in muscles



Trichinella Spiralis

Visceral Larvae Migrans seen in

- Toxocara canis
- Toxocara catis
- Angiostronglus cantonensis
  - o Leads to Eosinophilic meningitis

CLINICAL QUESTIONS

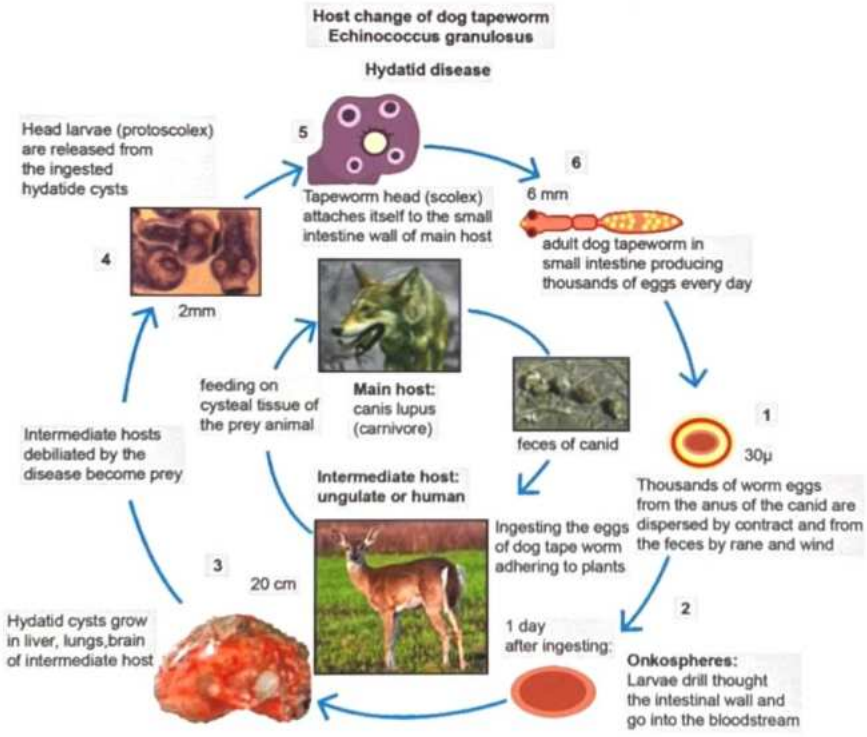
Q.1 A 37-year-old male presented to the OPD with RUQ pain and intermittent low-grade fever. Both USG and CT demonstrated a cystic lesion with vesicles at its periphery in segments 6 & 7 of the liver. The patient showed a positive Casoni's test. Which of the following is the intermediate host for the given condition?

A. Man
B. Dog
C. Cat
D. Foxes

Solution:

- **Man is the intermediate host** for the given condition.
- The clinical scenario is indicative of **hydatid disease.**
- **Echinococcus granulosus:**

1. Hydatid disease is caused by Echinococcus granulosus.
2. **Definitive host:** Dog and wild carnivores.
3. **Intermediate hosts:** Man and other herbivorous animals.
4. Man is an accidental host (dead end).
5. Eggs: Infective stage of the parasite.
6. Eggs transform to larva (hydatid cyst) that penetrate GIT and migrates to various organs like liver.



Q.2 A 26 year old male presented to the OPD with fever, lymphangitis and swollen lymph nodes. The clinical diagnosis of lymphatic filariasis was made by the physician. Which of the following organism cannot be the causative agent?

A. W. bancrofti
B. Brugia malayi
C. Onchocera volvulus
D. B. timori

Solution:

- **Onchocera volvulus** cannot be the causative agent. It causes **subcutaneous Filariasis.**
- **Lymphatic Filariasis is caused by:**
  - W. bancrofti
  - Brugia malayi
- B. timori

WUCHERERIA BANCROFTI:

- **INFECTIVE Form** → L3 Filariform larvae

Transmitted by:

- Culex [mainly]
- Aedes
- Anopheles

Acute Filariasis Features :

- Fever, lymphadenitis, lymphangitis

Chronic Filariasis Features:

- Elephantiasis
- Hydrocele
- Granuloma breast
- Kidney LN block up → Chyluria, Albuminuria

LAB DIAGNOSIS:

1. BLOOD SMEAR EXAMINATION: Taken at night time due to Nocturnal periodicity
2. DEC PROVOCATIVE TEST : Look for Microfilariae (Tail Tip)
3. RADIOLOGICAL:
   - X ray
   - CT Scan
   - USG of Scrotum → DANCING WORM SIGN Seen
4. PCR For W. bancrofti: Best investigation

LEARNING OBJECTIVES

UNIT 5: VIROLOGY

- Virology
  - General properties of virus
  - DNA virus: Herpes family, Pox virus, Parvo virus and Adeno virus
  - RNA virus
    - → Picorna viridae: Polio Virus, Coxsackie virus, Entero virus, ECHO virus and Rhino Virus
    - → Myxo viruses: Orthomyxoviridae and Paramyxoviridae
    - → Rabies virus
    - → Hepatitis virus
    - → HIV
    - → Arboviruses
  - Oncogenic viruses
  - Prions
  - Viral hemorrhagic fever causing viruses
- Covid 19

11 VIROLOGY PART-1 (GENERAL PROPERTIES AND DNA VIRUSES)

GENERAL PROPERTIES OF VIRUSES 00:01:10

- Viruses are obligate intracellular organisms
- Contains either DNA or RNA (never both)
- Structure
  - Consist of a Genome which is surrounded by a Capsid
  - Capsid is surrounded by caspomeres
  - Some times caspid is also surrounded by an Envelope

Important Properties

- Smaller in Size
- Lack the enzyme necessary for Protein & Nucleic acid synthesis
- Do not divide by binary fission
- Do not culture in any pure culture media
- Contain either DNA or RNA
- Resistant to antibiotics
- Host specificity present
  - Animal Virus
  - Plant Virus
  - Bacteriophage

Morphology of virus 00:07:01

- Structure
  - Central core of Nucleic acid of a Virion is called Genome
  - Genome is surrounded by a protein coat called Capsid [made of Caspomeres]
  - Genome & surrounded Capsid → nucleocapsid

- Functional of caspid
  - Impermeable shell around genome
  - Introduction of Viral genome into host cell by adsorbing on cell surface
  - Capsid envelope is a Bilayered lipoprotein
    → Virus codes glycoprotein subunits on the surface of envelope (Eg. Peplomers)
    → Peplomers – H(haemagglutinin) & N (neuraminidase) present in influenza virus

- Symmetry: Capsid is arranged into 3 types of symmetries
  - Icosahedral
    → Most of the viruses
    → It is having a polygonal shape with 12 vertices / corner having 20 facets (sides)
    → Facet is an equilateral triangle
  - Helical: Present in some RNA Viruses
  - Complex: Eg. Pox Virus

- Shapes of Viruses
  - Bullet Shaped: Rabies virus (speed of virus is 3mm/hr)
  - Rod Shaped: Tobacco mosaic virus
  - Brick shaped: Pox virus
  - Space vehicle: Adeno virus
  - Filamentous: Marburg & Ebola virus

Properties of virus 00:16:08

- Chemical properties
  - Inactivated at 56°C for 30 min
  - Stable & can be stored at -40 to -70°C by lyophilization
- Radiation: UV rays, X Rays, Gamma rays inactivate the virus
- Lipid solvent property: Chloroform, Ether & Bile salts destroy enveloped viruses easily i.e. enveloped viruses are less resistant than non enveloped viruses

Non-enveloped DNA viruses

- P – Parvo
- A – Adeno
- P – Papovaviridae

How to remember

- PAP

Non- Enveloped RNA viruses

- P – Picornaviridae (Eg. Hep A)
- A – Astroviridae
- R – Reoviridae
- C – Calciviridae (Eg. Hep E)

How to remember

- PARC

Replication 00:23:25

- Steps

1. A - Adsorption

2. P - Penetration
3. U - Uncoating
4. B - Biosynthesis/Assembly
5. M - Maturation
6. R - Release

How to remember

- A PUB MR

1. Adsorption
- Comes in contact randomly with the cells
- Depends on presence of specific receptors
- Eg
  - In HIV: Envelope proteins GP 120 / GP 41 binds to CD_4 T cell receptors
  - Influenza Virus A
    - → H (Hemagglutinin): Adsorption on Pharyngeal cells
    - → N (Neuraminidase): Receptor destroying enzyme

2. Viropexis / Penetration
- Viropexis: after attachment, the virus particle is taken inside the cell
- In enveloped viruses, envelop fuses with the plasma membrane of host cell releasing the Nucleocapsid into the cytoplasm

3. Uncoating
- Physical separation of Nucleic acid from Capsid occurs
- Enveloped viruses enter cell by receptor mediated Endocytosis & lose their outer covering by action of Lysosomal enzymes

4. Bio synthesis 00:32:44
- Viral genome directs, the biosynthesis machinery of the host cell to synthesize products for "New Progeny Virus"
- Viral proteins are synthesized mostly in cytoplasm
- Most of the DNA Viruses replicate in the Nucleus except Pox virus (replicate in cytoplasm)
- Most of the RNA viruses replicate in the cytoplasm except Influenza & Retro virus (replicate in Nucleus)
- Most of DNA Viruses are Double Stranded except Parvo virus
- Most of RNA viruses are Single Stranded except Reoviridae virus
- In case of SS DNA virus (Parvo virus)
  - Viral nucleic acid utilizes cellular DNA polymerase for making viral DNA double Stranded
- Double stranded DNA Virus
  - Transcription: Viral nucleic acid enters the Nucleus of host cell and undergo transcription of Parental DNA of virus producing Early mRNA
  - Translation: by the process of translation early non-structural proteins are produced which shutdown host cell machinery there by forming Progeny DNA
  - Progeny DNA undergo transcription to produce Late mRNA and then undergo translation to form Late structural proteins (Capsid & Envelope proteins)

- In case of RNA Viruses: Single stranded RNA Viruses are classified into 2 categories
  - Type-1: The Positive sense ssRNA [Plus strand]
    - → They have the same polarity as of mRNA → they directly translate into early proteins
    - → Early proteins have an RNA polymerase activity because of which they direct the replication of positive sense ssRNA → Negative sense ssRNA →and again to positive sense ssRNA → Late proteins are produced
    - → Eg. Picorna &Toga virus

  - Type-2: The Negative sense ssRNA[minus strand]
    - → They Posses RNA Polymerase activity to translate into Positive sense RNA which produce proteins
    - → This positive sense ssRNA act as a template to undergo replication and produce negative ssRNA

- Type-3: Double stranded RNA viruses
  - Eg. Reoviridae
  - Have segmented genome
  - Each segment codes for 1 Polypeptide
  - They contain a positive strand RNA which act as mRNA which undergo translation to produce Proteins
  - Both + & - strands serves as templates for synthesis of complementary strands to form a duplex

- Type-4 RNA virus: Retro Virus Eg. HIV
  - HIV and other Retrovirus have 2 copies of positive sense ssRNA genome (non-segmented)
  - After the entry into the host cell, the single stranded RNA gets reverse transcribed to produce a ssDNA by Viral Reverse transcriptase enzyme
  - Viral Reverse transcriptase enzyme also act as RNA dependent DNA polymerase
  - A genome having DNA : RNA Hybrid is being formed
  - Reverse transcriptase enzyme also has Ribonuclease activity by which it digest RNA from DNA : RNA Hybrid
  - This resulting ssDNA is converted into dsDNA by DNA polymerase activity
  - This dsDNA is transported to the nucleus where it

gets integrated into the host genome with the help of viral Integrase enzyme
  - This Integrated DNA serves as a template to produce mRNA which is later translated into proteins and genomic progeny RNA

5. Assembly
- Viral nucleic acid & proteins are packaged to form progeny viruses [Nucleocapsid]
- Assembly may takes place in host cell Nucleus or cytoplasm
- DNA viruses mostly assemble in nucleus except Pox & Hepadenavirus (Hep B)
- RNA viruses mostly assemble in cytoplasm

6. Maturation 01:05:42
- May take place in Nucleus or Cytoplasm
  - In Nucleus: Herpes, Adeno Virus
  - In Cytoplasm: Picorna, Pox virus
- In case of enveloped virus, the enveloped is derived from nuclear membrane (if virus assemble in nucleus) or form plasma membrane during the process budding (if virus assemble in cytoplasm)

7. Release
- Release can happen by Lysis and Budding
- Lysis: Lysis of host cell is done by non-enveloped viruses & Bacteriophage
- Budding: happens in enveloped virus and they acquire a part of host cell membrane to form Lipid part of envelope
- Eg. Envelope is acquired in influenza virus from plasma membrane & for herpes virus from Nucleus membrane
- Viral glycoproteins are inserted into the envelopes
- Excess viral glycoproteins are synthesized to saturate cell receptors so that virus will not stick to host cells following Release

Refer Figure 11.1

Abnormal Replicative Cycles 01:15:54
- Incomplete viruses
  - Result from defective assembly: proteins are assembled without nucleic acid
  - Eg. Von Magnus Phenomenon
    → High Hemagglutination titer but low infectivity

- Pseudo Viruses
  - During replication of viruses, Capsid occasionally encloses Host cell nucleic acid Instead of viral nucleic acid. They are neither infective nor replicative

- Abortive infection: d/t wrong selection of host cells by virus

Mutation
- Occurs at 10^{-4} to 10^{-10} per base pair per generation
- Mutation can happen by 5-Fluorouracil, UV light
- Conditional lethal mutants
  - Can grow only in some permissive conditions
  - Eg: Temperature sensitive mutants → can grow at 28-31°C but not at 37°C, used for making influenza vaccines

Reassortment
- Mostly happens in influenza virus A
- Reassortment happens in influenza due to its segmented genome
- Antigenic shift is due to reassortment

Viral interference 01:23:05
- Two viruses infect a host cell, one virus leads to inhibition of other virus k/a viral interference
- One virus can do blocking of the host cell receptor or destroys host cell receptor resulting in growth inhibition of other virus
- The 1st virus may compete with 2nd virus for replication apparatus & can induce Interferon production to kill second virus
- Eg. Rubella (Togaviridae), Polio (Picornaviridae)

Cultivation of Virus 01:24:28
1. Hen's Egg Inoculation
2. Animal Inoculation
3. Cell Culture / Tissue Culture

1. Hen's Egg Inoculation / Candling
- CAM(Chorio Allantoic Membrane)
  - Organisms which grow on CAM: Variola and Vaccinia (both belong to pox virus)
- Allantoic cavity
  - Influenza Vaccine production
  - Organism grown are
    → **Y** - Yellow fever [17D]
    → **I** - Influenza
    → **R** – Rabies

How to remember
- YIR

- Amniotic sac: primarily used for isolation of influenza virus
- Yolk sac: organism grown are
  - **C** - Chlamydia
  - **A** - Arbovirus
  - **R** - Rickettsiae

How to remember

- CAR

2. Animal Inoculation

- Newborn suckling mice (<48 hr old) used for proper cultivation of Arbovirus, Coxsackie Virus
- Routes of inoculation
  - Intracerebral
  - Intranasal
  - Intraperitoneal
  - Sub Cutaneous
- Animals are observed for the signs of illness & stained for inclusion bodies

3. Cell culture lines [Growth medium]

- Primary cell lines
  - Obtained from fresh organs of animals or humans
  - Allow only 5-10 divisions
  - Eg
    - → Rhesus monkey Kidney cell lines
    - → Chick embryo fibroblast cell lines
- Diploid / secondary cell lines
  - Allow 10-50 divisions
  - Can be used for longer storage of virus
  - Eg. Human Fibroblast cell lines (WI-38)
- Continuous cell lines
  - Unlimited divisions
  - Eg
    - → Hela: derived from Cervical cancer of a lady
    - → Hep 2: derived from Human Epithelioma of larynx
    - → BHK: derived from Baby Hamster Kidney
    - → Vero: derived from Vervet Monkey Kidney cell line

Cytopathic Effect 01:35:08

- Helps in identification of Virus
- Multinucleate Giant cell formation seen in: Measles Virus/ RSV
- Cell necrosis/lysis seen in: Enteroviruses
- Cell clumping [grape like clusters] seen in: Adenovirus
- Cytoplasmic vacuolation seen in: SV – 40

Inclusion Bodies 01:37:06

- Replicating aggregates of the virus either intranuclear or intracytoplasmic
- Can be seen with normal power microscope

Intra Cytoplasmic Inclusion Bodies [Eosinophilic]

- Negri bodies: Rabies
- Guarneri bodies: Vaccinia
- Paschen bodies : Variola
- Bollinger bodies : Fowl pox
- Henderson Peterson/ Molluscum bodies: Molluscum contagiosum

Intra Nuclear Inclusion Bodies

- Cowdry type A inclusions seen in
  - H - Herpes virus (Lipschultz bodies also seen)
  - A
  - Y - Yellow Fever virus (Torres bodies also seen)

How to remember

- HAY

- Cowdry type B inclusions are more circumscribed & seen in
  - B
  - A - Adeno virus
  - P - Polio virus

How to remember

- BAP

- Intra nuclear + Intra cytoplasmic Inclusion Bodies aka Warthin Finkelday Giant Cells Seen in: Measles
- Intra nuclear + Intra cytoplasmic inclusion bodies also seen in: CMV (Owl eye appearance of infected cells)

Previous Year's Questions

Q. Match the following? (AIIMS - Nov - 2019)

1. Molluscum contagiosum	A. Henderson Patterson bodies
2. Papilloma virus	B. Decoy cells
3. BK polyoma virus	C. Owl's eye
4. CMV	D. koilocyte

1. A
2. D
3. B
4. C

DNA VIRUSES 01:43:21

- H^2 – Herpes, Hepadena
- A – Adeno
- P – Parvo

- P – Papova
- P – Pox
- Y – Y

How to remember

- H^2APPPY

Herpes Famiy

01:45:00

αVirinae	βVirinae	γVirinae
• Includes: ○ HSV1 ○ HSV2 ○ HHV3 (Verizella zoster)	• Includes: ○ HHV 5 (CMV) ○ HHV 6 ○ HHV 7	• Includes ○ HHV 4(EBV) ○ HHV 8 (Kaposi sarcoma)
• Affects Ganglionic cells	• Affects salivary glands	• Affects B cells (d/t CD_{21}/CR_2 Receptors over B cells)

- HHV 3: Verizella zoster [chicken pox virus]
- HHV 4: EBV
- HHV 5: Cytomegalo virus
- HHV 8: Kaposi Sarcoma causing virus
- HSV 1: Herpes simplex virus 1
- HSV 2: Herpes simplex virus 2

Case scenario:
An irritable 18-month-old toddler with fever and blister-like ulcerations on mucous membranes of the oral cavity refuses to eat; the symptoms worsen and then slowly resolve over a period of 2 weeks. Six months later, the child develops a single vesicular lesion the resolves in 6 days, she does not have fever. Which of the following scenarios in most likely

Diagnosis: HSV 1

- HSV 1: Causes infection above the waist
- HSV 2: Causes infection below the waist
- While primary HSV-1 infections are usually asymptomatic, symptomatic disease occurs most frequently in small children (1-5 years old).
- Buccal and gingival mucosa is most often involved, and lesions, if untreated, may last 2 to 3 weeks.
- Reactivation results in sporadic vesicular lesions that heal more quickly than the primary lesions.
- Following primary oral infection, whether symptomatic or asymptomatic, the virus enters the trigeminal ganglia and remains latent there throughout life
- The virus may reactivate under conditions of physical, emotional, or mental stress and it the individual becomes immunocompromised from other infections or medical conditions.
- Each individual is unique regarding reactivation of HSV (type 1 or type 2), with some having no symptomatic reactivation, some one or two, and some multiple instances throughout their lives; therefore, one cannot say that the vesicular lesions will not recur.
- Guillain- Barre syndrome is more with other human herpes viruses (VZV, CMV, and EBC) and bacterial infections (campylobacter Jejuni and Mycoplasma pneumoniae)...

1. HSV 1 and 2

01:55:32

Mucosal involvement

- M/c site: Buccal mucosa
- Causes: Acute gingivo stomatitis
- M/c recurrent herpes lesion: Herpes labialis

Cutaneous involvement

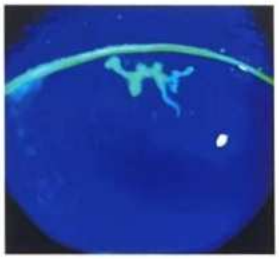
- M/c site: Face
- Herpetic whitlow: Occurs in Nurses, doctors
- Herpetic Gladitorum: Occurs in Wrestlers

CNS involvement

- Temporal lobe encephalitis
- Mollaret's Meningitis: Recurrent lymphocytic meningitis

Ophthalmic involvement

- Cause acute dendritic ulcers
- Steroids are contraindicated
- Topical Acyclovir indicated



Genital involvement

- Causes B/L Vesicular, Pustular, Erythematous lesions
- 36week Pregnant female with HSV2 infection near term: Cesarean section done to avoid transmission to newborn + Acyclovir to decrease viral load

Lab diagnosis

- Tzanck cell demonstration:
  - Tissue biopsy + Toluidine blue → can see Multinucleated giant cells / Tzanck cells
  - It is characteristic of all α Virinae (HSV 1, 2, and 3)
- PCR and ELISA for HSV-1, HSV-2

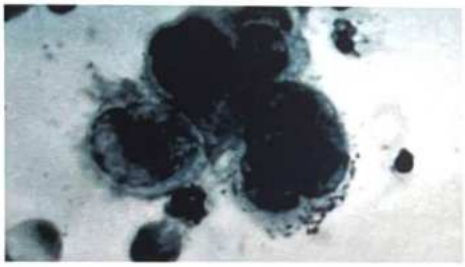
Previous Year's Questions

Q. In which of the following, viral load done by Real time PCR is of no role in investigation procedure? (AIIMS - May - 2018)

A. Person with hepatitis B on Tenofovir therapy
B. HSV causing temporal encephalitis
C. BK virus in patient of allograft renal transplant
D. CMV PCR in blood of patient of liver transplant

Important Information

- Tzanck smear • PCR is the IOC for whole α Virinae family



2. HHV-3

02:06:21

- Aka Varicella Zoster/Chicken Pox Virus
- Portal of entry: Respiratory tract / conjunctiva
- SAR (secondary attack Rate): >90%
- Infectivity period: 2 days before and 5 days after Rash (until the scab Falls)
- Characteristics of rash:
  - Centripetal Rash
  - Dew Drop Appearance on Rose petal
  - Pleomorphism seen
- IP: 14-16 days
- MC infectious complication: Secondary bacterial pneumonia (staph/strepto)
- MC extracutaneous involvement: Brain
- Most serious complication: Varicella Pneumonia

Congenital Varicella Syndrome

- Chorioretinitis
- Limb hypoplasia
- Microcephaly
- Cicatrizing lesions (scarring lesions)

Important Information

- If a pregnant female gets infected 5 days before EDD or within 2 days of delivery: Igs should be given to new born within 96 hrs of birth
- Vaccine strain for HHv-3: OKA strain

Lab Diagnosis

- PCR/ELISA For HHV-3
- Tzanck Smear

Previous Year's Questions

Q. A 5-year-old child came to OPD with fever, rashes on the body. There were rashes on axilla & flexor surface with various macule papule & vesicles. Most probable diagnosis is? (FMGE - Aug - 2020)

A. Measles
B. Chicken pox
C. Epstein Barr
D. Hand foot mouth disease

Previous Year's Questions

Q. Which vaccine is contraindicated in pregnancy? (NEET - Jan - 2019)

A. Hepatitis A
B. Hepatitis B
C. Rabies
D. Chicken pox

Herpes Zoster [Shingles]

02:17:42

- Reactivation of chicken pox
- Occurs in stress
- Types
  - HZ Ophthalmicus
    - → Single sided lesion present on Face
    - → Dermatomal involvement [T3-L3]
    - → Ophthalmic branch of Trigeminal nerve is involved

- HZ Oticus/Ramsay Hunt Syndrome
  - → Overnight Bell's palsy
  - → Vesicles over EAC, TM & cheeks
  - → Geniculate ganglion is involved
- Lab diagnosis: PCR / ELISA for Herpes zoster

Previous Year's Questions

Q. Herpes Zoster multiplies in? (NEET - Jan - 2019)

A. Peripheral nerve
B. Epithelium of skin
C. Dorsal root ganglion
D. Pharyngeal Epithelial cell

Previous Year's Questions

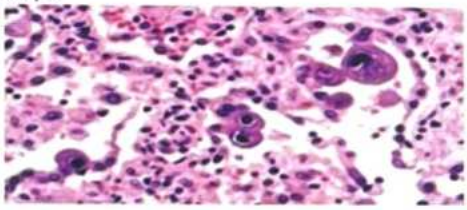
Q. Shingles is caused by which of the following? (NEET - Jan - 2018)

A. Varicella Zoster
B. Herpes Simplex
C. CMV
D. EBV

3. CMV (HHV-5)

02:25:24

- Largest Herpes virus
- Owl Eye Appearance of infected cell seen under microscope



Congenital CMV syndrome

- Features
  - Hepatosplenomegaly (mc feature)
  - Petechial lesions
  - Chorio retinitis
  - Calcifications
  - Convulsions
  - Microcephaly/ Macrocephaly
  - Mental retardation
- IOC: demonstration of CMV antigen on urine sample by ELISA / PCR (CMV DNA)

Mononucleosis like Syndrome

- Features
  - Atypical lymphocytes present [CD_8 T cells mainly]
  - Paul Bunnel Test: negative
  - Causes CMV Pneumonia in Post Kidney transplant patients(1-4 months post transplant are more vulnerable to develop CMV pneumonia)

CMV Retinitis

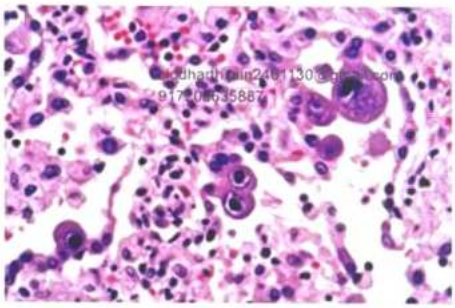
- Common in HIV positive patients when CD_4 count goes <50
- DOC: Ganciclovir

Lab Diagnosis of CMV

- Culture on Human Fibroblast cell line [WI-38]
- PCR: To demonstrate CMV DNA
- ELISA For CMV

Previous Year's Questions

Q. Owl eye inclusion bodies seen in? (FMGE - Dec - 2020)



A. HSV
B. HHV
C. EBV
D. CMV

Previous Year's Questions

Q. After kidney transplantation which organism infection is more likely to happen? (NEET - Jan - 2018)

A. CMV
B. EBV
C. Hepatitis virus
D. Polyoma virus

Previous Year's Questions

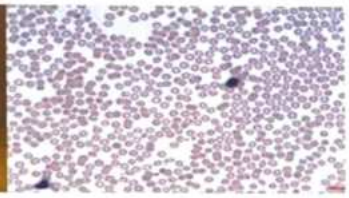
Q. MC Infection post solid organ transplantation? (AIIMS - Nov - 2019)

A. CMV
B. HSV
C. EBV
D. HPV

4. EBV / HHV-4

⏱ 02:33:51

- Affects B cells (d/t presence of CD21 / CR2) leads to Polyclonal proliferation of B cells because of which carcinomas can occur in EBV infected person
- Diseases
  - Nasopharyngeal Carcinoma [NPC]
  - Burkitt's lymphoma: Starry sky appearance
  - Hodgkin & Non Hodgkin's Lymphoma
  - Oral Hairy Leukoplakia [OHL]
  - Duncan Syndrome: X linked lympho proliferative syndrome
  - Tonsillar Carcinoma
  - Infectious Mononucleosis/Kissing Disease
    - → Common in male/female
    - → Presents with Fever, Rash, Lymphadenopathy
    - → CD_4: CD_8 Ratio reverses for sometime
    - → Atypical lymphocyte present: large size lymphocytes (CD_8 T-cell)
    - → Paul Bunnel test is positive

- Lab Diagnosis
  - PCR For EBNA (Epstein Barr Nucleic acid)
  - PCR For VCA (Viral Capsid Antigen)
  - ELISA For EBV

Previous Year's Questions

Q. Which of the following virus is from Herpes virus family? (FMGE - Jun - 2019)

A. Rubella
B. Measles
C. Rabies
D. EBV

Previous Year's Questions

Q. Oral hairy leukoplakia is caused by? (FMGE - Aug - 2020)

A. CMV
B. EBV
C. Kaposi Sarcoma
D. Varicella Zoster

5. HHV – 8

⏱ 02:41:12

- Aka Kaposi Sarcoma causing virus
- Common among HIV positive
- It is a Vascular tumor
- Also a/w Primary effusion lymphoma & Castle man's disease


Kaposi Sarcoma Lesions


Kaposi Sarcoma

Previous Year's Questions

Q. All of the following are associated with HHV 8 except? (AIIMS May 2018)

A. Kaposi Sarcoma
B. Primary effusion lymphoma
C. Castleman disease
D. T-cell leukaemia

6. HHV 6 & 7

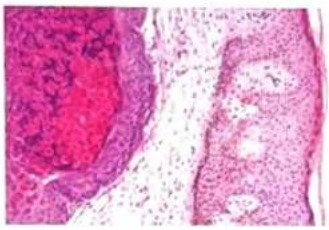
⏱ 02:43:00

- Causes Exanthem subitum: Nagayama spots
- Can ass with Focal Encephalitis

Pox Virus

02:44:02

- Molluscum Contagiosum
  - Caused by MCV-1, 2, 3 and 4
    - → MCV-1: M/c
    - → MCV-2: M/c in HIV positive
  - Clinical features
    - → Pearly white/ Greyish white / Pinkish white umblicated nodule over the skin/genitals present
  - Lab Diagnosis
    - → Tissue Biopsy: biopsy + stain shows Intra cytoplasmic Eosinophilic inclusion bodies/ Molluscum Bodies / Henderson Peterson bodies
  - Treatment: Podophyllin Resin + cryosurgery
- Vaccinia
  - Used to prepare Recombinant vaccine



Molluscum / Hp Bodies

Molluscum contagiosum

Previous Year's Questions

Q. A 22-year-old college boy with history for sex with commercial workers came to Derma OPD with lesion in genital region: tissue from this lesion was examined. which is the most likely finding?
(AIIMS - May - 2019)

A. Intracytoplasmic Vacuolation
B. Multinucleated giant cell
C. Belongs to Herpes family
D. It's an RNA virus

Previous Year's Questions

Q. Molluscum Contagiosum is a? (FMGE–June - 2019)

A. Adenovirus
B. Flavivirus
C. Rubivirus
D. Poxvirus

Parvo Virus

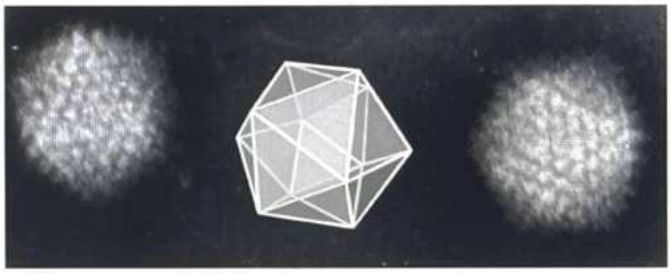
02:48:06

- Single stranded DNA virus
- Parvo B-19: Important serogroup
- Has an affinity towards Erythroid Precursor cells leading to aplastic crisis in Sickle cell anemic Patients
- Associated with
  - NIHF [Non Immune Hydrops Foetalis]
  - Polyarthalgia syndrome: Pain is more in upper limb joints than Lower limb joints
  - Erythema infectiosum /5th Disease: Slapped cheek appearance seen
- Lab diagnosis: PCR for Parvo B-19

Adeno Virus

02:51:49

- Double stranded DNA Virus
- Non enveloped virus
- Looks like space vehicle
- Type 3,4,7,14 causes: Pharyngo conjunctival fever or Swimming pool conjunctivitis
- Type 8,19,37 causes: Epidemic Kerato conjunctivitis
- Type 37 causes : STD
- Type 40,41 causes: Infantile diarrhea
- Type 11,21 causes: Hemorrhagic cystitis



- AAV (Adeno Associated Virus])& VACCINIA: Used for making recombinant vaccines using genetic engineering
  - Eg: sputnick (use human Adenovirus), Covishield (use chimpanzee Adeno virus)

Papovaviridae Family

02:57:18

- Double stranded DNA
  - Early region: Interacts with P_{53} & Rb genes
  - Late region: Capsid formation
- Non enveloped
- Papovaviridae consist of: Polyomavirinae and Papillomavirinae
  - Polyomavirinae consist of:

→ Polyoma virus
→ JC virus
→ BK virus
- Papillomavirinae consist of: HPV

1. Polyomaviridae

a. JC virus
- Cultured in human Fetal glial cells
- Infection: common
- Risk Factor: Immunosupression → JC virus crosses Blood Brain Barrier → Infects Oligodendrocytes & Astrocytes → Causes Subacute Demyelinating disease Progressive Multifocal Leukoencephalopathy (PML)
- Lab Diagnosis
  - PCR on brain biopsy tissue for JC virus
- No treatment, Fatal disease

b. BK Virus
- Post Kidney transplant infection [Borrowed Kidney]
- Causes BK nephropathy:
  - Decoy Cells are seen: Look like malignant cells
- Cultured in human diploid cell lines
- Lab Diagnosis
  - Culture: sample urine
  - PCR for BK virus

Decoy cell

c. SV – 40 [simian vacoulating – 40]
- Causes Progressive multifocal Leukoencephalopathy in Rhesus monkeys
- Cytopathic change: Cytoplasmic vacuolation

2. Papillomaviridae
- All are oncogenic
- Non – enveloped
- Icosahedral symmetry
- Double stranded DNA Genome (Circular)
  - Regulatory region:
    → Late region: L1 & L2 codes for late region which also codes for Capsid
    → Early region: E1, E2, E3, E4, E5, E6, E7
      – E6: Inactivates P53 gene
      – E7: Inactivates Rb gene

Infections caused by Papilloma virus 03:08:33
- Cause infections in the breaks of skin & mucous membrane, exposing the basement membrane
- In Cutaneous involvement
  - Low risk: 1, 2, 3
  - High risk: 5, 8
- In mucosal involvement:
  - Low risk: 6, 11
  - High risk: 31, 33, 16, 18

a. Skin Warts
- Most common types: Type 1 to 8
- HPV 5,8: Associated with Epidermodysplasia verruciformis / Tree Man syndrome

b. Genital Warts
- HPV 6,11: cause both Genital warts and Laryngeal warts
- Aka Condyloma accuminata

c. Carcinoma
- Females: Cervical Ca, CIN
- Males: Anogenital carcinoma [Penile, Anal]

Vaccines
- Cervarix (Bivalent vaccine): HPV 16,18
- Gardasil (Quadrivalent vaccine): HPV 6,11,16,18
- Gardasil -9: HPV 6,11,16,18,31,33,45,52,58
- These vaccines are prepared from L_1 region

Lab Diagnosis
- Histopathological examination shows:
  - Koilocytosis: cytoplasmic vacuolation along with Nuclear enlargement
- PCR For HPV DNA

Condyloma Accuminata

Previous Year's Questions

Q. Which sub-type of human papilloma virus has maximum chances of causing carcinoma cervix?
(FMGE June 2018)

A. HPV 16 & 18
B. HPV 6 & 11
C. HPV 31 & 32
D. HPV 1&2

Previous Year's Questions

Q. Blood from a woman at a local pregnancy clinic is analyzed for antibody titers. recently came in contact with nephew who is Varicella positive. A negative Varicella antibody titer in young woman signifies which of the following?

A. Suspectible to Zoster
B. Resistant to Zoster
C. Suspectible to Varicella
D. Resistant to chicken pox

Previous Year's Questions

Q. An otherwise healthy 65-year old male was in car accident and broke several ribs on the left side. Approximately 12 days later. he developed a painful. well-circumscribed vesicular rash over the left rib cage that persists for several weeks. The rash is most likely due to which of the following?

Ans: Herpes zoster

Previous Year's Questions

Q. A 42 year old male AIDS patient presents to the dermatology clinic for evaluation of skin lesions. He had numerous plaque-like lesions over his arms. chest. and neck. Oral examination revealed the lesion shown in the image on his palate. What is the most likely etiologic agent of these lesions?

A. Bartonella henselae
B. Human herpes virus B
C. HPV
D. Human T-cell leukemia virus

Ans: Option B

Diagnosis: Intra-oral Kaposi sarcoma

- Cause by Human herpes virus

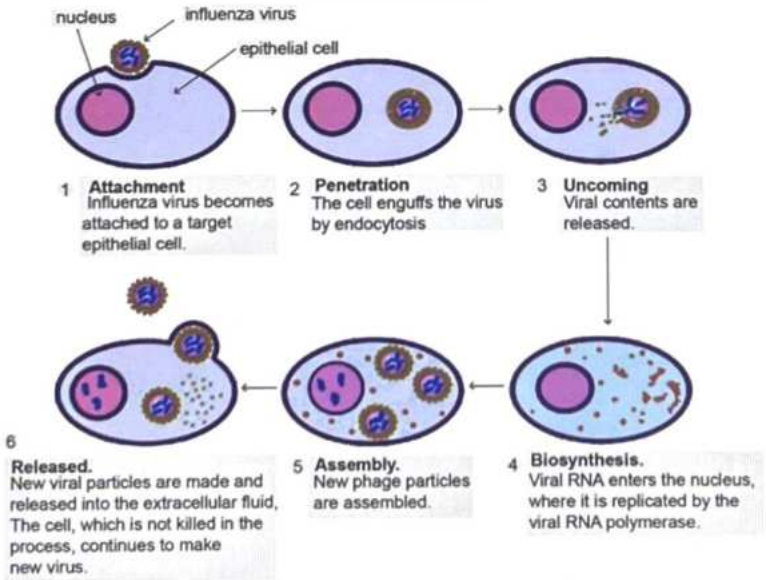
Previous Year's Questions

Q. A sexually active 17 year old man presents to the local free clinic to check some small papules that appeared on his penis. The papules are small. while lesions with a central depression. There is no discharge or pain on urination. What is the most likely causing these lesions?

A. Adenovirus
B. Coxsackievirus A
C. HPV type 6
D. Molluscipoxvirus

Diagnosis: Molluscum contagiosum

Figure 11.1



CLINICAL QUESTIONS

Q.1 A 40-year-old male who had a renal transplantation 2 months before developed a fever and came to your clinic for consultation. After a complete evaluation, bilateral diffuse interstitial pneumonitis was identified. Which of the following is the most likely etiological agent:

A. HSV
B. CMV
C. EBV
D. VZV

Solution:

- **CMV (Cytomegalovirus) is the most likely etiological agent.**

Option 1

Herpes simplex:

- **HSV 1 causes mainly oral herpes** in the form of **Herpes labialis (Refer to the image below)**
- HSV 2 causes genital Herpes.

Option 2

CMV is the most common viral pathogen complicating transplantation.

It most often manifests as pneumonitis in the first four months after transplantation.

It can also cause leukopenia in solid organ transplant recipients, obliterative bronchiolitis in lung transplants, and graft atherosclerosis after heart transplantation.

CMV (Cytomegalovirus) also causes:

- **Congenital CMV Syndrome:** hepatosplenomegaly, petechial lesions, chorio retinitis, calcification, microcephaly, mental Retardation.
- **Mononucleosis like syndrome**: Atypical lymphocytes [CD8 T cells mainly] and PAUL Bunnel Test negative.
- **CMV Retinitis**: CD4 count → <50 cells/mm^3

Option 3 **EBV (Epstein Barr Virus) causes:**

- Infectious mononucleosis
- Naso pharyngeal carcinoma
- Burkitts lymphoma
- Hodgkin's lymphoma / Non-Hodgkin lymphoma
- Tonsillar carcinoma
- Duncan syndrome.
- Oral hairy leukoplakia.

Option 4 **Varicella zoster** can cause chicken pox in primary exposure and Zoster or **Shingles on reactivation of primary lesion** in old age >60yrs.

Q.2 A 39-year-old homosexual HIV patient visited the OPD with complaints of visual disturbances. The patient was admitted and on evaluation, his CD4 count was 40 cells/mm^3. On Fundal examination, bilateral retinal exudates and perivascular hemorrhages were noted. On suspicion of viral etiology, microscopy of the sample was done in the lab. Which type of inclusion body is likely to be seen in this condition?

A. Intranuclear inclusion bodies
B. Intracytoplasmic inclusion bodies
C. Intranuclear + Intracytoplasmic inclusion bodies
D. None of the above

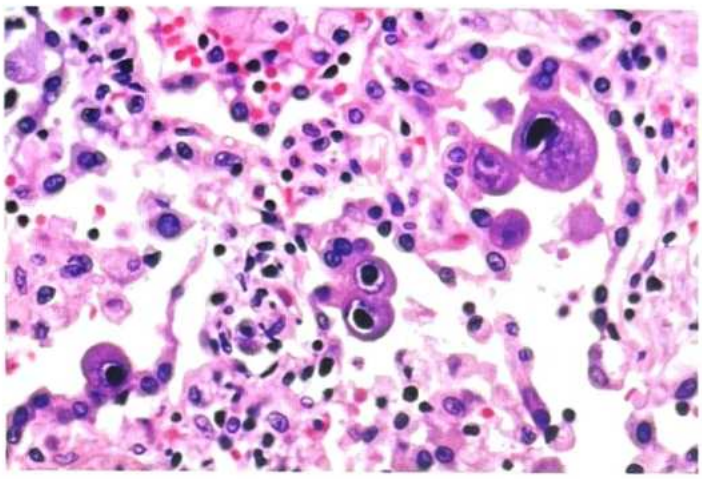
Solution:

- **Intranuclear and intracytoplasmic** inclusion bodies found in CMV infection.
- CD4 counts below which infections happen:

Cd4 count	Infection
< 500	TB
< 200	P. jiroveci
< 100	Cerebral Toxoplasmosis
< 50	CMV Retinitis M.A.C.

Features of CMV (Cytomegalovirus) :

- Largest herpes virus
- Characteristic features of the cell infected by **CMV: Owl eye appearance** (Image below).



CMV Causes:

- **Congenital CMV Syndrome:** hepatosplenomegaly, petechial lesions, chorioretinitis, calcification, microcephaly, mental Retardation.
- **Mononucleosis-like syndrome**: Atypical lymphocytes [CD8 T cells mainly] and PAUL Bunnel Test negative.
- **CMV Retinitis**: CD4 count → <50 cells/mm^3

LAB DIAGNOSIS of CMV:

- CULTURE on Human Fibroblast cell line (W1-38)
- PCR For CMV
- ELISA For CMV

12 VIROLOGY PART-2 (RNA VIRUSES)

RNA VIRUSES

Picornaviridae 00:00:18

- Includes small RNA Viruses
  - Polio Virus
  - Coxsackie virus
  - Entero virus
  - ECHO virus [Entero cytopathogenic Human orphan Virus]
  - Causes Aseptic meningitis
  - Rhino Virus: Common cold virus

1. Polio virus 00:01:29

- Types
  - Type 1: Most common wild type
  - Type 2: Most antigenic
  - Type 3: VAP [Vaccine associated Paralysis]
- Switch over: change of vaccines from Trivalent to Bivalent. Type 2 has been removed now
- Polio
  - Route: Feco oral contamination
  - >95%: Inapparent infections
  - <1 %: associated with paralysis (Anterior horn cells are damaged)
- Tonsillectomy should be avoided during a polio epidemic
- More physical activity should be avoided during polio epidemic
- Lab Diagnosis
  - Culture: stool sample cultured on Monkey Kidney cell lines
  - ELISA / PCR For Polio virus
- Antigens
  - C antigen (Coreless antigen)
  - D antigen (Dense antigen)

Previous Year's Questions

Q. Maternal antibodies do not provide protective immunity to neonate in? (NEET-Jan - 2020)

A. Diphtheria
B. Pertussis
C. Tetanus
D. Polio

2. Coxsackie Virus 00:07:00

- It is of 2 types: Coxsackie A and B

a. Coxsackie virus A

- Causes
  - Flaccid Paralysis in mice
  - Herpangina: Acute Vesicular pharyngitis
  - Acute Hemorrhagic Conjunctivitis: Cox A_{24} + Enterovirus – 70
  - HFMD (Hand Foot Mouth Disease): COX A_{16}
  - Aseptic meningitis

Hand, Foot & Mouth Disease

Herpangina

Previous Year's Questions

Q. An infant presented with the following lesions on his face & limbs. Which of the following is the most likely causative organism? (AIIMS–Nov - 2017)

A. HSV
B. HPV
C. Coxsackie
D. CMV

Previous Year's Questions

Q. Child present with oral & pharyngeal ulcerations & vesicular rashes of palms & soles which heal without Gusting. What could be the most probable diagnosis? (FMGE–Aug - 2020)

A. Measles
B. Hand foot mouth disease
C. Cutaneous drug reaction
D. Chickenpox

b. Coxsackie virusB

- Causes
  - Bornholm's disease (Acute Pleurodynia or Devil's grip)
  - DM (B4)
  - HFMD
  - Aseptic meningitis
- Lab Diagnosis
  - Culture: On New born Suckling mice (<48 hrs)
  - Arbovirus are also cultured on new born suckling mice

3. Enterovirus

- Enterovirus 68: Cause pneumonia
- Enterovirus 70: Causes Acute Hemorrhagic conjunctivitis
- Enterovirus 71: Causes HFMD
- Enterovirus 72: Hepatitis A

Previous Year's Questions

Q. Aseptic meningitis case fever of 4 days. with erythematous rash on chest with no h/o travel. tick bite. CSF findings – protein high. glucose normal. lymphocytosis monocytes 34%
(INICET–Nov - 2020)

A. Strep pneumoniae
B. Meningococci
C. West Nile
D. Enterovirus

Myxoviruses 00:14:04

1. Orthomyxoviridae	2. Paramyxoviridae
• Includes o Influenza A,B,C	• Includes o Para Influenza viruses o Mumps o Measles o Rubella (Togaviridae family, Arbovirus) o RSV o Metapneumo virus o Nipah virus

- Both are negative sense ssRNA virus

1. Orthomyxoviridae

- It is having a segmented genome
  - Influenza A: 8 segments of RNA
  - Influenza B: 8 segments of RNA
  - Influenza C: 7 segments of RNA

Important Information

- Other virus having segmented genome
  - B - Bunya virus
  - I - Influenza virus
  - R - Reo virus
  - A - Arena virus

- Important glycoprotein present in Orthomyxoviridae
  - Glycoprotein H (Hemagglutinin)
    → Helps in Adherence and Killing
  - Glycoprotein N (Neuraminidase)
    → Receptor Destroying Enzyme
    → Process is known as Elution

- Antigenic shift
  - Abrupt, drastic & discontinuous process
  - D/t Reassortment
  - Causes Pandemic
  - H3N2 changed to H1N1 (swine flu 2009)

- Antigenic drift
  - Single point mutation which is continuous in nature which firstly lead to Endemic then changes into Epidemic
  - H_3N_2 to $H_{3a}N_{2b}$

Refer Diagram 12.1

- H5N1 (bird flu)
  - Infected birds transmit infection to humans
  - Human to human transmission not seen
  - Carries high mortality
  - PB_1F_2 gene: Target the Mitochondria of cell & damages it

- H_1N_1 (swine flu)
  - Combination of Human influenza strain, Swine influenza strain, Bird flu strain
  - Common in Influenza A

  - Lab diagnosis: Nasopharyngeal swab
    → Taken with proper precautions
    → Extract RNA with RNA Extraction Kits and sent it for Real time PCR [<2 hrs]

  - Chemoprophylaxis: Tamiflu (Oseltamivir), given in initial stages of disease / during an Epidemic
  - Killed& live vaccines are available: Temporary immunity

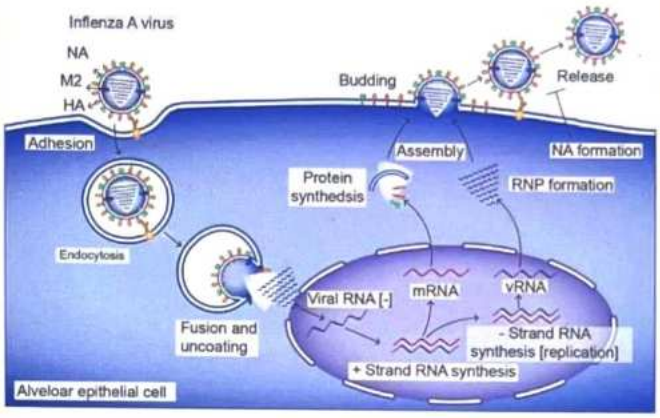
Previous Year's Questions

Q. Recent influenza-A pandemic was d/t?
(NEET Jan 2020)

A. H1N1
B. H5N1
C. H7N7
D. H3N3

Previous Year's Questions

Q. Identify the organism from the given life cycle?
(AIIMS May 2018)



A. Influenza A
B. Para-influenza
C. RSV
D. SARS virus

2. Paramyxoviridae 00:30:44

a. Para influenza virus

- Para influenza type 1 & 2 causes croup (Acute laryngotracheo bronchitis)
- Para influenza type 3 causes LRTI (Lower respiratory tract infection)

b. Mumps

- Mc presentation of Mumps: B/L Parotitis
- Atypical presentation: Meningoencephalitis can precede parotitis
- Complications
  - In young males: U/L Orchitis
  - In young Females: Oophoritis
  - Aseptic meningitis
  - Pancreatitis
- SAR: > 85%
- IP: 2-3 wks
- Lab diagnosis: PCR / ELISA for Mumps virus
- Vaccine strain: Jeryl Lynn Strain

Mumps

c. Measles 00:35:41

- Most characteristic Feature: Koplik Spot (opposite to lower 2nd molar)
- SAR: > 90%
- IP: 10-14 days
- Infectivity period: 4 days Before and 5 days after rash
- Up to 10th day of IP prodromal features come
- On 12th day of IP Koplik spot appears
- On 14th day of IP Rash appear
- Isolation period: from Prodromal features to 3rd day of rash

- Clinical features
  - Diarrhea
  - Pneumonia: Hecht's pneumonia aka Giant cell pneumonia is very common in HIV positive children
  - Sinusitis
  - Otitis media

- Chronic / Late complication: SSPE (sub sclerosing Pan encephalitis)
  - SSPE
    - → Schoolgoing child with H/o fever with rash during childhood presents with myoclonic seizure and on EEG there is a Burst pattern of waves indicate SSPE
    - → Diagnosis
      - IgG antibody to Measles virus on CSF examination
      - Biopsy
      - Radiological investigations can be done

- Lab diagnosis
  - PCR/ELISA For measles Virus
  - Warthin Finkelday giant cells: intracytoplasmic and intranuclear inclusion bodies

- Vaccine strain: Edmonston Jagreb
  - Vaccine is given by subcutaneous route (BCG: Intra dermal)
  - Diluent used: Distilled water (BCG: Normal saline)

Previous Year's Questions

Q. Warthin Finkeldey cells are seen in? (NEET Jan 2018)

A. Measles
B. Rubella
C. Rabies

Previous Year's Questions

Q. SSPE is a complication of? (FMGE Jun 2018)

A. Measles
B. Mumps
C. Rubella
D. Rabies

d. Rubella / German Measles 00:44:53

- Togaviridae Family & Arbovirus
- Infectivity period: 1 Week before and 1 week after Rash
- IP: 2-3 weeks
- Forcheeimer's Spots are seen over uvula
- Blue Berry Muffin Rash + Thrombocytopenia k/a Blue Berry muffin syndrome
- Congenital Rubella Syndrome
  - Classical triad
    - → **C** - Cataract
    - → **H** - Heart Disease (PDA > Pulmonary stenosis)
    - → **D** - Deafness (sensorineural)

How to remember

- CHD

- Lab diagnosis: PCR/ELISA For measles virus

Previous Year's Questions

Q. A neonate was found to have cataract, deafness & cardiac defects. Which group of viruses does the mother was infected with? (NEET–Jan - 2019)

A. Togaviridae
B. Flaviviridae
C. Bunyaviridae
D. Arenaviridae

Previous Year's Questions

Q. True about congenital rubella syndrome is? (NEET–Jan - 2019)

A. Blueberry Muffin rash is seen
B. Virus can be isolated only up to 6 months after birth
C. Triad of CRS are cataract, cardiac defects, cerebral palsy
D. Infection is most serious after 5 months of pregnancy

e. RSV (Respiratory Synctial virus)

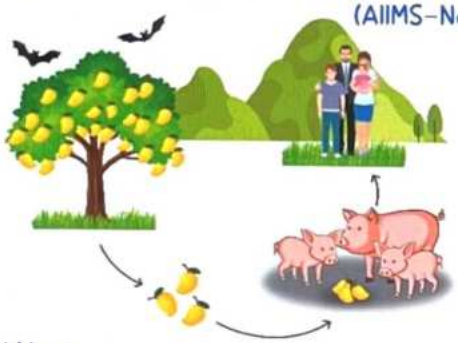
- Causes Acute Broncheolitis
- DOC: Ribavirin
- Age group: 1 month to 6 months
- Fusion proteins present aka F protein: Responsible for multinucleate Giant cell formation
- Lab diagnosis: PCR/ELISA For RSV

f. Nipah virus 00:50:12

- Subfamily: Henipavirinae
- Reservoir: Fruit Bats
- Amplifiers:Pigs
- MOT: contamination of food and water with urine of fruit bats or secretions of infected patients
- Mortality: > 70%
- IOC: RT PCR

Previous Year's Questions

Q. Which virus transmission shown in picture?
(AIIMS–Nov - 2018)



A. NipahVirus
B. Zika Virus
C. Ebola Virus
D. Corona Virus

Rabies Virus 00:54:17

- Family: Rhabdoviridae
- Sub Family: Lyssa Virinae
- SS RNA Virus: Negative sense changes to Positive sense with the help of RNA Polymerase
- Enveloped virus
- Shape: Bullet shape
- Speed of Rabies virus: 3 mm/hr
- Decrease in Ach at various sites & induce neural cell apoptosis
- IP: Months to years

- Types:
  - Natural / street virus
  - Fixed or dumb virus

Natural / street virus	Fixed / dumb virus
• Causes natural / furious rabies in 80% of cases and 20% develop Dumb rabies • Hydrophobia present • Negri bodies can be demonstrated	• Causes dumb rabies • Used in vaccine production • Hydrophobia and Negri bodies will be Absent

Types of bites

- Class-1 bites
  - Licks over intact skin
  - Management: clean the surface

- Class-2 bites
  - Licks over broken skin
  - Slight grazing present
  - No oozing of blood
  - Management: post exposure prophylaxis

- Class-3 bites
  - Facial Bites
  - Constant oozing of blood after bite
  - Untraceable animal bite
  - Wild animal bites
  - Management:
    → HRIg (20 IU/ Kg) / ERIg (40 IU/Kg) + Post Exposure Prophylaxis

Post exposure prophylaxis

- Essen Regime: 0,3,7,14,28 (I/M)
- Thaired regime: 2, 2, 2, 0, 2 (I/D)

Lab diagnosis

- Anti- mortem diagnosis
  - Done by taking up hair follicles from nape of neck or corneal smear
  - Direct Immuno fluorescence testing (antigen demonstration) is done on above sample: Rabies antigen is demonstrated
  - PCR for rabies virus
  - Antibody demonstration in CSF and Blood by indirect Immuno fluorescence testing
  - Animal inoculation

- Post mortem diagnosis
  - Demonstrations of Negri bodies in hippocampus
    → Brain Biopsy tissue + Basic fuchsin: Seller's technique
    → Negri bodies (intra cytoplasmic, Eosinophilic inclusion bodies) can be demonstrated
  - By animal inoculation

Previous Year's Questions

Q. Virus with high mortality rate?
(INICET–Nov - 2020)

A. Ebola (50%)
B. Influenza HINI (1-4%)
C. Rabies (100%)
D. Nipah (40-75%)

Previous Year's Questions

Q. Antemortem diagnosis of rabies is made with?
(FMGE–June - 2019)

A. Rabies virus specific antibodies
B. Inoculation in culture media
C. Negri bodies in hippocampus
D. Corneal impression smear

Hepatitis Virus

01:18:14

Refer Table 12.1

1. Hepatitis A

- Only hepatitis virus that can be cultured
- Lab diagnosis: ELISA
  - IgM anti HAV demonstration in
    - → Stool examination
    - → Serum examination (more diagnostic)

2. Hepatitis B

- Incomplete double stranded DNA virus
- Exists in 3 forms
  - Spherical: most common
  - Tubular
  - Dane particle: complete virus genome (42 nm)

- Genes
  - P gene codes for: Polymerase, Reverse transcriptase (largest gene)
  - C gene codes for: Precore and Core
  - S gene codes for: Surface Antigen
  - X gene codes for: Carcinogenicity

- Diagnosis
  - Acute Hep B
    - → HBs Ag positivity + IgM anti HBC
    - → IgM anti HBC: more diagnostic of acute Hep B
    - → Along with above 2 if HBe Ag +ve:
      - Indicate active Replicating phase
      - Maximum liver damage occurs
    - → Anti HBe +ve: Active replicative phase starts going down
  - Chronic Hep B
    - → 6 months HBS Ag +ve
    - → IgG anti HBc +ve
  - If vaccinate
    - → Anti HBs +ve
    - → Anti HBs should be > 10 IU / ml
    - → Hep B vaccine
      - Recombinant vaccine
      - Prepared in yeast
    - → Schedule → 0, 1, 6 months

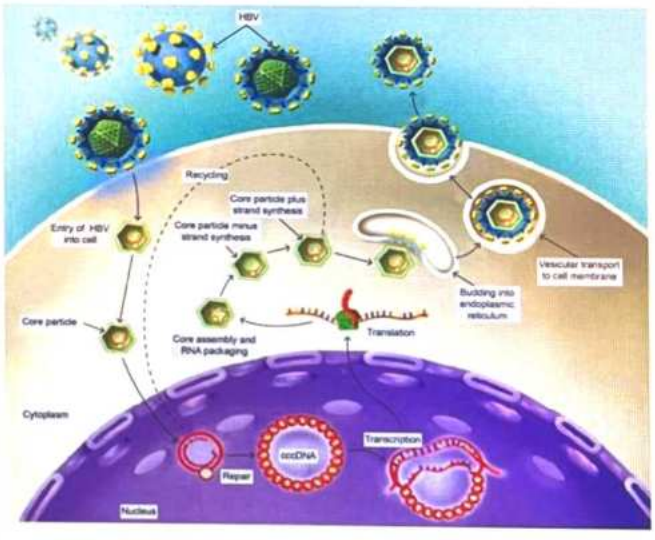
Hepatitis B virion

Refer Diagram 12.2

- Pre core mutants: lack HBe Ag & carry poor prognosis
- Escape mutants: No HBs Ag
- Lab Diagnosis
  - IOC: PCR For HBV DNA (series of PCR done)
  - Hepacard Test For HBs Ag

Previous Year's Questions

Q. The following diagram show life cycle of?
(AIIMS – Nov - 2017)



A. HIV
B. Hep B
C. Influenza virus
D. Rabies

Previous Year's Questions

Q. Which is not true about hepatitis B viruses?
(FMGE – Aug - 2020)

A. DNA viruses
B. Transmitted by focal-oral route
C. Can be transmitted from mother to child (Perinatal transmission)
D. Contains reverse transcriptase

3. Hepatitis C

01:48:26

- No Good vaccine available d/t Quasi Species (Antigenic variations)
- 85% Chronicity (Maximum): a/w more with
  - Cirrhosis
  - Hepatocellular carcinoma
- Lab Diagnosis
  - IOC: HCV RNA PCR
  - ELISA: IgM anti HCV can be demonstrated
  - Hepatitis C tridot Testing

Previous Year's Questions

Q. Cryoglobulinemia is associated with?
(NEET – Jan - 2018)

A. Hepatitis A
B. Hepatitis B
C. Hepatitis C
D. Hepatitis D

4. Hepatitis D

- Exist in 2 forms
  - Co infection: Hep B+ Hep D
    → When Hep B and Hep D co exist it will reduce each other's virulence
  - Super infections: Hep D infection happens over the already present Hep B
    → More dangerous
    → 5-20% Fulminant
    → Lab diagnosis: ELISA For IgM anti HDV

5. Hepatitis E

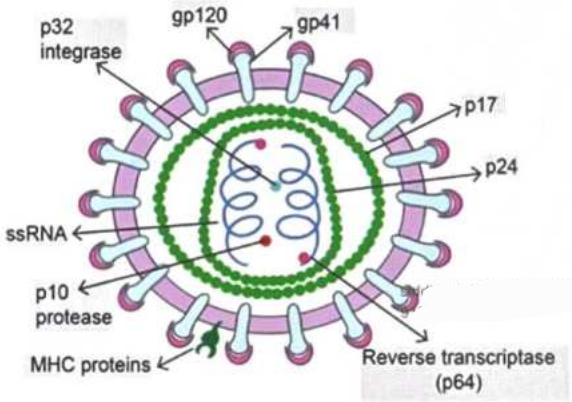
- Feco-oral transmission
- Fulminancy
  - 1-2% in normal condition
  - Increase to 20% in pregnancy
- Lab diagnosis: ELISA For IgM anti HEV (serum > stool)

HIV

01:55:43

- Family: Retroviridae
- Sub Family: Lentivirinae
- Prevalencein India:
  - HIV-1 is More common than HIV-2
  - M/c variant in HIV-1: group 'M', sub type 'c'
- Structural genes of HIV-1
  - Gag: Codes for core (p24) as well as matrix protein (p17, p18)
  - Env: Codes for Envelope (Gp 120 and Gp 41)
  - Pol: Codes for polymerase and reverse transcriptase
- Non structural genes
  - Tat: Transcriptional activator
  - Rev: Regulator
  - Nef: Negative factor gene
  - Vif: Viral infectivity factor gene
  - Vpu: promotes CD4 degradation
    → Important entity of HIV-1
  - Vpr: Help in transport of viral genome
  - Vpx: Specific to HIV-2
  - LTR: Provide promoter, enhancer and integration signals

HIV virus



HIV Transmission

- Route
  - Sexual: 01-1% (mc in India)
  - Blood Transfusion: > 90%
  - Needle stick injury: 0.5 -1% (0.3-3%)
  - Mother to child: 30%
- After entry
  - HIV virus affect the CD4 T-cell and Macrophages via Co-receptors
    → Co-receptor for
    – CD4 T-cell: CXCR4
    – For Macrophage: CCR5

- Stages
  - Initially Acute HIV syndrome (Mononucleosis like syndrome) happens which includesFever and LAP
    - → It takes around 10 years for HIV +ve to change into AIDS disease
    - → Following infection HIV gets carried to lymph nodes and other lymphoid tissues
    - → After destroying CD4 T-cells it spills over into blood stream which presents as mononucleosis like syndrome
      - Initially Flu like illness
      - After 3 to 6 weeks of infection there is a significant drop in CD4 T-cell
  - Asymptomatic stage: Aka Clinical latency
    - → It is due to adequate immune response of the body against HIV Infection
    - → Happens within 1 month
    - → Both CMI and Humoral immunity prevents further destruction of T-cells because of which the viremia gets decreased
    - → This clinical latency can continue for up to 10 years
  - PGL: Persistent generalized Lymphadenopathy
    - → It is enlarged LN of >1 cm in 2 or more contiguous sites which persist for >3months
    - → 25 to 30% of the infected people develop PGL
  - Symptomatic HIV disease: aka AIDS related complex
    - → After variable period of clinical latency the CD4 T-cells start decreasing and patients start developing opportunistic infections
    - → Signs and symptoms include
      - Unexplained weight loss (>10%), fatigue
      - Unexplained diarrhea for >10months
      - Oral thrush
  - AIDS
    - → There will be a rapid fall in CD4 T-cells (<200 cells/ µL)
    - → High viral load present
    - → Lymphoid tissue is destroyed

Previous Year's Questions

Q. Approximate time interval between HIV infection and manifestation of AIDS is? (NEET Jan 2018)

A. 7.5 years
B. 10 years
C. 12 years
D. 11 years

OPPORTUNISTIC INFECTIONS

CDC AIDS case definition with/ without HIV serology

02:19:02

- Candida: cause infection in Esophagus, Trachea, Bronchus
- Cryptococcus neoformans
  - M/c cause of meningitis in HIV +ve
  - DOC: Fluconazole
  - In Cryptococcal meningitis: LAMB + Flucytosine
- Cryptosporidium parvum: Cause chronic diarrhea which last for > 1 months
- CMV colitis/ esophagitis: Serpiginousulcers are produces
- Cerebral toxoplasmosis
  - Causes seizures in HIV +ve
  - Treatment
    - → For prevention: Cotrimoxazole
    - → With encephalitis: pyrimethamine + sulphadiazine + Leucovori
- CNS lymphoma: NHL
  - On NCCT
    - → Cerebral edema
    - → Seizures, BBB gets damaged
    - → Basal ganglia involved
    - → Eccentric dot sign

Important Information

- **To confirm AIDS, HIV serology should be +ve for:**
  - **Coccididomycosis**
  - **TB**
  - **Extrapulmonary TB**

- P.jerovecii: when CD4 <200
  - B/L Perihilar shadows on X-Ray
  - O_2 saturation less
  - BAL is taken and GMS
  - Treatment: Cotrimoxazole
- Progressive PML
  - Done by JC virus
  - Subcortical white matter lesion seen in parietal and temporal lobe
  - CT shows Hyper intense lesion on CT (T2 image)
- HSV esophagitis
  - Produces Mucocutaneous ulcer > 1month
  - Punched out ulcers seen
  - Painful

- Kaposi sarcoma
  - In HIV +ve
  - Age <60 years
  - Cause by HHV-8
  - DD: Bacillary Angiomatosis (vascular tumour) done by Bartonella hensela
  - Reddish brown lesions on body which bleeds on touch

- M.A.I (Mycobacterium Avium Intracellulare)
  - CD4 <50
  - Treatment: Clarithromycin + ethambutol + Rifabutin

Cd4 count	Opportunistic infection
• CD4 <500	• TB
• CD4<200	• P. jerovecii
• CD4<100	• Cerebral toxo
• CD4<50	• CMV retinitis • MAC

- For Verizella zoster infection in HIV positivity: VZ immunoglobin should be administered to prevent fulminant infection
- For Histoplasma: Itraconazole is DOC
- For salmonella: Ciprofloxacin
- Bartonella: Doxycyclin
- CMV: vancyclovir, Ganciclovir, Acyclovir
- Person with TB having HIV positivity: First treat TB then HIV after 2 weeks

Respiratory disease in AIDS patient 02:39:37

- Organism: S. pneumoniae > Mycobacterium TB > P. jerovecii
- Perihilar opacities: PJP
- Consolidation patch: Pneumococcus pneumonia
- Multiple snowstorm appearance / hazy appearance: Miliary TB
- Fever, shortness of breath, chest pain, weight loss, on CXR Perihilar opacities, LDH increased and β 1,3 glucan test +ve, DLCO decrease → PJP

MTB with HIV 02:43:24

- More serious
- When MTB co exist with HIV positivity the cavity size tend to be smaller so it disseminate more easily into the blood stream
- First start with ATT then CART after 2 week
- For Latent TB infection: INH + $VitB_6$ for 9 months
- For MAI (CD4< 50): B/L infiltrate may mimic MTB
- IOC: culture from BAL
- Treatment: REC (Rifampicin + Ethambutol + Clarithromycin)

Important Information

- Invasive Aspergillosis is not seen in HIV +ve patients

CVS involvement

- Associate with MI,
- Cardiomyopathy is common

GIT involvement

- Oropharyngeal Candidiasis
- Odynophagia in AIDS

CD4 T cell count	Opportunistic infections
• 400 – 200	• Mycobacterium TB • Candida (oral Thrush) • Herpes zoster • Kaposi sarcoma • Hairy Leukoplakia • Herpes simplex
• Below 200	• **P** - **P**neumocystis carinii • **C** - **C**ryptococcus neoformans **C**occidioidomycosis **C**ryposporidiosis • **T** - **T**oxoplasma gondii
• Below 50	• MAC [mycobacterium Avium Intracellulare] • CMV retinitis o DOC: Ganciclovir

Lab Diagnosis

- Most sensitive test: ELISA
- Rapid / simple test: <30 min

- Supplemental test: antibody detection
  - Western blot
  - Immunoblot assay

- Confirmatory test
  - Most confirmatory: HIV RNA PCR
    → Can detect 40 copies
  - For Pediatric HIV: HIV DNA PCR

- Non specific test
  - Low CD4 T-cell count: measured with the help of flow cytometry

- Detection of Hypergamma globulinemia: in this we measure
  - → β_2 macroglobulin
  - → Neopterin
- Altered CD4: CD8 ratio

ELISA 02:55:37

- 1st gen ELISA: Crude antigens
- 2nd gen ELISA: Synthetic antigens
- 3rd gen ELISA
  - Synthetic Peptides (IgM and IgG)
  - It reduce the window period to 22 days
- 4th gen ELIAS
  - Can measure IgM and IgG antibody
  - Can estimate P24 antigen by using Recombinant/ synthetic peptide

Previous Year's Questions

Q. Approximate time interval between HIV infection and manifestation of AIDS is? (NEET – Jan - 2018)

A. 7.5 years
B. 10 years
C. 12 years
D. 11 years

Western Blot Test

- Works on the principle of Immunoblot technique
- Find out Ab in serum against specific antigenic fragments
- Ab against
  - Gag gene component: P24, P17, P18
  - Envelope gene component: Gp120, Gp41, Gp160
  - Pol gene component: P68, P53, P31
- According to WHO criteria, western blot is said to be +ve when:
  - At least 2 envelope bands (Gp120, Gp160, Gp41) with or without Gag/Pol bands
- As per CDC: presence of any 2 bands (P24, Gp120, Gp41, Gp160) then it's said to be +ve
- P24 antigen is 1st to come (12 to 26 days)

Latest Post exposure prophylaxis of HIV 03:05:32

- Start < 2 hours / maximum <72 hrs of exposure
- If exposed person is an Adolescent and adult (>10 years) and >30 kg weight give
  - Fixed dose combination of:Tenofovir + Lamivudine + Dolutegravir
- In Children ≥6 yrs, ≥20kg weight
  - Fixed dose combination of: Zidovudine + Lamivudine + Dolutegravir (50 mg)
- In children <6 years, <20 kg weight
  - Fixed dose combination of: Zidovudine + Lamivudine + Lopinavir / Ritonavir

ARBO Viruses 03:09:01

- **Togaviridae**
  - Chickungunya
  - Eastern Equine Encephalitis virus
  - Western Equine Encephalitis virus
- **Flaviviridae**
  - JE virus
  - Dengue
  - Yellow Fever
  - KFD virus
  - Zika virus
  - Hep C
  - West Nile fever
- **Bunyaviridae**
  - Can be further classified into:
    - → Bunya virus
      - California encephalitis
    - → Phlebo virus
      - Sand Fly Fever virus
      - Rift valley Fever virus
    - → Nairovirus
      - Crimean Congo Hemorrhagic virus
      - Ganjam virus
    - → Hanta virus: associated with 2 syndrome (associated with rodent Fecal material)
      - Hantaan renal syndrome
      - Hantaan pulmonary syndrome
- **Reoviridae**
  - Contains Orbivirus: associated with Colorado Tick fever
- **Rhabdoviridae**
  - Contains Vesiculo virus
    - → Chandipura virus
    - → Vesicular stomatitis virus

Previous Year's Questions

Q. Viral haemorrhagic fever in India caused by virus? (AIIMS–Nov - 201[illegible])

A. Ebola
B. Yellow virus
C. Crimean Congo
D. Marburg

Incubation Periods

- Dengue : 5-6 days
- Chickungunya: 5-6 days
- Japanese encephalitis: 5-15 days
- Yellow Fever: 3-6 days
- Kyasanaur Forest Disease: 4-8 days

Japanese Encephalitis 03:18:53

- Reservoir: Ardeid birds [white cranes]
- Amplifiers: Pigs
- Transmitters: Culex tritaeniorhyncus, Culex Vishnui (in India)
- Mortality: 30 %

Previous Year's Questions

Q. Which of the following is not true about JE?
(NEET–Jan - 2020)

A. Human is reservoir
B. Pig is amplifier host
C. Transmitted by culex vishnui
D. Vaccination available

Dengue 03:22:28

- Serotypes: 1,2,3,4,5
  - Most dangerous: serotype 2
- Transmitted by: Aedes aegypticus
- ADE [Anti body Dependent Enhancement]: Serotype 1 infected 1st time & recovered→Gets infected with Serotype 2 →Leads to increase chance of
  - DHF [Dengue Hemorrhagic fever]
  - DSS [Dengue shock syndrome]
- DHF
  - Fever: Acute, high & continuous
  - Tourniqet test Positive: > 20 Petechial lesions / Sq. inch
  - Thrombocytopenia: platelet count < 1lakh
  - Haemoconcentration: Haematocrit raises by 20%
- DSS
  - DHS + Weak & rapid pulse
  - Narrow pulse pressure observed
  - Management: IV fluids + antibiotics
- Break Bone Fever occurs in Dengue
- Lab Diagnosis
  - NS-1 antigen testing
    - → Done in the initial 2 days
    - → NS: Non Structural or Non Specific protein
    - → Done by ELISHA
    - → Up to 5th day: IgM capture ELISA can be done
  - PCR For Dengue virus & serotypes

Yellow Fever 03:32:40

- 1st viral disease to be discovered
- Not present in India
- Prevented by Yellow Fever vaccination
- 17 D vaccine is used
- YF vaccine can be given to a pregnant female
- Transmitted by Aedes aegypticus
- Aedes aegypticus index should be <1 around seaports and airports
- Vaccination certificate is valid For life time
- Presents with fever, jaundice, mild hemorrhagic features
- IOC: PCR

KFD

- Aka monkey fever
- Reservoirs: Squirrels / Rats
- Amplifiers: monkeys
- Transmitter: Hard Tick
- Prevalent in Karnataka
- Mild hemorrhagic Fever

Chikungunya 03:37:33

- Means Bending upon
- Immense joint pain present
- Never complicates
- Chik sign: Hyperpigmentation
- IOC: PCR for chikungunya virus

Transmission

Aedes	Culex
Y - Yellow Fever	B - Bancroftian Filariasis
D - Dengue	E - Encephalitis Japanese
R - Rift valley Fever	W - West Nile Fever
C - Chikungunya	A - Arthritis like Features

How to remember

- Aedes: YDRC
- Culex: BEWA

Filoviridae 03:41:31

- Important species: Marburg and Ebola virus

Ebola virus

- Filamentous in Shape
- MOT: Contact with the secretion of infected patients
- Most virulent: Zaire Ebola
- Least virulent: Ivory coast

- IP: 3-15 Days
- BSL 4 [Bio safety level 4] is maintained
- IOC: RTPCR For Ebola virus

Previous Year's Questions

Q. What is not true regarding Zika Virus?
(AIIMS – Nov - 2018)

A. Belong to family Flaviviridae
B. Transmission happens by mosquitoes
C. Not transmitted from mother to new born
D. Possibility can cause microcephaly

Arena Viridae 03:44:21

- Segmented genome is present
- Sandy appearance on electron microscopy
- Important Viruses
  - Lymphocytic chorio meningitis virus
  - Lassa Virus
  - Junin Virus
  - Machupo virus

Reo Viridae 03:45:30

- DS RNA Virus
- Have segmented genome
- Important Viruses
  - Rota Virus
  - Orbi virus
  - Colti Virus

Rota Virus 03:46:21

- On Electron Microscopy: Wheel with spoke appearance
- MC cause of diarrhea in children
- IOC: Rota Virus antigen detection in stool by ELISA
- Vaccines
  - Rotateq
  - Rotavarix
- NSP – 4: Responsible for secretory diarrhea

Previous Year's Questions

Q. A 6-month child presented with diarrhoea and vomiting for three days. Which of the following enterotoxin is most likely responsible for this condition? (NEET – Jan - 2019)

A. NSP4
B. NSP6
C. VP3
D. VP7

Virus associated with Gastroenteritis

- Enteric Adenovirus Type 40, 41: infantile diarrhea
- Norwalk Virus
- Calci virus
- Astro virus

Oncogenec Viruses 03:49:53

DNA viruses

- Papova viridae
  - HPV: Genital warts
- Herpes viridae
  - EBV [HHV-4]: causes
    - → Nasopharyngeal Ca
    - → Burkitt's lymphoma
    - → Hodgkin and Non Hodgkin lymphoma
  - HHV 8: Kaposi sarcoma
  - Hep B: HCC

RNA viruses

- Retro Viridae: HTLV 1 (Adult T cell leukemia)
- Flavi Viridae: HCC

Zika Virus 03:52:10

- Family: Flaviviridae
- Transmitted by: Aedes mosquito
- Also have Sexual Transmission
- Mother to child transmission with the help of Hofbauer cells are seen
- If pregnant mother infected then the new born can develop
  - Microcephaly
  - GBS [Guillain Barre Syndrome]
- IOC: RTPCR for Zika

Previous Year's Questions

Q. What is not true regarding Zika Virus?
(AIIMS – Nov - 2018)

A. Belong to family Flaviviridae
B. Transmission happens by mosquitoes
C. Not transmitted from mother to new born
D. Possibility can cause microcephaly

Prions

- Proteinaceous infectious particles
- Most resistant form
- Killed or Denatured at 135°C for 1hr
- Mechanism of virulence: Abnormal Folding (PrP→PrPSC)
- Human diseases
  - K - Kuru
  - F - Familial Fatal insomnia

- C - CJD
- Gaye - Gerstman Strassler Schienker syndrome

How to remember

- KFC Gaye

- Animal diseases
  - Scrapie disease of goat
  - Bovine spongiform encephalopathy
  - Wasting disease of deer

Viral Haemorrhagic Fever Causing Viruses 03:58:26

1. **Family Arena Viridae**
- Lassa Virus
- Junin virus
- Machupo virus

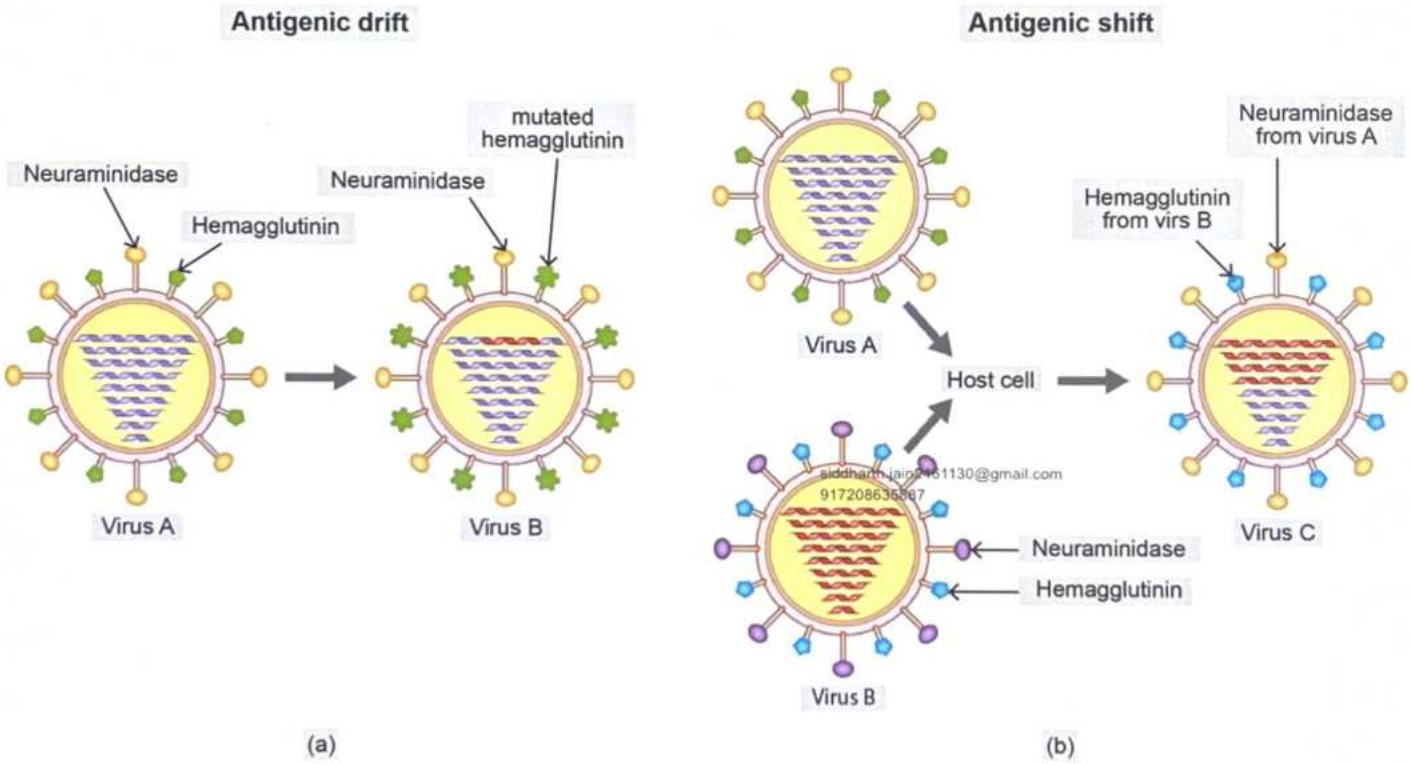
2. **Family Bunyaviridae**
- Hantaan Virus
- Rift valley Fever

3. **Family Filoviridae**
- Ebola virus
- Marburg virus

4. **Family Flaviviridae**
- Dengue
- Yellow Fever and
- Tick borne encephalitis
- KFD

5. **Family Rhabdoviridae**
- Chandipura virus
- VS virus

Diagram 12.1

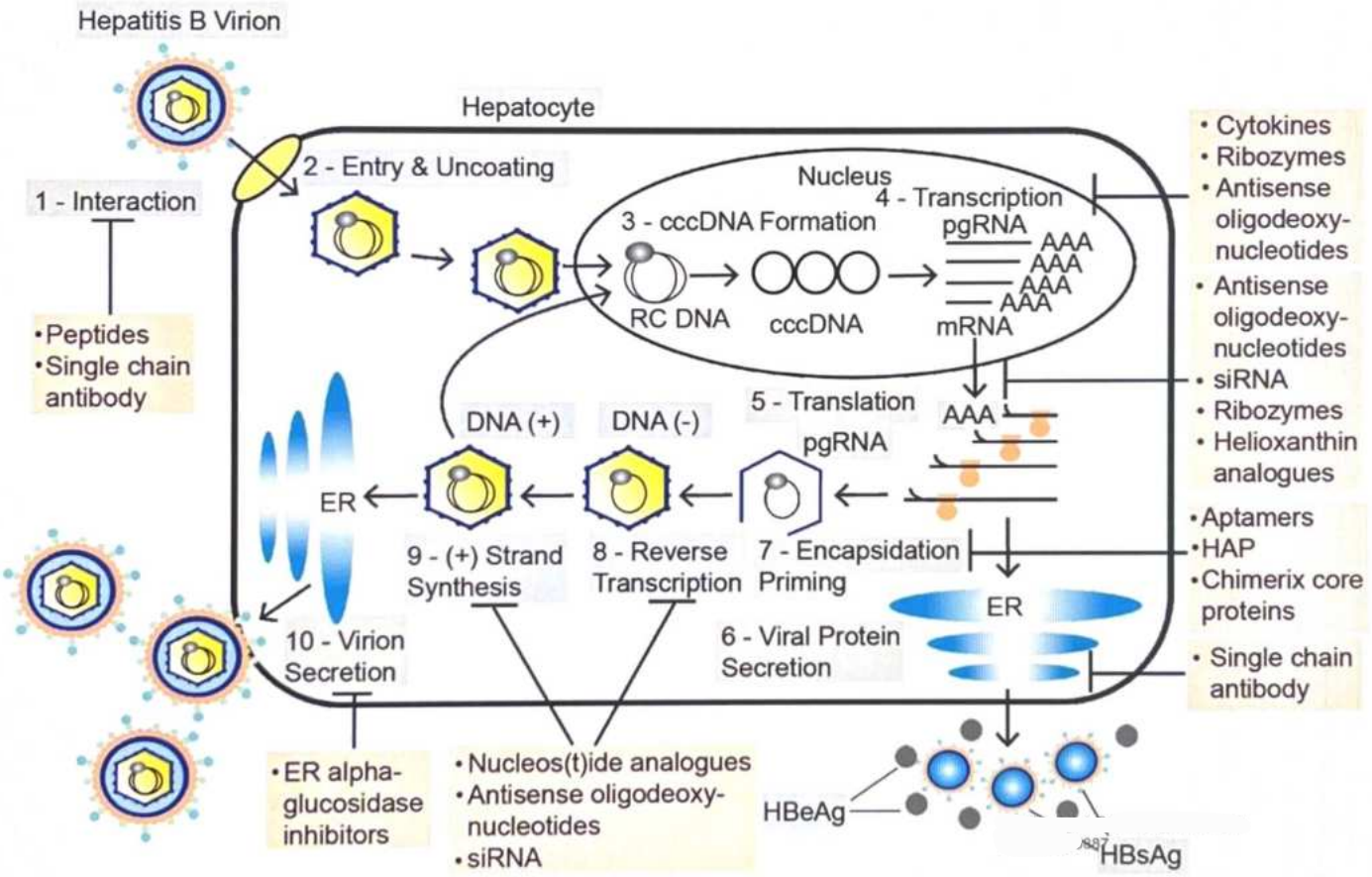


Antigenic shift & drift

Table 12.1

Hep A	Hep B	Hep C	Hep D	Hep E
• Picornaviridae	• Hepadenaviridae • only DNA virus	• Flaviviridae	• Dependent over B	• Calciviridae
• Feco oral transmission	• Parenteral, sexual & vertical transmission	• Parenteral		• Feco oral transmission
• IP: 15-50days	• IP: 50-150days	• IP: 15-150days	• IP: 50-150days	• IP: 15-50days
• Fulminancy 0.1% (adults)	• 0.1%-1%	• 0.1%	• 5-20%	• 1-2% →20% (pregnancy)
	• Chronicity 1-10%	• Chronicity >85%		
	• Carrier 01-30%	• Carrier 1.5-3.2%		

Diagram 12.2



CLINICAL QUESTIONS

Q.1 A forest officer with high-grade fever, chills, intense frontal headache, severe myalgia and body aches visited the OPD. The doctor susupected it as a haemorrhagic fever. Which of the following is true about the given condition?

A. Transmitted by tick
B. It is arboviral infection
C. Also known as monkey disease
D. All of the above.

Solution:

- All of the above are true of KFD (Kyasanur forest disease)
- **Kyasanur forest disease**: KFD is an example of tick-borne hemorrhagic fever. It was first reported in Kyasanur forest of Karnataka, Shimoga district, n 1957.
- **Vector: Hard ticks** (Haemaphysalis spinigera).

Hosts:

- **Reservoirs** are the **rats and squirrels.**
- **Amplifier hosts** are the monkeys (KFD is known as **Monkey's disease**).
- Man is an incidental host and considered as dead end.
- **Clinical Manifestation**: Incubation period varies from 3–8 days. First stage (hemorrhagic fever) occurs followed by second phase of meningoencephalitis.
- **Killed KFD vaccine**: It is recommended in endemic areas of Karnataka (all villages within 5 km of endemic foci).

Q.2 A patient with chills, sore throat, runny nose, cough and body aches visited to the OPD. On further testing, single stranded RNA virus with segmented genome was found to be the causative agent. Which of the following strain is associated with the given condition?

A. H1N1
B. H5N1
C. H2N2
D. H3N2

Solution:

- Swine flu is due to **H1N1.**

Structure of Influenza virus:

- Single stranded RNA
- Have **segmented** genome

- Surface proteins:
  - Glycoprotein H [Hemagglutinin] → adheres to cell
  - Glycoprotein N [neuraminidase] → Receptor Destroying Enzyme. The process is known as **elution.**
- **Transmission:** from person to person primarily in droplets released by sneezing and coughing.
- **Antigenic variations**:
  - The surface antigens **HA and NA show variations** and are primarily **responsible for antigenic variations** exhibited by influenza viruses.
  - Antigenic variations are of two types: antigenic **shift** and antigenic **drift.**

Option 1

The 2009 flu pandemic or swine flu was an influenza pandemic caused by H1N1 and the second of the two pandemics involving H1N1 influenza virus (the first was the 1918 for pandemic.)

H1N1 2009 Flu:

- Started in April 2009.
- Pandemic – affected the whole world including India.
- It is made by reassortment of 4 strains (1 Human + 2 Swine + 1 Avian Strain) in pigs.
- Human to human transmission seen – hence more infectious.
- H1N1 was less virulent (as it lacks PB1F2 protein gene) compared to H5N1 therefore it had more morbidity but less mortality.
- Diagnosis – by RT PCR detecting Hemagglutinin and Neuraminidase genes.

Option 2

H5N1: Avian influenza or **bird flu.**

- Infected birds transmit to humans.
- Human to human transmission not possible.
- **$PBIF_2$ gene** → target the mitochondria of cell & damages it.

Option 3

H2N2: 1957 pandemic (**"Asian Flu"**).

Option 4

H3N2: 1968 pandemic

Q.3 A patient visited the hospital with non-specific symptoms like fever, mild cough, rashes, etc. On further examination and testing, the physician is suspecting it to be an arboviral infection. Which of the following is true regarding arboviral infections?

A. Hantavirus pulmonary syndrome is caused by inhalation of rodent feces
B. KFD is caused by the bite of wild animal
C. Lyssa virus is transmitted by ticks
D. Yellow fever is transmitted by Anopheles mosquito

Solution:

- All the above are false statements **except** Hantavirus pulmonary syndrome is caused by inhalation of rodent feces.

Option 1: Hantaviruses:

- Found worldwide.
- Cause two serious and often fatal human diseases:

- **Hemorrhagic fever with renal syndrome**
- **Hantavirus pulmonary syndrome (HPS).**

- **Rodents are the reservoirs** for hantaviruses.
- The virus infections in rodents are lifelong and without deleterious effects.
- Transmission to humans occurs by inhaling **aerosols** of rodent excreta **(urine, feces, saliva)**.
- **Hantavirus pulmonary syndrome** is generally severe, with reported mortality rates of 30% or greater.
- The disease begins with fever, headache, and myalgia followed by rapidly progressive pulmonary edema, often leading to severe respiratory compromise.

Option 2, 3: KFD (Kyasanur forest disease) is **transmitted** to humans **through** the bite of infected hard **ticks.**

Option 4: Yellow fever is transmitted by Aedes mosquito.

13 COVID-19

- Aka Wuhan Virus
- Caused by SARS COV 2
- Family: Coronaviridae
- Genus: Beta Coronovirus (SARS COV 2)
- Suborder: Cornidovirinae
- Order: Nidovirales
- Realm: Riboviria
  - Gp. of viruses having RNA dependent RNA Polymerase (RDRP)

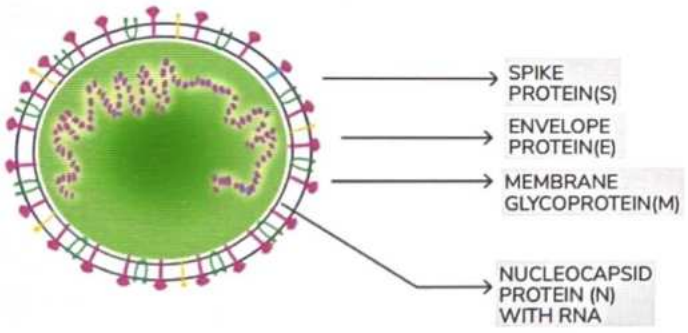
Morphology of SARS COV2

00:02:25

- Virion: Spherical & Enveloped
- Diameter: 100-150 nm
- Most important feature: Club shaped Spike projections (coded by S Gene)
- EM (Electron Microscopy): Crown like appearance (CORONA)

Genome

- Non – segmented, Positive sense ssRNA
- Large genome (30 kbs)
- Contains genes coding for
  - Structural proteins
  - Envelope: coded by E gene
  - Nucleocapsid protein: coded by N gene
  - Membrane Glycoproteins: coded by M gene
  - Accessory proteins



Mode of Transmission

00:07:54

- Spreads through infected droplets
- Aerosol spread (more efficient route of transmission)
- Contact transmission
- Spread through (yet not proved)
  - Stool contamination
  - Plasma
  - Serum
- Maximum virus shedding occurs 5-8 hrs before onset of symptoms
- Virus can be cultured& can be isolated for next 6-8 days
- Concentrations of SARS Cov 2 RNA in upper respiratory specimens have shown decline after onset of symptoms
- For Mild to Moderate COVID-19 patients RCV (Replication Competent Virus) has not been recovered after 10 days of symptom onset
- For severe COVID-19 infection patients, 95% of patient's specimens no longer yield RCV after 14 days
- Patient can be considered non infectious after 2 weeks of symptoms onset

Pathogenesis

00:15:51

- Virus enters upper respiratory tract (URI) & binds to specific receptors
- In the Initial Asymptomatic phase (1-2 days) Virus binds to ACE 2 receptor present in human airway with the help of Receptor binding domain of Spike protein
- Spike protein binds to the receptors and leads to Conformational change & fusion of Virus & Host cell membrane
- Following this Type II Transmembrane Serine protease (TMPR SS2) present on surface of Host cell clears these ACE 2 receptors & further activate the Spike glycoprotein of SARS Cov 2
- Because of this there is a Conformational change which helps in Virus entry
- After virus entry Uncoating of Virus happens which release Nucleic acid in cytoplasm and further Replication & Translation in Nuclei
- Upon complication of Machinery this Virus gets assembled in cytoplasm & release by Exocytosis
- Secondary targets: Depend upon ACE 2 receptor numbers
- Max number of ACE II receptor are present in
  - Lungs (Type II Pneumocytes)
  - Gut
  - Heart & kidney

Clinical Features

- 80% of infected person develop mild disease
- 20% of infected person develop moderate to severe disease

- Out of this 20%
  - 15%: Respond well to O_2 therapy
  - 5%: May need Ventilatory support

Symptoms in COVID-19 00:25:58

Refer Table 13.1

- Day 5: Dyspnea
- Day 7: Hospitalization
- Day 6-9: Cytokine storm
  - Release of Interleukins, TNF
  - Mx by Tocilizumab
- Day 9: Patient develops ARDS if not managed properly

Lab diagnosis 00:35:35

- Gold standard test: Real time (Reverse transcriptase) PCR
- Direct tests
  - Antigen detection
  - RTPCR
  - Gene xpert (CBNAAT)
  - True NAT
- Indirect test
  - Antibody detection kits
- Supplementary test
  - CXR
  - HRCT
  - Other lab tests: Eg: Look for Interleukin levels in case of Cytokine storm

Previous Year's Questions

Q. ...biology testing for COV-2 depends upon? (AIIMS – June - 2020)

A. Viral load
B. Time of incubation
C. Past infection & asymptomatic patient
D. Currently infected asymptomatic

RTPCR

- Take 2 swabs
  - 1 Nasopharyngeal swab (NP swab)
  - 1 Oropharyngeal swab (OP swab)
- Nasopharyngeal swab is better than Oropharyngeal

Steps to take NP swab

- Ask the Pt. to take off the mask to take NP swab
- Remove swab from packaging
- Tilt the patient's head back slightly
- Insert swab along Nasal septum, just above floor of Nasal passage to the Nasopharynx until resistance is felt
- Gently Rub & Roll the swab, leave swab in place for several seconds to absorb secretions
- Remove swab slowly while rotating

Steps to take OP swab

- Insert swab into Posterior pharynx & Tonsillar areas
- Rub swab over both Tonsillar pillars & Posterior Oro-pharynx

Viral Transport Media (VTM)

- Eagle minimum essential medium (commercially available)

- VTM can be easily prepared using
  - 10gm Veal infusion broth + 2gm Bovine Albumin fraction V
  - Add into sterile distilled water (400ml)
  - Add 0.8 ml Gentamycin sulfate solution (50 mg/ml) & 3.2 ml Amphotericin B (250 µg/ml) to avoid Bacterial & Fungal contamination
  - Sterilize by Filtration

Genes targeted in RTPCR

- Screening genes (Semi confirmatory genes)
  - Nucleocapsid
  - Envelope
  - Spike proteins
- Confirmatory genes
  - ORF 1a: Open reading frames 1a
  - RDRP: RNA dependent RNA Polymerase
- On RTPCR

• E Gene → +ve • RDRP Gene →-ve	• Not COVID 19
• E Gene → +ve • RdRP Gene → +ve	• COVID 19 +ve
• E Gene →+ve • ORF 1a Gene →+ve	• COVID 19 +ve

RTPCR vs TrueNAT

Refer Table 13.2

Cycle Threshold Value (CT VALUE)

- Relative viral RNA estimation
- CT value depends on PCR kits
  - CT value is Inversely proportional to Viral RNA levels
  - CT value 16: suggests high viral RNA
  - CT value30: suggest less viral RNA

Immuno Chromatographic Test (ICT)

- Can estimate Antigen or Antibody
- Various card tests work on Immuno chromatographic principle
- Rapid methord

Masks

- N_{95} is Integral point of PPE

• N - Not resistant to oil	• R -Resistant to oil	• P - oil proof
• Types: N_{95}, N_{99}, N_{100}	• Types: R_{95}, R_{99}, R_{100}	• Types: P_{95}, P_{99}, P_{100}

- 95 indicates:95% of Airborne particles filtered
- 99 indicates: 99% of Airborne particles filtered
- 100 indicates: 100% of Airborne particles filtered
- HEPA Filter: High Efficiency Particulate Air Filter
  - Filter 99.97% of Airborne particles

Basic structure of N_{95} mask 01:03:29

- Outer layer: Spun bond
  - Made up of Non-woven Polypropylene which blocks droplets & water
- 2nd & 4th layer
  - Made up of Melt blown Non-woven fabric
  - Protective & Holding layer for 3rd layer (PTFE)
- 3rd layer
  - Made up of PTFE (Polytetra flouro ethylene)
  - Main filter

N_{95} MASK

R_{95} MASK

P_{95} MASK

HEPA FILTER MASK

Filter efficiency tests

- Sodium Chloride Aerosol Challenge Test
- Dioctyl phthalate challenge test

Uses of N_{95} 01:08:12

- Single use (Recommended)
- Decontamination & Reuse of N_{95} mask (as per CDC protocol)
  - Vaporized H_2O_2 (Best method)
    - → Time of decontamination: 10 min
    - → Mask can be used for 3 cycles
  - UV light (260-285 nm)
    - → Time of decontamination: 60 min
    - → Mask can be used for 3 cycles
  - Autoclave
    - → Time of decontamination: 121°C x 15 min
    - → Mask can be used for 2-10 cycles
  - Dry heat (Hot air oven)
    - → Time of decontamination:70°C x 1 hr
    - → Mask can be used for 2 cycles
  - Air out (Most commonly done)
    - → Time of decontamination: 72 hrs
    - → Mask can be used for 5 cycles

Drugs used for COVID 19 01:13:00

1. Inhibit Viral RNA Polymerase (RNA synthesis)
- Includes
  - Remdesivir
  - Favipravir
- MOA
  - Adenosine/ Guanosine Nucleotide Analogue
  - RdRP inhibitor

2. Inhibitors of Viral protein synthesis
- Lopinavir
- Ritonavir

3. Viral entry inhibitors
- Hydroxychloroquine
  - Increases endosomal pH & inhibits fusion of SARS COV 2 & Host cell membrane
  - Also have immunomodulatory action

4. Immunomodulator
- Ivermectin
  - Inhibits the Nuclear Symport (Import) of Host & Viral proteins through inhibition of Importin-1 receptors
- Azithromycin
  - Acts by Induction of Interferon which stimulate genes that attenuate Viral Replication
  - Enhance Neutrophill activation
  - Attenuation of Inflammatory cytokines mainly IL-6 & IL-8
- Hydroxychloroquine
  - Increases endosomal pH & inhibits fusion of SARS COV 2 & Host cell membrane

Important Information

- Drugs known to prevent/ decrease mortality in COVID 19 patients → Steroids

Previous Year's Questions

Q. Bio safety levels required for propagative work dealing with SARS-COV 2 vaccine is?
(AIIMS - June - 2020)

A. BSL-2
B. BSL-4
C. BSL-1
D. BSL-3

Table 13.1

Systemic symptoms	Prevalence	Respiratory symptoms	Prevalence	Miscellaneous symptoms	Prevalence
Fever	75-81%	Cough	54-60%	Chest pain and tightness	4-10%
Fatigue	27-35%	Dyspnea	19-28%	Otalgia	1-11%
Myalgia	14-19%	Wheeze	9-26%	Diarrhea	8-12%
Rigors	13-22%	Hemoptysis	1-2%	Nausea	3-10%
		Loss of smell (Hyposmia)	4-55%	Abdominal pain	2-7%
		Sore throat	10-14%	Vomiting	2-8%
		Nasal congestion	3-7%	Headache	10-16%
		Rhinorrhoea	5-12%	Confusion	7-15%
		Pneumonia		Ataxia	Rare
		Respiratory failure		Conjunctivitis	1-4%
				Opthalmoplegia	3-6%

Table 13.2

Factors	RTPCR	TrueNAT
• Sensitivity	• 70-80%	• 70%
• Specificity	• 95%	• 90-95%
• Genes targeted	• Screening genes ○ N, S, and E genes • Confirmatory genes ○ ORF 1a, RdRP	• Screening genes ○ E gene • Confirmatory genes ○ RdRP
• Copies detection	• Can detect even 10 copies/ml	• Detects 486 copies/ml
• TAT	• 4-6 hrs	• 60-90 min
• Type of system	• Open system	• Cartridge based test
• Cost	• Cost effective than TrueNAT	• Expensive kits

LEARNING OBJECTIVES

UNIT 6: IMMUNOLOGY

- Immunology
  - Innate and acquired immunity
  - Active and passive immunity
  - Antigen
  - Antibodies: property, types
  - Antigen antibody interactions, Precipitation reaction
  - Agglutination, Card test, neutralization reaction, compliment fixation test
  - ELISA, Immunofluorescence
  - Hypersensitivity
  - Compliment system
  - Structure of immune system
  - Graft, Graft vs host reaction
  - Autoimmunity and Immunodeficiency disorders

14 IMMUNOLOGY PART-1

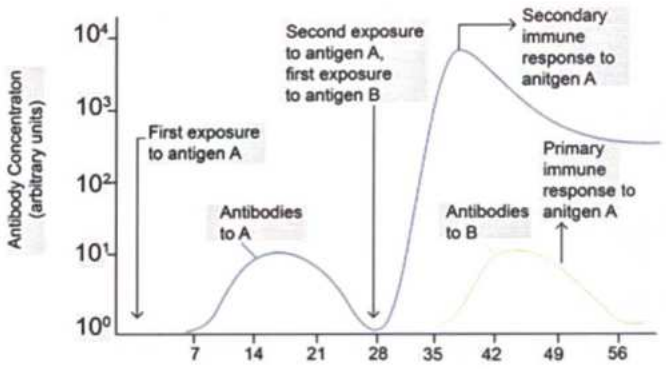
- IMMUNITY → Resistance offered by the host against offenders 00:01:51

Innate immunity	Acquired immunity
• Resistance to the infection from birth by its Genetic or constitutional make up	• Resistance to infection acquires during life time
• Depends on o S - Species specific o I - Individual specific o R - Race specific	• Different Forms o Natural Active: Infections o Natural Passive: mother to child o Artificial Active: vaccines o Artificial Passive: Immunoglobulins
• Components o Phagocytes (monocytes, macro phages, neutrophils) o NK (natural Killer) cells o Alternate complement pathway o Acute Phase Proteins (CRP, MBP Serum amyloid protein) o Normal resident Flora o Inflammation, Fever o Skin & mucosal barrier	• Components o B cell o T cell o Classical complement pathway o Antigen presenting cells [APC]

Active immunity	Passive immunity
• Produced actively by host immune system	• Received passively
• Induced by infection or immunogen	• Induced by readymade antibody
• Long lasting	• Short
• Lag period present	• No lag period
• Memory present	• No memory
• Booster dose useful	• Subsequent dose – less effective
• Negative phase may occur	• No negative phase
• In immunodeficiency: not useful	• Useful

00:17:45

PIR [Primary Immune Response]	SIR [Secondary Immune Response]
• Slow to start, Sluggish, Short lived	• Prompt, Powerful, Prolongs
• IgM produced mostly	• IgM change to Ig G k/a Isotype/Class Switching • Occurs d/t change in the constant portion of heavy chain



Antigen 00:12:28

- Any Foreign substance which induces antibody formation under optimum conditions
- HAPTEN
  - Incomplete antigen
  - Requires carrier protein to be immunogenic

Factors affecting immunogenicity

- Nature of the substance
- More Molecular weight & size: more immunogenic

- Tissue enzyme response
- Foreignness
  - More foreign then more immunogenicity
  - More foreign then more Hypersensitivity
- Nature of substance
  - Proteins > carbohydrates > Lipids > Nucleic Acids

Heterophile antigen

- Two different families share the same antigen
- E.g
  - In Weil-felix test: Test between OX-19, 2, and K which are proteus antigens. These proteus antigen share the Rickettsial antigen due to which they cross react with Rickettsial antibodies. Eg OX-K is strongly positive for scrub typhus
  - Paul Bunnel Test: done to diagnose infectious mononucleosis (caused by EBV)

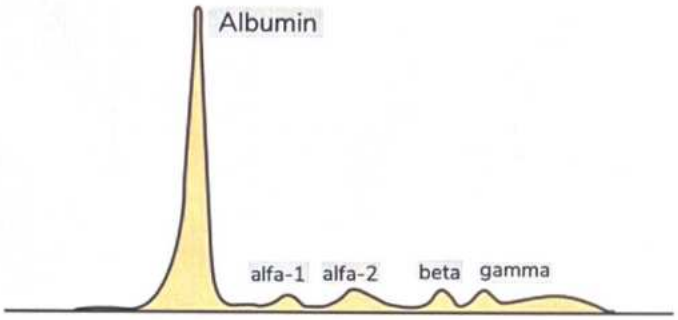
Super Antigens 00:28:17

- Non MHC restricted
- Attached to TCR on Lateral side on Vβdomain, which Induces > 20% T lymphocytes activation→Leads to massive Lymphokine release→ Shock
- Examples
  - Staphylococcal Toxins
    - → Enterotoxin A: Food Poisoning
    - → Exfoliate Toxin A, B: SSSS [Staphylococcal skin scalded syndrome]
    - → Entero Toxin F: Toxic shock syndrome Toxin
  - Streptococcus: scarlet Fever/Erythrogenic toxin A & C / Pyrogenic toxin
  - Mycoplasma arthritis
  - Malasezzia Furfur
  - Yersinia enterocolitica
  - EBV, CMV, HIV, Rabies virus

T-Independent Antigen	T-Dependent Antigen
• Simple antigens like: o LPS o Capsule o Flagella	• Complex
• Dose dependent	• Immunogenic over wide range of dose
• IgM &IgG3 produced more	• All Ab rise
• No memory	• Memory +nt
• No macrophage processing	• Macrophage processing needed
• Directly acts on B cells & causes polyclonal B cells activation	• Involves the CMI and Humoral immunity comes into action

ANTIBODIES 00:39:01

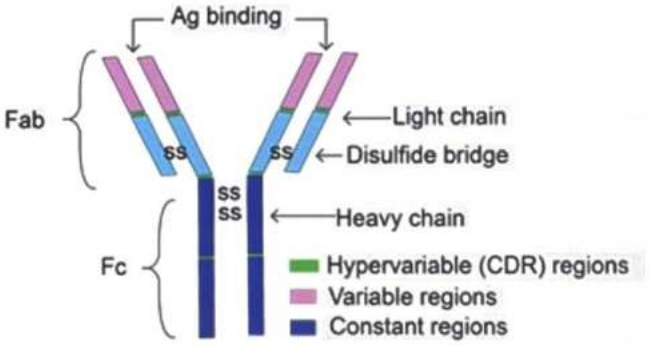
- Antibodies are γ-globulins
- Electrophoretic mobility was discovered by: Kabat & Tiselius
- Helps to differentiate between various forms of Immunoglobulins

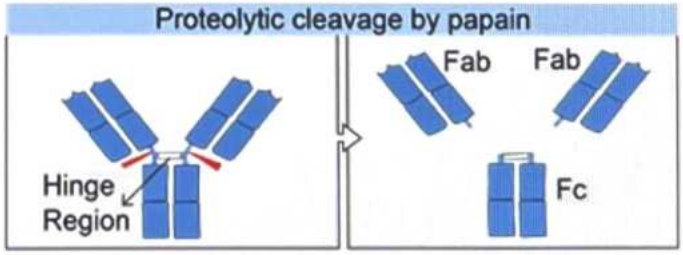


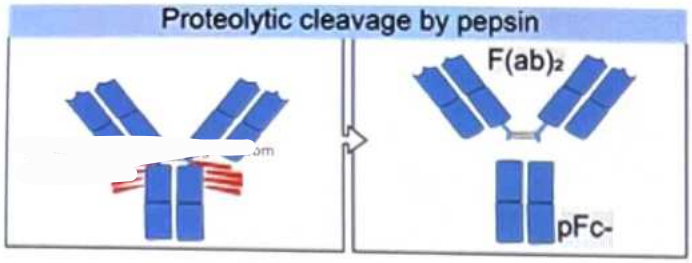
Normal Electrophoretic Graph & Blood Proteins

Antibody Structure 00:40:20

- Any antibody consists of 2 Light chains and 2 heavy chain
- Light chain and heavy chain are bonded together by disulfide bond
- 2 Heavy chains are bonded by disulfide bond
- One light chain have 1 variable & 1 constant
- One heavy chain have 1 variable & 3 or 4 constant
- Upper portion: Fab [ab = antigen binding]
- Lower portion: Fc [c= constant portion]
- CDR: present in both light and heavy chain, in the Hyper variable region aka Paratope
- Epitope of an Antigen attaches to Paratope of an Antibody







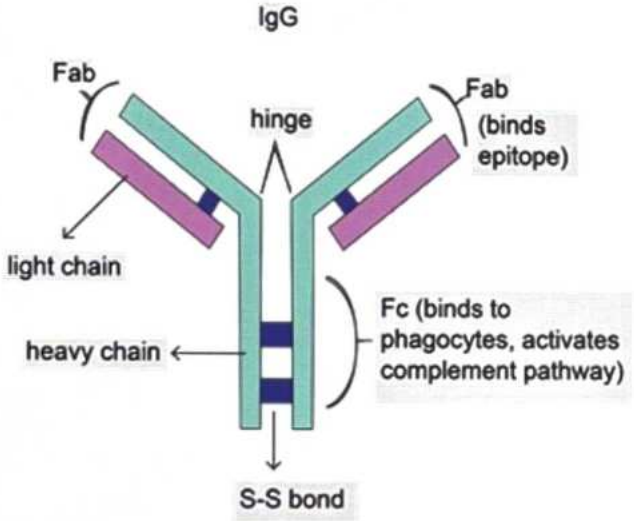
Gerald M. Edelman

Rodney R. Porter

- Proteolytic cleavage is done by both papain and pepsin
  - Papain cleaves above the hinge region: 3 Fragments (2Fab + 1 Fc) released
  - Pepsin cleaves Below the hinge region: 2 Fragments [$F(ab)_2$ + Fc1] released

Properties of Antibodies

IgG



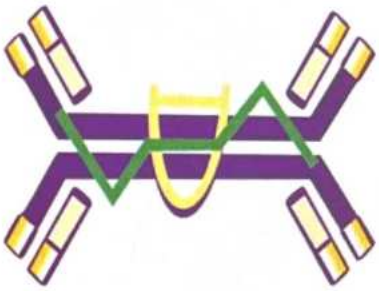
- Maximum in concentration
- Maximum t1/2: 23 Days
- Sub Types: Ig G_1, G_2, G_3, G_4
  - M/c: IgG_1
- Mainly involved in precipitation reaction
- Involved in classical compliment pathway
  - IgG_4 does not play any role in classical compliment pathway
- IgG_3 does not involve in co-agglutination
- IgG_2 does not cross placenta

Previous Year's Questions

Q. A child is suffering from recurrent chronic infection with encapsulated bacteria. Which subclass of IgG does the child has deficiency?
(NEET Jan 2019)

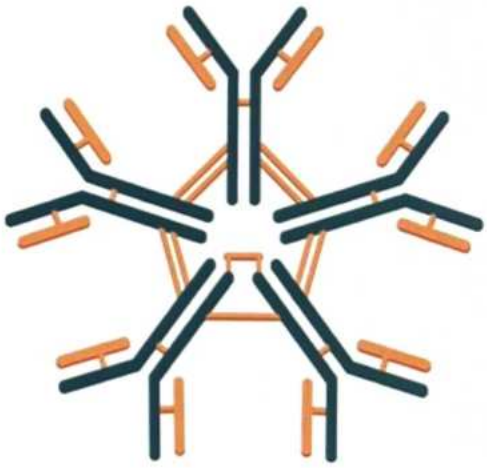
A. IgG1
B. IgG2
C. IgG3
D. IgG4

IgA



- IgA1: present in serum
- IgA2: present in mucosal surface & body secretions
- Dimer Form bounded by Joining chains
- Mc selective Ig deficiency: Selective IgA

IgM



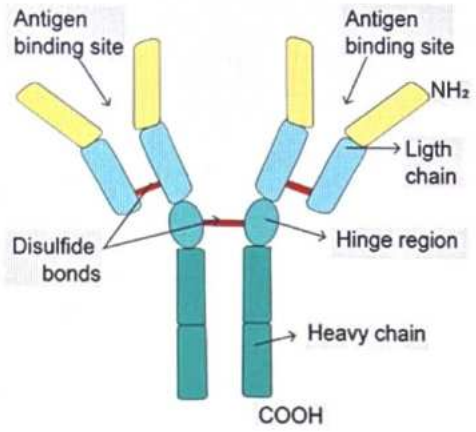
- Valency: 10 (Pentamer)
- Maximum molecular weight
- Has maximum sedimentation co-efficient
- 1st Ig to be produced in any immune response
- Involved in classical compliment pathway
- They are having 'j'chains aka joining chains

Previous Year's Questions

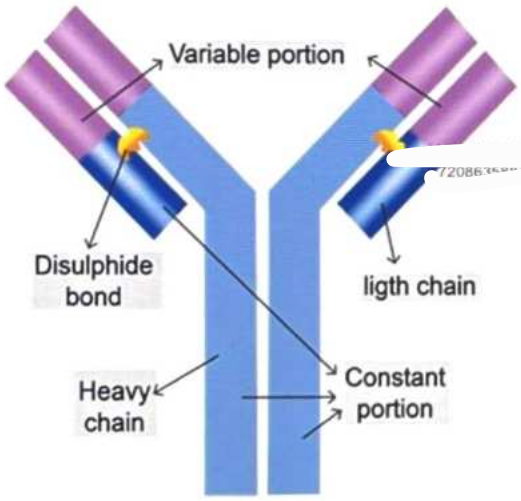
Q. 10-year-old child Jaundice &diarrhea reported to hospital and within same period of time, many children from same school presented with same symptom. Which of the following is most probable finding? (AIIMS June 2020)

A. IgM antibody hepatitis A
B. IgG antibody hepatitis B
C. IgM antibody hepatitis B
D. IgG antibody hepatitis A

IgD



IgE



- Lowest concentration in the body
- Shortest t1/2:< 2 Days
- Shows a particular phenomenon K/a Homocytotropism (movement towards mast cells). This property is being used in Type 1 Hypersensitivity
- Only heat labile antibody

Previous Year's Questions

Q. In chronic allergy which Ig are more persistent in the body? (AIIMS – May - 2019)

A. IgG
B. IgE
C. IgA
D. IgM

Light Chains

00:58:32

- Every Immunoglobulins contain 2 light chains which can be kappa : lambda (2:1)
- In Multiple myeloma: IgG light chains comes in urine
- In Waldenstrom macroglobulinemia: IgM light chains comes out in urine
- Naive B cells: Over their surface IgM and IgD are present and that's why IgM is the 1st Ig to get released in any immune response

Antigen Antibody Interactions

01:01:38

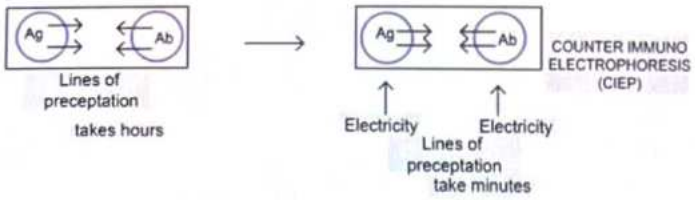
- Reaction happens in 3 zones
  - Prozone
    - → Antibody excess
    - → Prozone phenomenon is seen in standard agglutination test for the brucellosis in which Prozone is removed by dilution
  - Zone Of Equivalence
    - → Antigen and Antibody in equal concentration
    - → Lattice is being produced
  - Post Zone: Antigen Excess
- Most reactions happen in Zone of Equivalence
- Affinity:intensity of attraction between the antigen & antibody
- Avidity: strength of bond after the formation of antigen & antibody complex

Precipitation Reaction

- Soluble antigen + Antibody produce insoluble precipitate
  - Eg. Ascoli's thermo-precipitation test for Anthrax antigen

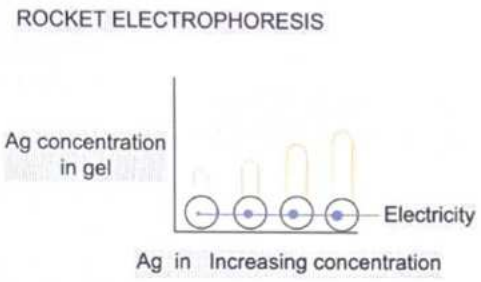
Flocculation

- Modification of precipitation
- In flocculation floccules are being produced
- Used in VDRL (slide flocculation test)



Rocket electrophoresis

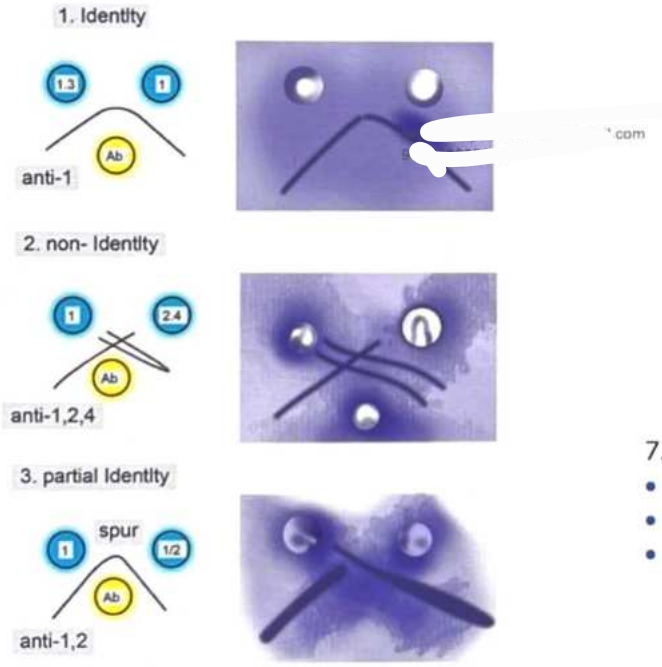
- Increase in concentration of Antigen there is an increase in height of the rockets



Immuno diffusions

01:12:55

1. Single Diffusion in one Dimension
- Aka Oudin procedure
2. Double diffusion in one dimension
- Aka Oakley-fulthorpe Procedure
3. Single diffusion in two dimensions
- Aka radial Immunodiffusion
- Can estimate the Ig levels in serum
4. Double Diffusion in two Dimension
- Aka Ouchterlony Procedure
- Eg. Elek gel precipitation test for Toxin demonstration in diphtheria



Ouchterlony procedure

Agglutination

01:20:10

- Insoluble antigen + Antibody → CLUMPS (Particulate antigen)
- Agglutination >>> Precipitation

Examples

1. Blood group estimation
2. WIDAL Test: Antibody against 'O' & 'H' are measured
3. CAT: Cold Agglutination Test for Mycoplasma
4. SAT: Standard Agglutination Test for Brucellosis
5. MAT: Microscopic Agglutination Test for Leptospirosis
6. COOMB'S TEST

Coomb's test

- Determine Incomplete IgG Antibodies
- Coomb's sera: Anti-human globulin
- It is of 2 Types
  - Direct Coomb's test:
  - Indirect Coomb's test:
- Direct Coomb's test

Refer Figure 14.1

  - Done for Autoimmune hemolytic anemia
  - Done For Fetal Ab
  - In vivo sensitization

- Indirect Comb's test

Refer Figure 14.2

  - Done For Maternal
  - In vitro Sensitization
  - Rh^+ Father + Rh^- mother → Rh^+ 1st child and during time of birth fetal blood enter maternal circulation → mother produce Anti Rh antibody (IgG incomplete antibodies) which are free in circulation
  - If 2nd child is Rh^+ these Anti Rh antibody try to destroy the antigen and erythroblastosis fetalis occurs
  - To determine If mother is having Anti Rh antibody: Do incomplete IgG Antibodies testing
    → If Free incomplete IgG Antibodies in maternal serum+ O Rh^+ RBCs [For tagging incomplete IgG Ab] +Comb's Sera → Agglutination

7. Passive Latex Agglutination Test
- Precipitation → Agglutination
- Soluble antigen + Latex particle: insoluble antigen
- Examples
  - ASO test
  - CRP
  - Rheumatoid Factor Test

Card Tests

- Based on Immuno chromatographic principle
- Sample runs on nitrocellulose paper
- Eg.
  - Hepa Card: Test becomes positive if HBs Ag present
  - Malaria Card Test
  - Pregnancy card test
- If control is negative, then card test is not valid

Neutralization Reactions

01:40:18

- Toxin + Anti toxin→ Neutralization
- Examples
  - Schick Test: done for Diphtheria
  - Nagler's Reaction: done for clostridium Perfringens [Lecithinase]

Complement Fixation Test

01:41:20

1. Wasserman Reaction: done For Syphilis
2. Sabin Feldman Dye Test: done For Toxoplasma

- Antigen + Antibody + RBC system + complement
  - If Ab present→Ag + Ab complex →Activate complement &Complement attaches to complex→ No Hemolysis [Ab present]

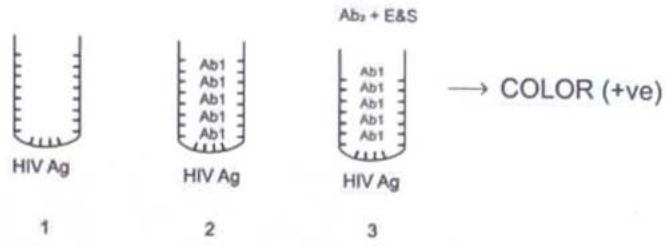
Elisa [Enzyme Linked Immuno Sorbent Assay]

01:45:40

- Types
  - Direct ELISA
  - Indirect ELISA
  - Competitive ELISA
  - Sandwich ELISA
- Components
  - Enzyme conjugate
    a. Horse Radish peroxidase [HRP] - more widely used
    b. Alkaline phosphatase [AP]
  - Substrate & Chromogens
    → Commonly used substrates
    a. p- Nitrophenyl phosphate [PNPP]
    b. Hydrogen peroxide – 0.003-0.015%
    → Chromogen: are light sensitive so it is necessary to incubate in dark
    a. TMB: Tetra Methyl Benzidine
    b. ABTS
    c. OPD: O-Phenylene Diamine
- Wave length commonly used is 450nm
- Other wavelength used are 480nm, 620nm, 405 nm etc

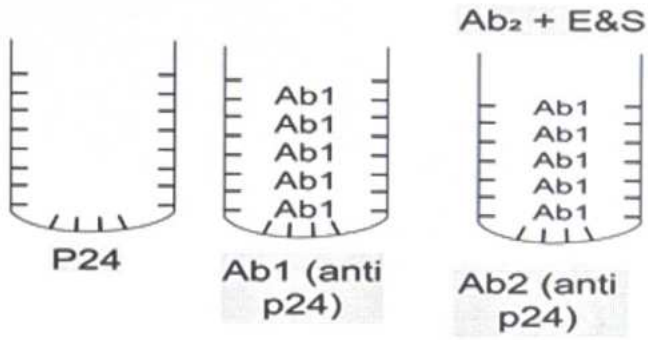
Indirect ELISA

- A well tagged with HIV Ag taken
- Patient serum with Ab1 added to it
- Ab1 injected into Goat which produce Ab2
- If color appears: +ve
- Can measure OD values (Optical Density Values)



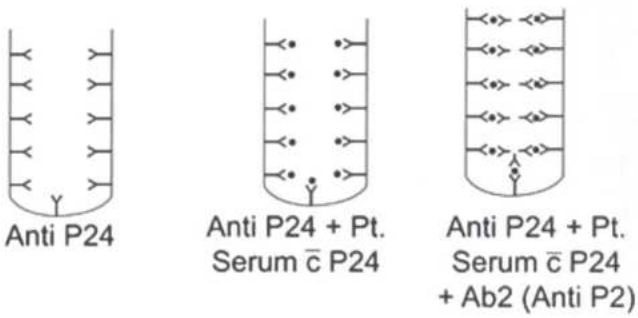
Competitive ELISA

- A well tagged with P24 Ag taken
- Patient serum with Ab1 (anti P24) added too it
- P_{24} antigen is injected into a Goat which produce Ab_2 [anti P_{24}]
- Ab_2 + Enzyme + Substrate added
- No Colour: positive



Sandwich ELISA

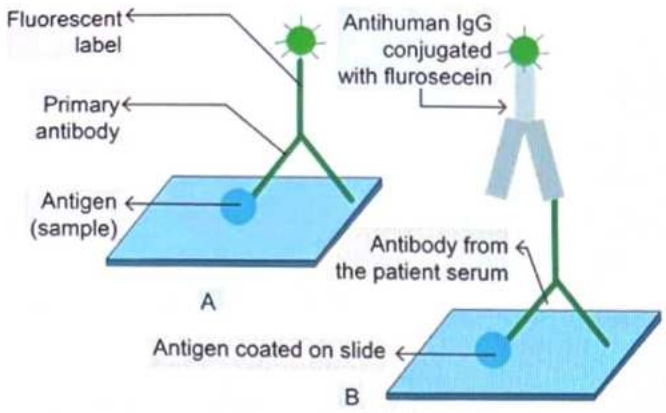
- A well tagged with anti P24 Ab taken
- Patients serum with P24 added to it
- Ab2 [antiP24] + Enzyme + Substrate added
- If colour appears: +ve
- Can measure OD (optical density with optical densitometer)



Immuno Fluorescence

02:02:22

- Types
  - Directimmuno fluorescence
    → Done for Antigen
  - Indirect immuno fluorescence
    → Done for Antibodies



Direct IF

- Detect antigens
- Principle: Antibodies tagged with fluorescent dyes→Detect unknown antigen

Indirect IF

- Detection of Antibody
- Known antigen→Unknown antibody (serum) → If Ab presents it binds with antigen→ To visualize this Ag-Ab complex Fluorescence tagged Ab to human immuno globulin is added→Fluorescence occurs

Hyper Sensitivity

- An altered state of reactivity to an Antigen / Hapten in a sensitizes host resulting in an unexpected, exaggerated reaction which is injurious to the host leading to tissue injury/ death
- Types
  - **A** - **A**naphylactic hypersensitivity (Type-1)
  - **C** - **C**ytotoxic hypersensitivity (Type-2)
  - **I** - **I**mmune complex mediated hypersensitivity (Type-3)
  - **D** - **D**elayed hypersensitivity (Type-4)

How to remember

- ACID

Type-1 HSN 02:09:53

- Occurs in 2 phases
  1. Sensitization Phase
  2. Effector Phase
- Sensitization Phase: Allergen →Processed by APC &Peptides of it is presented to CD_4 T cells→Activates TH_2 cells→Releases IL_4→Activates B cells→IgE Production→ Homocytotropism (movement towards the mast cells)
- Effector Phase →Shocking Dose→ Mast cell Degranulation → Release of primary mediators
- Primary mediators
  - **P** - **P**rotease
  - **E** - **E**osinophilic chemotactic Factor
  - **N** - **N**eutrophilic chemotactic Factor
  - **Hai** - **H**istamine & Serotonin (Leads to BC and VD)

How to remember

- PEN Hai

- Secondary Mediators:
  - **B** - **B**radykinin
  - **P**- **P**rostaglandins
  - **L**- **L**eukotrienes
  - **P**- **P**AF (Platelet Activating Factor)
  - **C**-**C**ytokines (IL-1, TNF)

How to remember

- BPL PC

- Localized Anaphylaxis / Atopy
  - Run in families
  - Eg.
    - → Food allergies
    - → Allergic rhinitis
    - → Atopic dermatitis

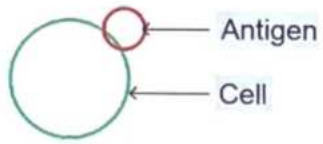
- Examples
  - P-K Reaction
  - Theobald smith Phenomenon
  - Casoni Test

Previous Year's Questions

Q. 55-year-old patient presented with difficulty in breathing & rashes after ingestion of sea food. He has shown similar reaction in post following consumption of some food item. Which type of Hypersensitivity reaction? (FMGE- Dec - 2020)

A. Type I
B. Type 2
C. Type 3
D. Type 4

Type-2 HSN 02:18:53



- Antigen + Antibody → Complement Activated
- This Activated compliment causes
  - Compliment mediated Cytolysis
    → Cytolysis is mediated by MAC (C_5-C_9)
  - Inflammation
    → Mediated by C_{3a} and C_{5a}
  - Phagocytosis
    → Mediated by C_{3b}, C_{5b}
    → C_{3b} is a major Opsoniser
- The Antigen is a part of the cell
- Examples
  - **B**
  - **P**ositive - **P**ernicious anemia
  - **B**lood - **B**lood Transfusion Reaction
  - **G**roup - **G**ood Posture Syndrome, **G**raves Disease
  - **IS** – **I**TP, **I**nsulin resistant DM
  - **M**ine - **M**yasthenia gravis
  - **D** - **D**rug induced Hemolytic anemia
  - **A** - **A**uto immune Hemolytic anemia
  - **T** - **T**ransfusion Reaction
  - **E** - **E**rythroblastosis Fetalis
  - **P**alm - **P**emphigus vulgaris

How to remember

- **B positive Blood group Is Mine DATE Palm**

- Antibody Dependent Cellular Cytotoxicity [ADCC]
  - Ig G Ab attach to target cell antigens through Fab region
  - Fc portion of IgG binds to Fc receptors of NK cells
  - Leads to destruction of the target cell
  - Used to Killing & destruction of
    → Parasites
    → Tumor cells
    → Graft tissue

- Antibody Dependent Cellular Dysfunction [ADCD]
  - Auto Ab mediated
  - Types
    1. Stimulatory ADCD
    2. Inhibitory ADCD
  - Stimulatory ADCD
    → Eg. Grave's Disease
      - Activation of receptors takes place
      - Auto Ab (Long Acting Thyroid stimulators LATS) produced
      - Stimulate Thyroid to produce hormones
  - Inhibitory ADCD
    → Eg. Myasthenia gravis
      - Inhibition of receptor takes place
      - Anti Ach receptor Ab are produced
      - Blocking of Ach receptors happens & Leads to Profound muscular weakness

Type-3 HSN 02:31:35

- Immune complex mediated
- Antigen is not a part of the cells
- Antigens can be exogenous [bacteria & virus] or endogenous [DNA etc]
- Ab Excess
  - Immune complexes are large & insoluble & tend to localize near the site of antigen administration
    → Eg. Arthus reaction (localized form of immune complex mediated vasculitis)
- Ag Excess (Monovalent Antigens)
  - Small soluble complexes tend to travel through blood to various sites & get deposited leads to Generalized Reaction
    → Eg. Serum Sickness
      - Occurs following serum therapy, mostly with Horse anti-tetanus serum
      - Manifest after 7-8 Days
      - Edema, Erythema, Glomerulonephritis can occur
- Examples
  - **H** - **H**ypersensitivity pneumonitis, **H**SP
  - **A** - **A**rthus Reaction
  - **R** - **R**eactive arthritis
  - **P** - **P**AN, **P**SGN
  - **E** - **E**pstein Barr Infection
  - **R** - **R**heumatoid Arthritis
  - **S** - **S**erum sickness, **S**LE
  - TYPE II Lepra Reaction / ENL

How to remember

- **HARPERS**

Type-4 HSN 02:39:29

- Delayed Hypersensitivity
- Sensitization Phase
  - APC present peptide fragment with MHC_2 to TH_1 cells

- This TH_1 cells differentiate to from T delayed Hypersensitivity (T_{DTH}) cells
- Other T cells are also involved which include
  - → CD_8 T cells
  - → CD_4TH_{17} cells

- Effector Phase
  - T DTH cells on subsequent antigen exposure release cytokines
  - The cytokines attract & recruit inflammatory cells at the sites
  - Cytokines
    - → **I**- Interferon [most important]
    - → **I** - IL_2
    - → **M** - MCAF (Macrophage chemo Attractant Factor)
    - → **To** - TNF (Tumor Necrosis Factor)
    - → **Medical** - MIF (macrophage Inhibition Factor)
    - → **Institute** - IL_3
    - → **Gujarat** - GMCSF (Granulocyte monocyte colony stimulating Factor)

How to remember

- IIM To Medical Institute Gujarat

  - IFN → activates resting Macrophages
  - IL_2 → Autocrine action & helps in proliferation of T_{DTH} cells

- Pathology
  - Continuous TDTH reaction For Killing intracellular microbes leads to formation of granuloma (TB, Leprosy etc)
  - T helper cell infiltrate is replaced by macrophages in 2-3 weeks
  - Macrophage transforms into
    - → Epitheloid cell
    - → Epitheloid cells fuse to form Multinucleated giant cells
- Examples
  - Tuberculin test→Delayed type hypersensitivity
  - Lepromin test
  - Frie's test
  - Type-1 Lepra reaction
  - Contact dermatitis
    - → Nickel, oak poison act as Haptens→Haptens + Skin proteins→Internalized by skin APC [Langerhan's cells]→Presented to TDTH cells→Activates macrophages→Release lytic enzymes→Causes skin redness & pustule formation

Previous Year's Questions

Q. Type IV Hypersensitivity is due to?
(AIIMS - June - 2020)

A. Innate immune response
B. Cell mediated immunity
C. Antibody & cell mediated immunity
D. Humoral mediated immunity

Figure 14.1

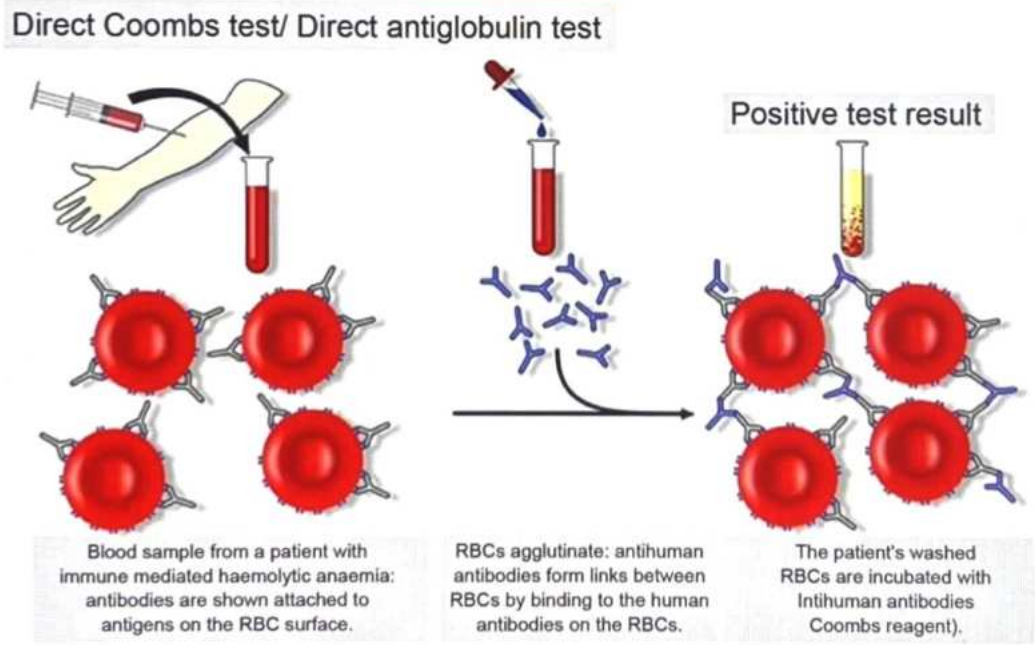
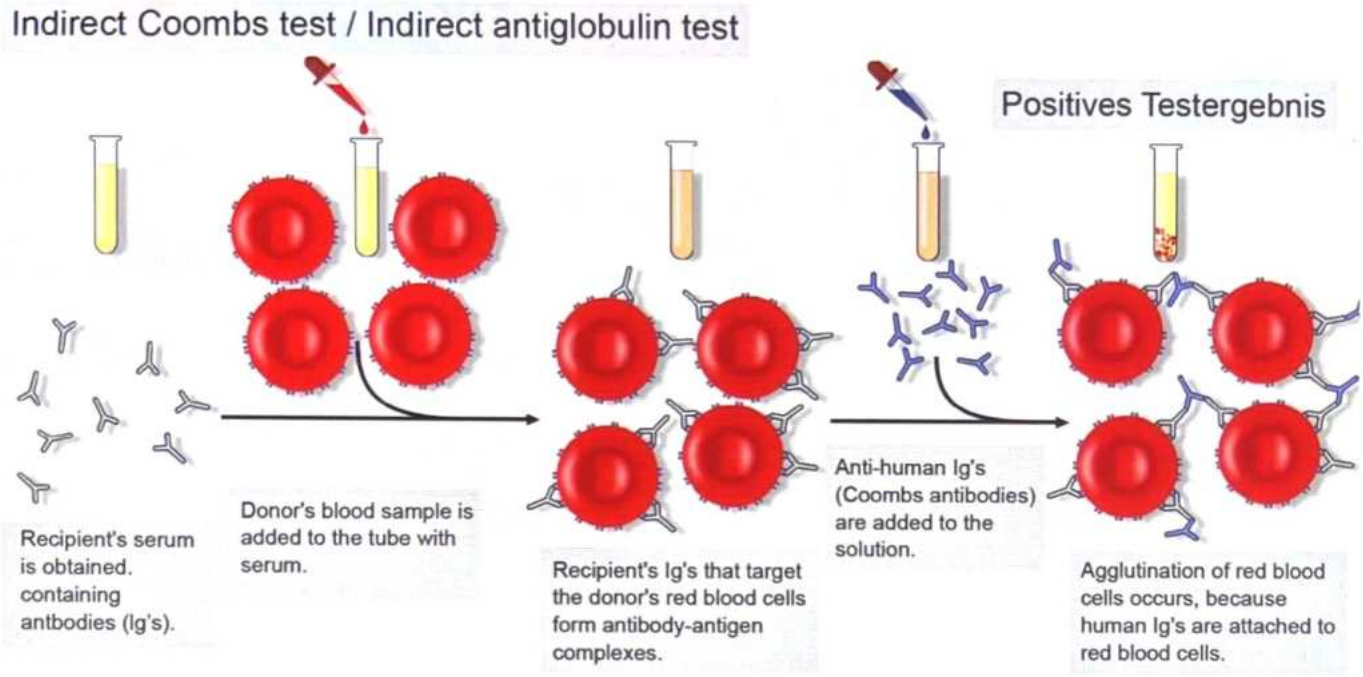


Figure 14.2


Indirect Coombs test / Indirect antiglobulin test
Positives Testergebnis
Recipient's serum is obtained. containing antbodies (Ig's).
Donor's blood sample is added to the tube with serum.
Recipient's Ig's that target the donor's red blood cells form antibody-antigen complexes.
Anti-human Ig's (Coombs antibodies) are added to the solution.
Agglutination of red blood cells occurs, because human Ig's are attached to red blood cells.

CLINICAL QUESTIONS

Q.1 A 55-year-old patient with complaints of pain, stiffness, and swelling in all the fingers of both her hands visited the OPD. The doctor sent her sample for immunoglobulin testing to the microbiology laboratory. All of the following are false regarding classes of immunoglobulin except?

A. IgM has maximum half-life of 23 days
B. IgE has least serum concentration of 0.0003 mg/ml
C. IgG has maximum sedimentation co-efficient of 19
D. IgD has a carbohydrate concentration of 8 %

Solution:

- Highest serum half-life and serum concentration is of IgG.
- Least serum half-life and serum concentration is of IgE.
- Highest sedimentation coefficient is of IgM.

	IgG	IgM	IgA	IgD	IgE
Serum concentration	**12 mg/ml**	1.2 mg/ml	2 mg/ml	0.03 mg/ml	0.0003 mg/ml
Half-life	**23 d**	5 d	6 d	2-8 d	1-5 d
Molecular wt.	150,000	**900,000**	160,000	180,000	190,000
Carbohydrate	2-3%	12%	8%	**13%**	12%
Sedimentation Coefficient	7	**19**	7	7	8

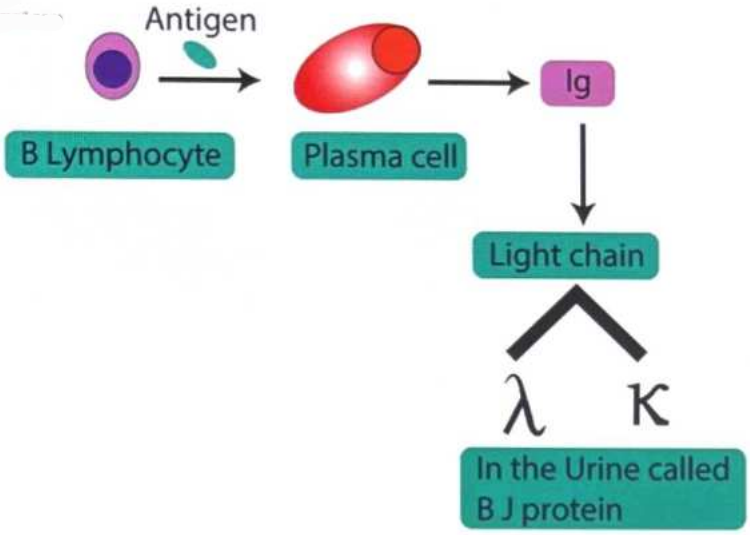
Q.2 A 55-year-old male presented with symptoms of exertional fatigue. His Hb was 10.6g/dl, with an MCV of 92fL. He had normal s.ferritin; vitamin B12 and folic acid levels. A serum protein electrophoresis demonstrated a monoclonal IgA protein of 1.5 g/dL and a skeletal survey showed occult lytic lesions in the skull and B/L humeri. The physician recommended to do a Bence Jones proteins testing. How are Bence Jones proteins best described?

A. μ chains
B. γ chains
C. Kappa & Lambda chains
D. Fibrin split products

Solution:

- Bence Jones proteins are best described as **Kappa & Lambda chains.**

- **Bence Jones proteins:**
  - Excretion of **light chains** in the **urine** has been referred to as **Bence Jones proteinuria**.
  - **Light chains includes κ and λ (kappa and lambda)**
  - Detection of Bence Jones protein may be suggestive of multiple myeloma or Waldenstrom's macroglobulinemia.
  - Bence Jones proteins are light chains of immunoglobulins so may occur as Lambda or Kappa forms, but in any patient the chain is either Kappa or Lambda, never both.



Other Options:

- Fibrin split products are seen in a case of thrombolysis or DVT.
- μ and ϒ are the types of heavy chain.

15 IMMUNOLOGY PART-2

HYBRIDOMA TECHNOLOGY

00:00:15

- Done for mass production of Monoclonal antibodies
- Principle: B cells fused up with Myeloma cells on HAT media
  (Hypoxanthine Aminopterine Thymine media)
  - Mouse is injected with desired Antigen → Produces B cells (HGPRT +ve)
  - B cells [HGPRT +ve] + Myeloma cells [HGPRT-ve] on HAT media produce
    → Unfused B cells (HGPRT +ve)→Do not survive
    → Unfused Myeloma cells (HGPRT –ve)→ Do not survive
    → Fused cells (Hybrid cells)→Survive
- Aminopterine
  - Blocks denovo pathway for Purine synthesis
  - Now every cell will go through salvage pathway to synthesize Purine
- Hybrid cells → Mice → Desired Monoclonal antibodies

COMPLEMENT SYSTEM

00:07:02

- Non-Specific Proteins
- Constitute 5% of serum proteins
- Heat labile (56°C x 30 min)
- Can blind to Fc portion of Ig

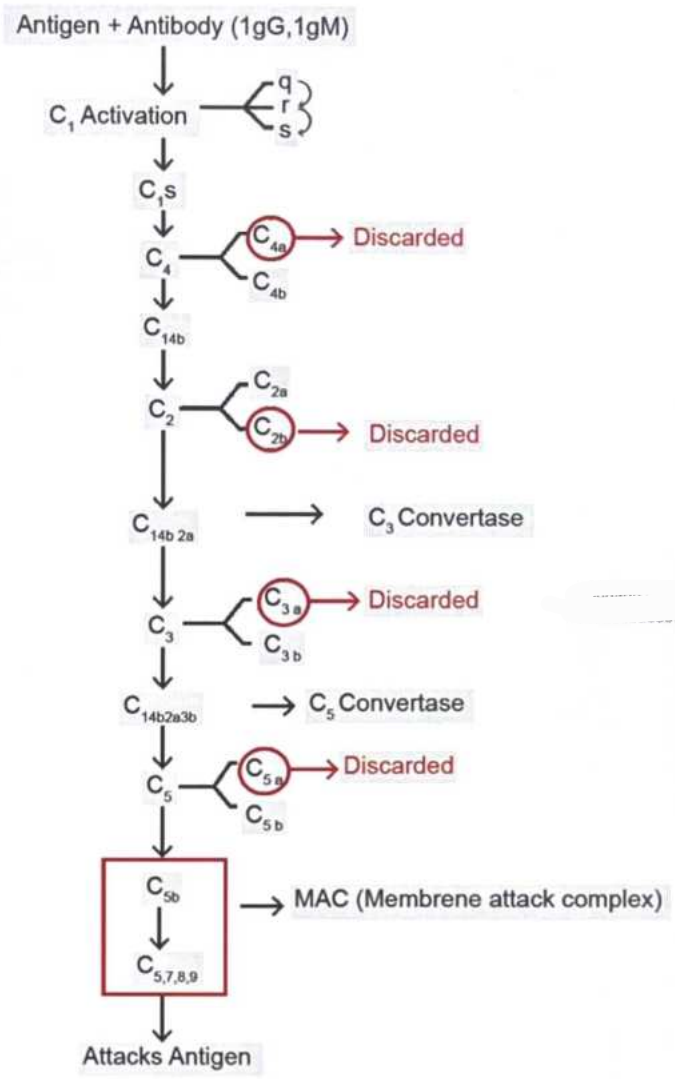
Sites of Synthesis

- GIT→C_1
- Macrophage→C_2, C_4
- Spleen→C_5, C_8

Types

	Classical	Alternative	Lectin
• Activators	• Ag + Ab complex	• Zymogen • Endotoxin • Cobra venom • Nephritic Factor	• Mannose binding Lectin
• 1st Activated	• C_1	• C_3	• C_4
• Part of	• Acquired immunity	• Innate immunity	• Innate immunity

Classical Complement Pathway

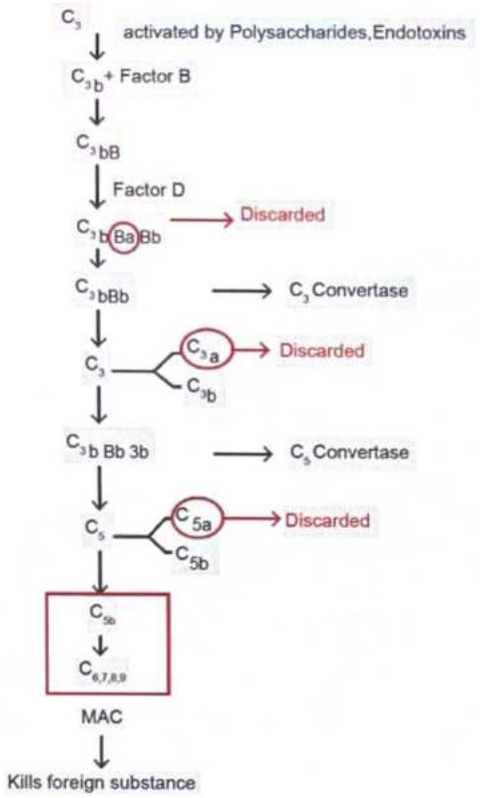


Previous Year's Questions

Q. Classical complement activated by?
(FMGE– June - 2019)

A. Cl
B. C3 convertase
C. IgA
D. Ag – Ab complex

Alternate Complement Pathway



- C_1, C_4, C_2 are not consumed in alternate pathway

Previous Year's Questions

Q. Which of the following is a major component in activation of the complement via alternative pathway? (FMGE – June - 2018)

A. C1
B. C2
C. C3
D. C4

Lectin Pathway

- Similar to Classical complement pathway, but starts with C_4
- It happens in GNB as the contain mannose binding lectin in their cell wall
- C_1 is not consumed

Biological Role of Complement System

1. Lysis of target cells
2. Chemotactic role → C3a, C5a
3. Anaphylactic role → C3b (best Opsonin)
4. Mediate Type 2 & Type 3 Hypersensitivity Reactions
5. Auto immune diseases
6. Endotoxic shock

Deficiency

00:23:42

- MAC Deficiency → Leads to Recurrent Neisserial infections
- Factor D & I absence → Leads to Pyogenic infections
- Factor H absence → leads to Hemolytic Uremic syndrome
- C_1 esterase inhibitor deficiency → Hereditary Angioneurotic Edema (HAE)
  - Side effect of ACE Inhibitors → HAE (d/t C_1 esterase deficiency)

Previous Year's Questions

Q. Meningococcal meningitis is seen with which of the following deficiency? (FMGENov2017)

A. Clq
B. C2
C. C4
D. C5

STRUCTURE OF IMMUNE SYSTEM

00:26:11

Myeloid System	Lymphoid System
• RBC • WBC • Platelets	• Lymphoid cells • Thymus • Bone marrow

Lymphoid Organs

00:27:31

- Central lymphoid organ
  - Thymus
    - → Maturation of T cells occurs in Thymus
  - Bone marrow
    - → B & T cells originates From BM
    - → B cells mature in BM

- Peripheral lymphoid organ
  - Spleen
    - → T dependent area in Spleen: Area around Periarteriolar lymphoid sheath
    - → B dependent area in Spleen: Germinal follicle [Marginal Zone]
  - Lymph nodes
    - → T dependent area in a LN: Para cortical area
    - → B dependent area in a LN: Cortical follicle
  - MALT [Mucosa Associated Lymphoid Tissue]
  - GALT [Gastric Associated Lymphoid Tissue]
  - Liver

CD Markers

- T cells→CD_1 to CD_8
- B cells →CD_{19} to CD_{23}
- NK cells → CD_{16} and CD_{56}

B-Lymphocytes 00:32:35

- Constitute 10-20% of total lymphocytes
- Also acts as APC (Antigen presenting cells)
- CD_{21} / CR_2→EBV receptor for B cells
- On Electron Microscopy →shows Microvilli on surface of B-cell
- Blast Transformers of B cells → Endotoxin (mainly), EBV Ag
- Blast Transformers of T cells → Concavalin and Phytohaemagglutinin
- Rosettes Types on RBC
  - B cells → EAC Rosettes
  - T cells→ E Rosettes

T Cells 00:36:05

- Constitute 60-70% of total lymphocytes
- Types
  - CD_4 T-cells
    - → TH_1:secretes IL-2, IFN
    - → TH_2:secretes IL-4, 5, 6, 10
    - → The earliest identifiable T cell precursor in bone marrow: CD_{34} + Pro T cells
  - CD_8 T-cells (Cytotoxic T-cells): causes lysis of target cells

NK Cells

- Constitutes 5-10% of all lymphocytes
- Kills Virus infected cells & Tumor cells
  - APCs → Secretes IL-1→Stimulates TH_1 cells and releases IL-2
  - This IL-2 changes NK cells into LAK cells (Lymphokine Activated Killer cells)
- Play a role in Antibody Dependent cellular Cytotoxicity

MHC [Major Histocompatibility Complex] 00:40:27

- Present on chromosome 6p

Class-I MHC Genes

- Present in all nucleated cells
- Provides peptide fragments of cell to CD_8 T cells
- Helps in processing of Virus infected cells & Tumor cells

Class-II MHC Genes

- Present over all APCs which are depicted by DP/DQ/DR
- They attaches to exogenous peptide fragments and present them to CD_4 T cells
- Plays a major role in Transplantation antigens immune response

Class-III MHC Genes

- A/w Complements → C_2, C_4, Properidin factors,TNF, HSP (Heat shock protein)

MHC Restriction

- Exogenous antigen (Peptide fragments with MHC II) are handled by: CD_4 T cell
- Endogenous antigen (Peptide fragments with MHC I) are handled by: CD_8 T cell

Macrophages 00:45:50

- Play a role in Phagocytosis
- Act as APCs
- Modified Macrophages
  - Peripheral Blood→Monocytes
  - Liver→Kupffer cells
  - Brain→Microglia cells
  - Kidney→Mesangial cells
  - Bone→ Osteoclast
  - Lungs→Alveolar macrophages
  - Skin→ Langerhans cells
  - Placenta →Hofbauer cells
- Acts/ Kills by
  - O_2 free radical killing: because of Phagolysosomal fusion

Dendritic Cells

- APCs
- Derived from BM
- Transported to lymph nodes

Interleukins 00:49:49

Refer Table 15.1

Chemokines 00:53:25

- CxC Chemokines
  - Act on Neutrophils
  - IL8, IL1, TNF
- CCChemokines
  - Aka Eotaxin, RANTES
  - Monocyte chemoattractant protein 1
- C Chemokines
  - Specific For lymphocytes & lymphotactin
- CX_3C Chemokines
  - Specific for Fractalkine

Table 15.1

	Secreted By	Function
• IL 1	• Monocytes • Macrophages	• Induces inflammation, fever
• IL 2	• TH_1 Cells	• NK cells →LAK cells
• IL 3 • [GMCSF]	• TH Cells	• Granulocyte Monocyte Colony Stimulating Factor (GMCSF)
• IL 4	• TH_2 cells	• Class switching to IgE
• IL 5	• TH_2 cells	• Class switching to IgA • Eosinophil attractant
• IL 8	• Macrophages	• Neutrophill chemoattractant
• IL 12	• Macrophages	• Promotes TH_1 induction & inhibit TH_2

CLINICAL QUESTIONS

Q.1 A 44 year old woman currently working in a bakery, presents with a 1 year history of asthma and allergic rhinitis. She especially finds red bran to worsen her symptoms almost immediately on exposure. This condition points towards type I hypersensitivity reaction. What are the most important cells in type I hypersensitivity?

A. Macrophages
B. Mast cells
C. Neutrophils
D. Lymphocytes

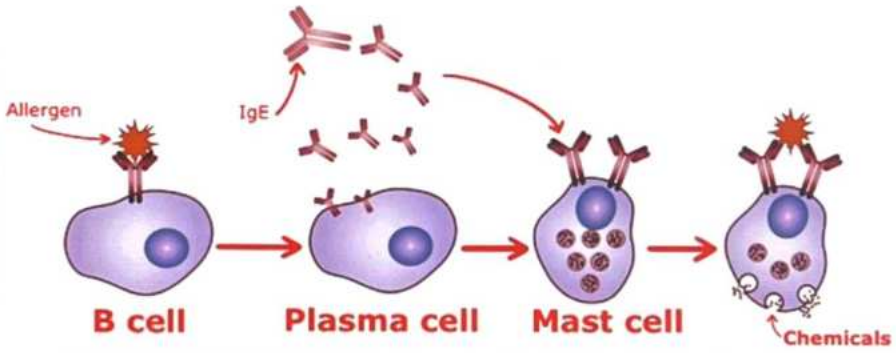
Solution:

- **Mast cells** are the most important cells in type I hypersensitivity.

Type I hypersensitivity reaction:

- Commonly called **allergic or immediate** hypersensitivity reaction.
- This reaction is always rapid, occurring within minutes of exposure to an antigen, and always involves **IgE-mediated degranulation of basophils or mast cells.**

Option 1, 2, 3, 4



- **occurs in 2 phases:**

1. Sensitization Phase
2. Effector Phase

Sensitization Phase:

- Allergen is processed by APC & Peptides of it presented to CD4 T cells → Activates TH_2 cells → Releases IL4 → Activates B cells → **IgE** Production → HOMOCYTOTROPISM [movement towards the mast cells].

Effector Phase:

- Shocking Dose → **Mast cell Degranulation** → primary and secondary **mediators.**
- **Primary Mediators:**
  - P → Protease
  - E → Eosinophil chemotactic Factor
  - N → Neutrophil chemotactic Factor
  - Hal → Histamine & Serotonin
- **Secondary Mediators:**
  - B → Bradykinins
  - P → Prostaglandins
  - L → Leukotrienes
  - P → PAF [Platelet Activating Factor]
  - C → Cytokines [Il1, TNF α]

Type I reaction can occur in two forms:

- anaphylaxis
- atopy

Q.2 A sample from an HIV positive patient was sent to the microbiology laboratory. The microbiologist is explaining his intern about different tests which can be performed for HIV. ELISA test when compared to Western blot technique for diagnosis of HIV infection is?

A. Less sensitive, less specific
B. More sensitive, more specific
C. Less sensitive, more specific
D. More sensitive, less specific

Solution:

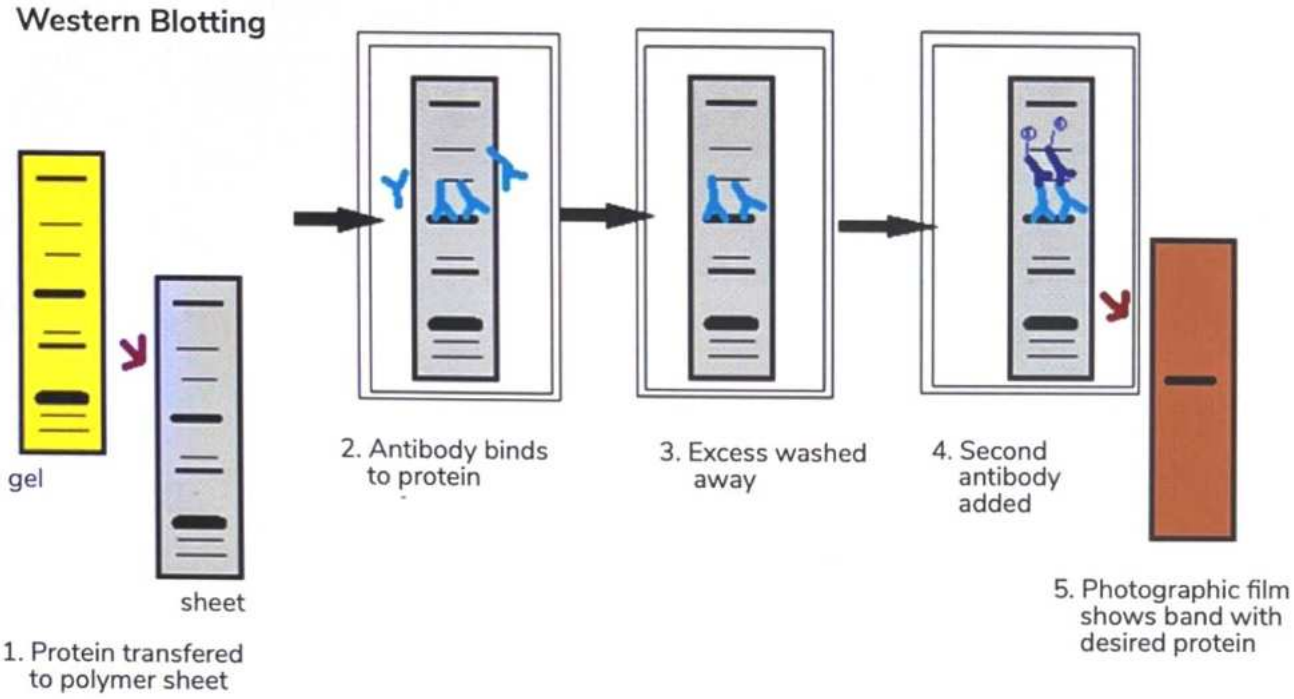
- **ELISA test is More sensitive, less specific** when compared to Western blot technique.
- **ELISA**:
  - ELISA (screening test) is the most frequently used test for detection of both HIV-1- and HIV-2-specific antibodies in the serum.
  - **Highly sensitive**, but less specific.
- **Supplementary or confirmatory tests**: These tests are **designed for a higher specificity than the screening tests**, hence are used as the test of choice **to verify the results of screening tests**. Western blot, line immunoassay, and immunofluorescence assays are the most commonly used.

HIV RNA:

- Gold standard' for confirmation of HIV diagnosis
- Reverse transcriptase PCR (RT-PCR)
- Branched DNA assay
- NASBA (Nucleic acid sequence based amplification)
- Real time RT-PCR for estimating viral load

⇒Most sensitive test → ELISA

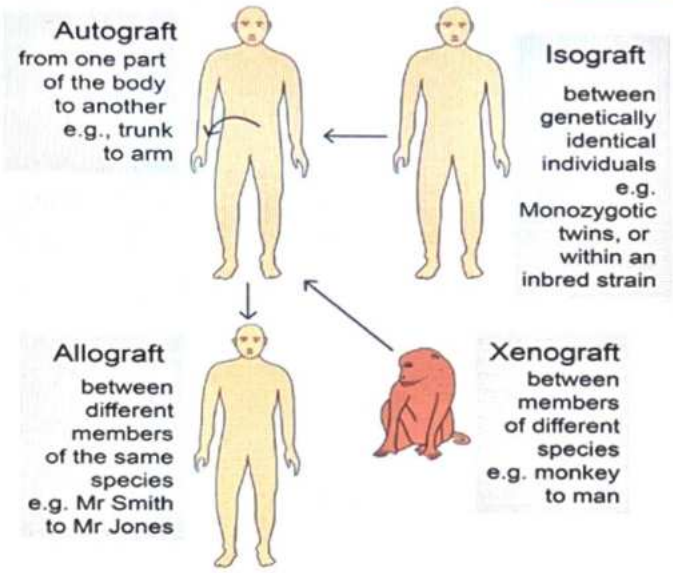
⇒**Most specific test → HIV RNA PCR** & Western blot.



16 IMMUNOLOGY PART-3

GRAFT

00:00:16



Types

- Autograft: from one part of the body to another
  - Eg. Trunk to Arm
- Allograft: b/w different members of same species
  - Eg. Mr. Smith to Mr. Jones
- Xenograft: b/w members of different species
  - Eg. Monkeys to Man
- Isograft: b/w genetically identified twins
  - Eg. Monozygotic Twin or with an inbred strain

Previous Year's Questions

Q. An elderly male patient with some clinical scenario is undergoing kidney transport from his twin brother. What kind of graft is it?
(FMGE – Dec - 2020)

A. Isograft
B. Allograft
C. Xenograft
D. Autograft

Allograft Rejection 00:02:15

Series of Events

- Graft is vascularised, healthy in first 3 days
- From 4th day
  - There is inflammation with Lymphocytes, Macrophages
  - Blood vessels in Graft are occluded by thrombi
- By 10th day
  - Vascularity diminishes & graft undergoes Ischemic necrosis
  - Necrosis & scab like appearance and Slough off K/a 1st set response
- If another graft from the same donor, it will be rejected in an accelerated fashion→ Graft slough off by 6th day

Graft rejection 00:06:12

Type	Time Taken	Mechanism
• Hyperacute • Acute • Chronic	• Minutes to hours • Days to weeks • Months to years	• Due to Preformed Ab • CD_4 T cell activation • Fibrosis & Wide spreadArteriopathy [T cell]

Previous Year's Questions

Q. Nude mice is not resistant to Xenograft due to absence of?
(NEET Jan 2018)

A. B cell
B. T cell
C. NK cell
D. LAK cell

Graft VS Host Reaction

- **Billing Ham's Criteria**
  1. Graft contains immune competent cells
  2. Host is immunosuppressed
  3. MHC incompatible

- Organs damaged
  - Skin, Liver & GIT are the main targets
  - GVH model in animal: Runt Disease
    → Cachexia occurs and later on Animal dies

Typing Methods 00:13:00

HLA Typing

- DNA PCR Typing: Best
- Primitive Methods: Not done nowadays
  - Micro Cytotoxicity
  - Mixed Leukocyte Reactions

AUTOIMMUNITY 00:14:29

- Immune response to our self antigens leads to various pathological conditions/diseases

Mechanisms of Autoimmunity

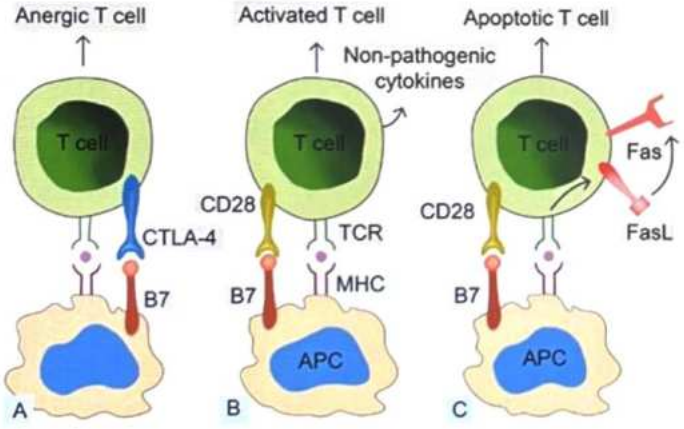
1. Exposure of sequestrated antigen
2. Formation of Neo antigens
3. Immunological damage by cross reactivity
4. Mismatch in CMI and HMI
5. Exposure of forbidden clones

Immunological Tolerance

- It is due to 2 things
  - Central tolerance: in the central lymphoid organs
  - Peripheral Tolerance

- Central tolerance
  - In Thymus: Self reactive T-cells are negatively selected
  - BM: 2 mechanisms happen
    - → Receptor Editing
    - → Negatively selected by Apoptosis

- Peripheral tolerance: by
  - Ignorance
  - Anergy
  - Phenotypic skewing
  - Apoptosis
  - Dendritic cells
  - Sequestration of self antigens



1. Exposure of Sequestrated antigen
- Lens protein and Sperm protein

2. Formation of Neo Antigens
- Because of any Biological agent or Chemical agent

3. Immunologic reaction by cross reactivity
- Carbohydrate 'C' cross reacts with Cardiac valves
- 'M' Protein cross reacts with Myocardium

4. Breakdown of immunological homeostasis
- No coordination b/w Cell mediated immunity & Humoral immunity

5. Exposure to Forbidden Clones

Examples of Autoimmune diseases

1. AIHA (Auto Immune Hemolytic Anemia)
2. Myasthenia Gravis
3. Graves disease
4. SLE [Cause of Death is due to Kidney lesions mainly]
5. Rheumatoid Arthritis: IgM is produced against Fc fragment of IgG
6. Reactive Arthritis
7. Hashimoto's Thyroiditis
8. Sjogren's Syndrome
9. Multiple sclerosis
10. Good pastures syndrome

IMMUNO DEFICIENCY DISORDERS 00:39:01

Disorders of Phagocytosis

1. **CGD (Chronic Granulomatous Disease)**
- Deficiency of NADPH Oxidase
- Leads to recurrent Catalase positive infections (Staph & Coliforms)
- Nitro Blue Tetrazolium Test negative

2. **LAD syndrome**
- Absence of beta chain of integrin
- Leads to recurrent infection

3. **Chediak Higashi Syndrome**
- Defective Phagolysosomal fusion
- ↓↓↓ in Melanocytes

4. **Job Syndrome/ Hyper IgE Syndrome**
- Leads to frequent cold and recurrent Staphylococcal abscess
- ↑ IgE

B Cell Disorders 00:44:34

1. **Bruton's Agammaglobulinemia/ X Linked Agammaglobulinemia**
- Defect in B Tyrosine kinase gene (BTK gene)
- Due to BTK gene deletion the Pre B-cells cannot change into immature B-cells

- Upto 6 months of neonatal life, nothing happens to child [d/t Maternal Antibodies]; After that severe infections occur
- Treatment: external Immunoglobulins

2. X Linked Hyper IgM Syndrome
- Deficiency of CD_{40L} on activated T cells due to which Isotype switching fails
- ↑ Serum IgM
- T cells are normal

3. Selective IgA Deficiency
- M/c selective Ig deficiency
- Associated with Giardiasis infection

4. Common Variable Immuno Deficiency
- Unknown defect
- B Cells fails to mature into Plasma cells
- ↓↓Immunoglobulins
- Late onset
- Commonly a/w Giardiasis

T Cell Defects 00:50:55

1. Digeorge Syndrome
- Absent Thymus
- Presents with
  - C - Cardiac abnormality
  - A - Abnormal Facies
  - T - Thymic Hypoplasia
  - C - Cleft palate
  - H - Hypocalcemia: Recurrent episodes of Tetany
  - 22- Chromosome 22q11 defect

How to remember

- CATCH22

2. Chronic Mucocutaneous Candidiasis
- Also a T cell Disorder
- Causes Recurrent Candidiasis

Combined B & T- Cell Disorders 00:54:48

1. Wiskott Aldrich Syndrome
- WASP gene defect
- Triad
  - R - Recurrent infections
  - E - Eczema
  - T – Thrombocytopenia

How to remember

- RET

- IgM: decreased
- IgG & IgA: Normal
- IgE: increased

2. Ataxia Telangiectasia
- Defect in Kinase involved in cell cycle
- Deficiency of IgA & IgE
- Leads to
  - Cerebral ataxia
  - Telangiectasia
  - Ovarian dysgenesis

3. Severe Combined Immuno Deficiency[SCID]
- Etiology
  - Defect in IL-2 receptor gamma chain
  - 2nd MC defect: Adenosine deaminase (ADA) deficiency
  - Rag 1 & Rag 2 genes defect → Total absence of T & B cells
  - TAP-1 and TAP-2 defect
  - Deficiency of PNP [Purine Nucleoside Phosphorylase]
- Treatment: Bone marrow transplantation

Previous Year's Questions

Q. Gene not involved in SCID? (AIIMS – Nov - 2019)

A. BTK
B. ZAP70
C. IL2RG
D. JAK3

Previous Year's Questions

Q. T cell is associated with? (AIIMS – May - 2018)

A. Cd4
B. Cd5
C. Cd8
D. First line defense against bacterial peptides

CLINICAL QUESTIONS

Q.1 A patient presented with complains of fever, red and tender skin and swollen lymph nodes to the outpatient department. The doctor diagnosed the condition as granulomatous disease in which cell mediated immunity is affected with involvement of T cells. What are the type of receptors present on T cells?

A. IgG
B. IgD
C. CD4
D. Prostaglandins

Solution:

- **CD4** receptors are present on T cells.
- **T cell receptor for antigen:**
  - The TCR is the recognition molecule for T cells.
  - The TCR is a transmembrane heterodimeric protein containing two disulfide-linked chains.

Two different classes of TCR:

- Alpha-beta (α and β): majority of the T cells
- Gamma-delta (γ and δ): a smaller percentage of T cells

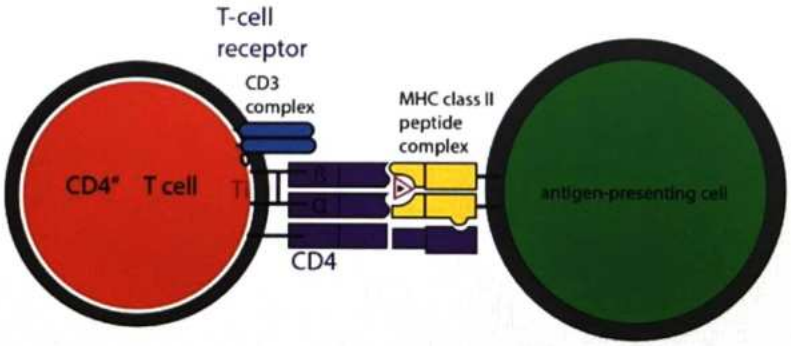
The **αβ T cells** are subdivided by their surface markers:

- CD4 : bind to MHC II
- CD8: bind to MHC I

They recognize peptide antigens presented by MHC molecules on the surfaces of APC's (antigen-presenting cells).

γ δ T cells:

- Primarily located in the **epithelial linings of the reproductive and GI tracts.**
- They recognize different type of antigens without the requirement of MHC.



Other options:

- Prostaglandins (eicosanoids) are the lipid compounds acting as mediators of pain and inflammation. They have diverse action such as bronchodilation, vasodilataion etc.
- IgG and IgD are immunoglobulins involved in defence mechanism.

Q.2 A 68 year old man who developed a harsh productive cough four days prior to being seen by a physician. On examination the sputum is thick and yellow with streaks of blood. He developed fever, chills and malaise along with the cough. One day ago he developed pain in his right chest that intensifies with inspiration. Chest X-ray shows cavitation's which confirms pneumonia. The given condition affects humoral immunity. Which cells are involved in humoral immunity?

A. B-cells
B. T-cells response
C. Helper cells
D. Dendritic cells

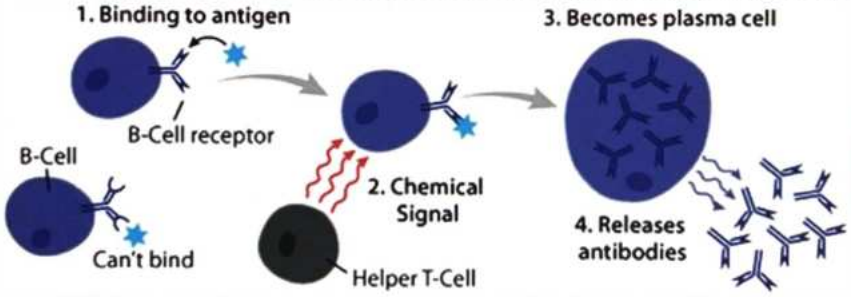
Solution:

- **B cells are involved in humoral immunity.**

Option 1

B cells:

- B cell **form plasma cell → antibody.**
- The antibodies recognize microbial antigens, combine specifically with the antigens, neutralize the infectivity of microbes, and target microbes for elimination by various effector mechanisms.



Major Functions of Antibody-Mediated Immunity (B Cells):

- Host defense against infection (opsonize bacteria, neutralize toxins and viruses).
- Allergy (hypersensitivity) (e.g., hay fever, anaphylactic shock)
- Autoimmunity

Option 2, 3

T cells: cell mediated Immunity

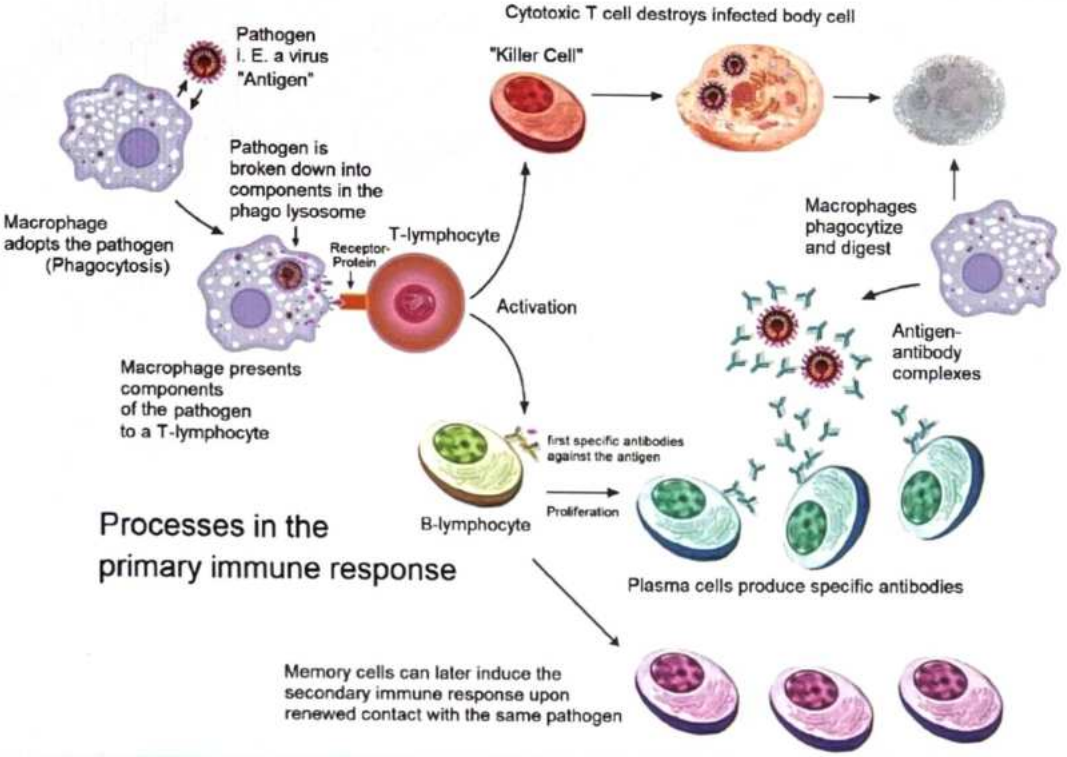
- Cell mediated Immunity is mediated by both activated T helper cells (CD4 T cells) and cytotoxic T cells (CD8 T cells).

Major Functions of Cell-Mediated Immunity (T Cells):

1. Host defense against infection (especially M. tuberculosis, fungi, and virus-infected cells).
2. Graft and tumor rejection.
3. Regulation of antibody response (help and suppression)

CD4+ T helper cells perform following functions:

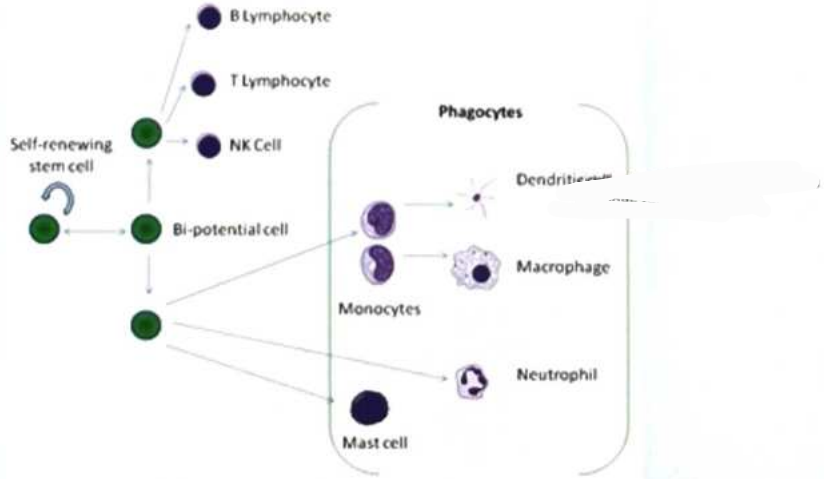
- They help B cells to be transformed into plasma cells.
- They help CD8+ T cells to become activated cytotoxic T cells.
- Cytokines secreted by helper (Th) T cells activate various phagocytic cells, enabling them to phagocytose and kill microorganisms.
- They help macrophages to mediate delayed type hypersensitivity reactions.



Option 4

Dendritic Cells:

- Antigen presenting cells (APCs).
- Derived from Bone marrow.
- Transported to lymph nodes.



LEARNING OBJECTIVES

UNIT 7: GENERAL MICROBIOLGOY

- General microbiology
  - Scientists in microbiology
  - Microscopy: Light microscope, Phase contrast microscope, Dark field microscope, fluorescent microscope, and Electron microscope
  - Bacteria
    - → Prokaryotes vs eukaryotes
    - → Shapes, Cell wall demonstration, Capsule, Flagella, Bacterial spores
    - → Physiology of bacteria
  - Sterilization and disinfection
    - → Sterilization methods
    - → Testing of disinfectants
    - → Sporicidal agents
    - → Spaulding classification
  - Bacterial genetics: Bacterial variations, Mutation, Gene transfer and Jumping genes

17 GENERAL MICROBIOLOGY PART-1

SCIENTIST IN MICROBIOLOGY

00:00:15

Louis Pasteur

- Father of microbiology
- Proposed Fermentation principle
- Disapproved Abiogenesis theory
- Coined Germ theory
- Gave concept of Autoclave & Hot air oven
- Vaccines
  - **C** - **C**holera
  - **A** - **A**nthrax
  - **R** - **R**abies → 1st rabies vaccine given to Joseph Meister

How to remember

- CAR

Robert Koch

- Father of modern Microbiology
- Proposed Koch Postulates
- Discovered TB, cholera bacilli
- Proposed Hanging Drop method
- Proposed solid media concept
- Koch Postulates
  - Constant association with disease must be present with causative organism
    → Eg. MTB always causes TB
  - Isolation in a pure culture media should be possible
  - Culture growth inoculated in animal should cause same lesion
  - Re-isolation should be possible from experimental animal
  - Antibodies should be produced from antigens
- Bacteria which does not follow Koch Postulates
  - Mycobacterium Leprae
  - Treponema Pallidum
  - Gonococci

Antony Von Leeuwenhoek

- Father of microscopy
- Coined the term "Little Animalcules" For micro organism

Alexander Fleming

- Discovered Penicillin

Paul Ehrlich

- Proposed Acid Fast staining
- Toxin & anti toxin standardization
- Father of Chemotherapy

Joseph Lister

- Father of antiseptic surgery
- 1st person to introduce Carbolic acid as antiseptic for surgery

Edward Jenner

- Father of vaccination

Kary B Mullis

- Father of PCR

Important Information

- **Kleb-Loffler's Bacilli: C. diphtheriae**
- **Preisz Nocard Bacilli: C. pseudotuberculosis**
- **Koch Week Bacilli: H. aegypticus**
- **Whitmore Bacilli: Burkholderia pseudomallei**
- **Pfeiffer's Bacilli: Haemophilus influenza**
- **Friedlander's Bacilli: Klebsiella Pneumoniae**

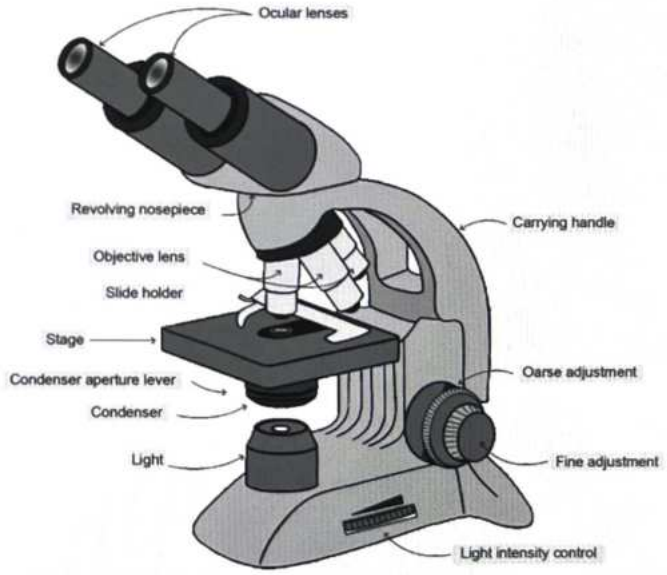
Characteristics	Viruses	Bacteria	Fungi	Protozoa/ Helminths
• Cells	No	Yes	Yes	Yes
• Diameter [µm]	20 to 200 nm	1-5 µm	3-10 µm (yeast)	15-25 µm [Trophozoite]
• Nucleic Acid	DNA or RNA	DNA & RNA	DNA & RNA	DNA & RNA
• Type of Nucleus	None	Prokaryotic	Eukaryotic	Eukaryotic
• Ribosomes	Absent	70s [50s + 30s]	80 s [60s + 40s]	80s [60s + 40s]
• Mitochondria	Absent	Absent	Present	Present
• Outer surface	Capsid	Rigid wall Peptidoglycan	Rigid wall Chitin	Flexible Membrane
• Motility	None	Some	None	Most
• Replication	-	Binary Fission	Budding mitosis	mitosis

MICROSCOPY

00:29:04

- Depends on
  - Magnification
  - Resolution: ability to distinguish two points clearly as separate
    - → Human eye→0.2 mm
    - → Light microscope→0.2 µ
    - → Electron microscope→0.2 nm

Light Microscope



- White light is used as source
- Used For

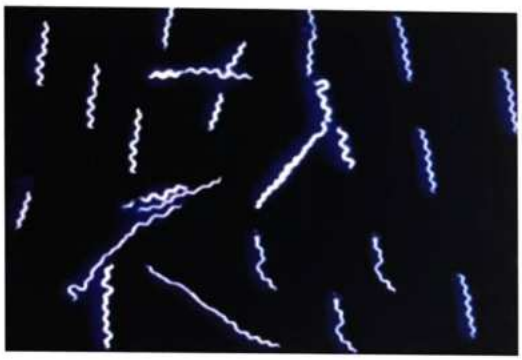
- Motility demonstration [hanging drop method]
- Gram staining, Zn staining
- Stool examination (cyst and eggs can be seen)

- Objective lens can be 10x, 40x, 100x
- 100X: Oil immersion Lens
- 10X, 40X: Demonstrate motility or stool examination
- Eye piece 10x
- Highest magnification achieved by oil immersion lens [100x X 10x = 1000x]
- By oil immersion lens we can see
  - Gram staining, Zn staining

Phase Contrast Microscopy

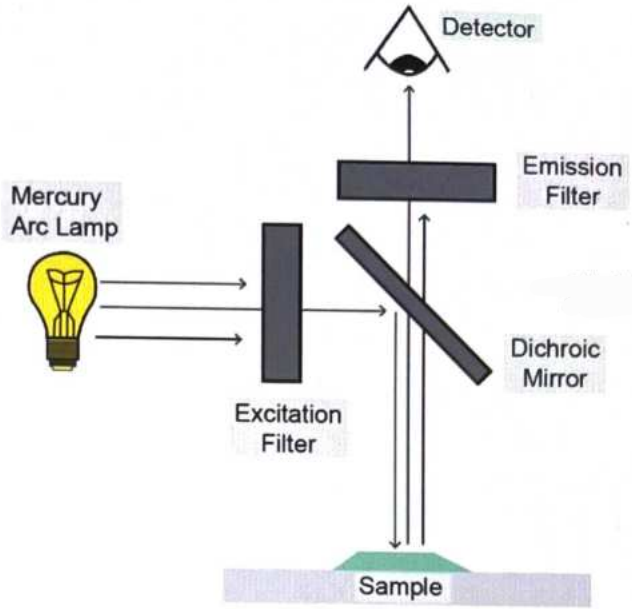
- Light emerges in different phases →Difference in intensity →Reveal internal structures of living cells
- Can be used for motility demonstration

Dark Field Microscopy



- Background is dark by dark Field condensers
- Reflected light is used
- Visualize spirochetes

Fluorescent Microscopy



- When UV rays fall on fluorescent substance, wave length of invisible light increases &Organism becomes Luminous
- Stains
  - Auramine / Rhodamine for →MTB
  - Calcofluor white for →Fungus
  - Acridine orange for →Malarial parasite
    → Acridine orange stains organism Green

Electron Microscope

- Beam of electrons used instead of Light
- Beam focused by circular magnets in vacuum
- Object is held in path of beam
- Maximum resolution can obtained [0.2nm-0.5nm]
- Scatters the electrons by object
- Image is formed on fluorescent viewing screen

BACTERIA

Prokaryotes Vs Eukaryotes 00:50:49

	Prokaryotes	Eukaryotes
• Nuclear membrane, Nucleolus, RNP	• Absent	• Present
• Cell division	• Binary Fission	• Mitosis and meiosis
• Chromosome	• One, circular	• Many, linear
• Cell wall	• NAM, NAG	• Sterols [Mycoplasma also]
• Membrane bound organelles	• Absent	• Present
• Respiration	• Mesosome	• Mitochondria
• Cytoskeleton	• Absent	• Present
• Ribosomes	• 70s	• 80s
• mRNA	• No introns	• Introns & Exons + nt

Shapes 00:57:12

- Cocci in clusters: staphylococcus
- Cocci in chains: streptococcus
- Cocci in pair [diplococcic]: Pneumococcus
- Kidney shaped: Gonococci
- Lens shaped: Meningococci
- Cocci in Tetrads: Micrococci
- Cocci in octet: sarcina
- Spectacle like appearance: Enterococcus

Various patterns observed in Bacilli

- Bamboo stick appearance: Bacillus anthracis
- Diplobacilli: Klebsiella pneumonia
- Branching: Actinomycetes
- Curved GNB: Campylobacter and Helicobacter
- Spiral: Spirochetes
- Pleomorphic: Hemophilus influenzae and Brucella species

Gram staining

- All cocci are Gram positive except
  - Meningococci
  - Gonococci
  - Veillonella
  - Moraxella
- All Bacilli are Gram negative except
  - M - Mycobacterium species
  - A - Anthracis Bacillus
  - C - Clostridium species
  - D - Diphtheriae corynebacterium
  - O
  - N - Nocardia
  - A - Actinomycetes
  - L – Listeria species
  - D–Diphtheroids

How to remember

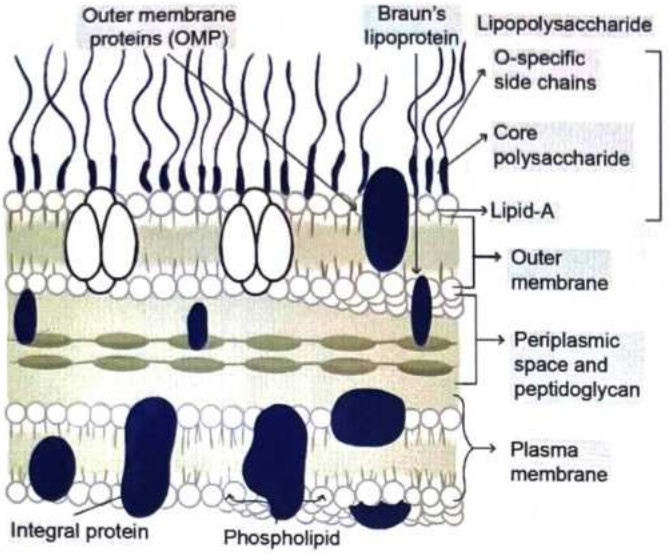
- MAC DONALD

Important Information

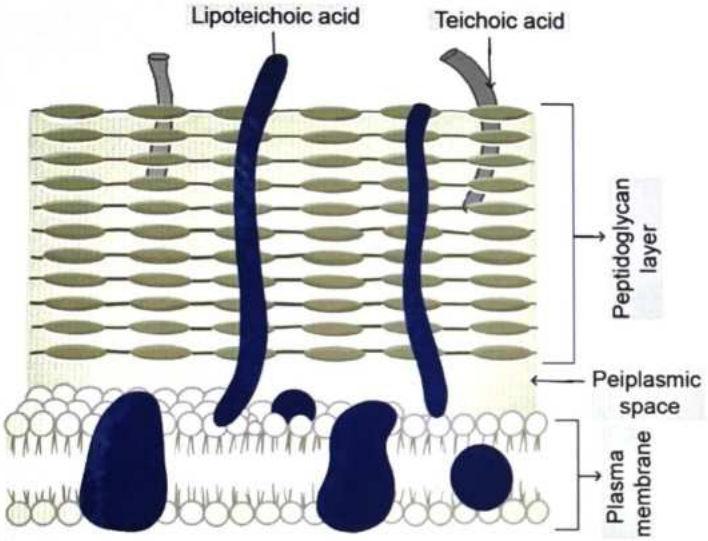
- Diphtheroids: Non pathogenic, are normal commensal of throat
- Corynebacterium diphtheriae: pathogenic and cause diphtheria

Gram Positive vs Gram Negative Bacteria 01:08:23

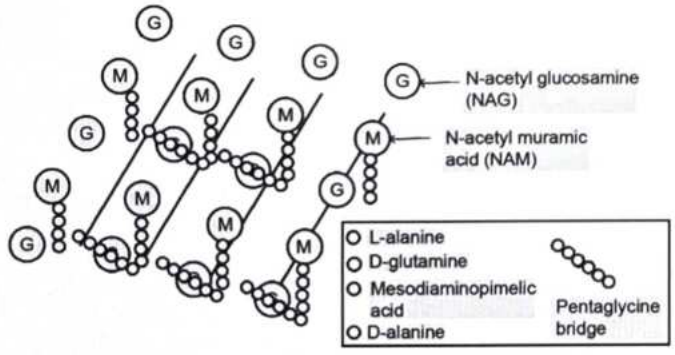
	Gram Positive	Gram Negative
• Thickness	100 layer thick	1-2 layer thickness
• Lipid Content	2-5 %	15-50 %
• Teichoic Acid	Present	Absent
• Variety of AA	Few	Several
• LPS [Endotoxin]	Absent	Present
• Nag-Nam	Tetra peptide is having side chain formed by pentapeptide crossbridge	Tetra peptide are directly linked to each other



Gram negative Bacteria cell wall

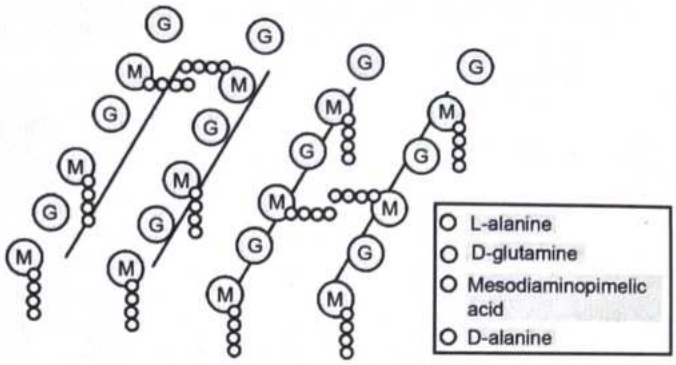


Gram positive Bacteria cell wall



NAG and NAM in Gram Positive Bacteria

- Tetra peptide is having side chain formed by Pentapeptide crossbridge



NAG and NAM in Gram Negative Bacteria

- Tetra peptide are directly linked to each other

Cell Wall Demonstration

- **R** - **R**eaction with specific antibody
- **M** - **M**icro dissection (Mechanical Rupture of cell)
- **P** - **P**lasmolysis
- **D**octors - **D**ifferential staining

How to remember

- RPM Doctors

Inclusions 01:21:40

- Inclusions are the Storage granules of bacteria by which they derive energy
- They play an important role in Corynebacterium diphtheria
  - Storage granules in Corynebacterium diphtheria are K/a Polymetaphosphate aka Volutin granules aka Babes Ernest granules
- Granules are stained by
  - **P** - **P**onders Stain
  - **A** - **A**lbert Stain
  - **N** - **N**eisser Stain

How to remember

- PAN

Capsule 01:23:32

- Mainly made up of Polysaccharide except
  - Polypeptide in Bacillus anthracis
  - Hyaluronic acid in Streptococcus Pyogenes

- Capsulated organism
  - **P**-Pneumococcus, pseudomonas, S. Pyogenes
  - **A**-Anthracis bacillus
  - **K**-Klebsiella species
  - **I**-Influenza Hemophilus
  - **Y**-Yersinia species
  - **B**-Bordetella species
  - **M**-Meningococci
  - **C**-Clostridium perfringens [C. welchii], Cryptococcus neoformans
  - **V**-Vibrio parahemolyticus

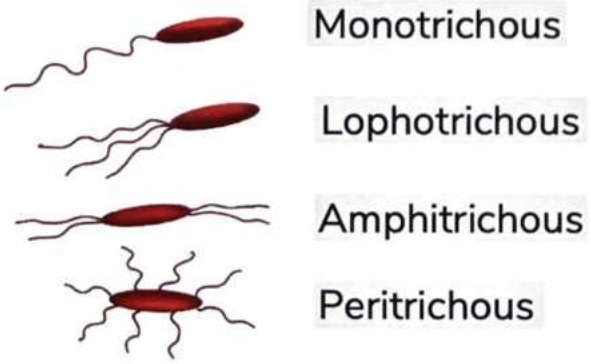
How to remember

- PAKIYB MCV

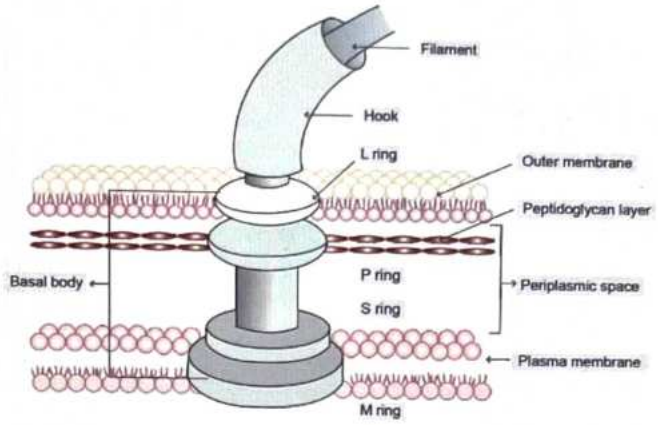
- Function of capsule
  - It avoids Opsonisation thereby avoiding Phagocytosis
  - Modification of capsule k/a Biofilm contribute to Antibiotic resistance and Adherence
- Capsule is Demonstrated by
  - Mc Faydean's Reaction for B. Anthracis capsule
  - Quellung Reaction for Pneumococcal capsule
    - → Quellung reaction is a serological reaction in which the capsular antigen reacts with capsular antibody causing Capsular swelling / Capsular Delineation

Flagella 01:31:31

- Thread like structures which arise from the cell wall
- Made up of a protein K/a Flagellin
- Types
  - Monotrichous: Eg. V. Cholerae, Pseudomonas
  - Amphitrichous
  - Lophotrichous
  - Peritrichous: Eg. E. coli, proteus, Listeria



- Structure of flagella



- Motilities
  - **T** – **T**umbling by Listeria
  - **D** – **D**arting by Vibrio
  - **S** – **S**warming by Proteus
  - **Falling** - **F**alling leaf like by Giardia lamblia [d/t 4 pairs of Flagella]

How to remember

- TDS Falling

- Motility can be demonstrated by
  - Hanging drop method
  - Semisolid Agar method

Fimbria/ Pili 01:39:02

- Helps in Adherence
- Short hair like structure
- Detection of Fimbriae can be done by
  - Hemagglutination: Shown by E. coli, Klebsiella, Gonococci
  - Surface pellicle: thin layer of this Pellicle is produced at the surface of broth

L-Forms

- Cell wall deficient forms
- Named by Klinberger
- Seen in Streptobacillus Moniliformis
- Two Types
  - Unstable L forms
    - → Lose their cell wall in presence of penicillin
    - → They can Revert back so K/a Unstable L-form

→ They are 2 Types
- Protoplast: seen in GP bacteria
- Spheroplast: Seen in GN bacteria

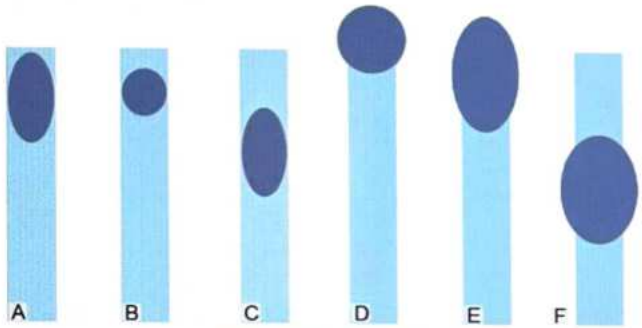
- Stable L-forms: seen in Mycoplasma

Bacterial Spores 01:47:13

- Highly resistant resting Forms
- Dormant or resting form
- Seen in Unfavorable condition
- Bacterial spore comprise of
  - Innermost layer is core followed by cortex, Coat, Exosporium (outermost)

Refer Diagram 17.1

- Types
  - Non bulging terminal and oval, seen in clostridium tertium
  - Subterminal round
  - Central and Oval
  - Terminal spherical and Bulging



Types of spores

Important Information

- **Non-Bulging spores**
  - **Clostridium Tetani: terminal and round**
  - **Most of the clostridia: Subterminal spore**
- **Bulging spore is a feature of**
  - **Bacillus Anthracis**

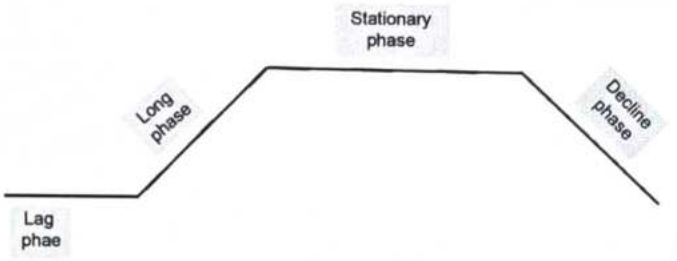
- Demonstration of spores
  - By Gram staining: Spores remain unstained
  - Modified Zn stain: spores are 0.25% Acid fast
  - Special stains
    → Schaeffer Fulton method
    → Moeller stain

- Uses
  - Geobacillus stearothermophilus: Used as Biological control for Autoclave
  - Bacillus subtilis spore: control of Hot air oven

Physiology of Bacteria

- 80% of total bacterial cell is water
- Carbon, nitrogen, hydrogen are also required for growth

Bacterial Growth Curve 02:04:20



- Lag Phase
  - Adapts to the surroundings
  - Accumulation of nutrients and Increases in size
- Log Phase
  - Exponential division occurs
  - Smaller in size
  - Maximum metabolic
  - Best phase for Biochemical reactions
  - Best phase for doing Gram stain
  - $(2)^n$ exponential division
- Stationary Phase
  - Bacterial growth ceases due to depletion of the nutrients
  - Number of progeny cells formed replace the number of cells that dies
  - Total count increase but viable count remains same
  - Start of accumulation of toxic products
  - Sporulation happens in stationary phase
  - Exotoxin production happens
  - Antibiotic production [some bacteria] happens

Previous Year's Questions

Q. **Sporulation occurs in this phase of bacterial growth curve?** (FMGEJun 2019)

A. **Stationary phase**
B. **Lag phase**
C. **Log phase**
D. **Decline phase**

- Decline Phase
  - Complete accumulation of toxic metabolites
  - Involution forms Formed

Factors affecting growth of the Bacteria

- Oxygen
- Co_2 (Capnophiles)
- Temperature
- PH (7.2 to 7.6)
- Light
- Osmotic effect
- Mechanical and sonic stress
- Moisture: Essential requirement

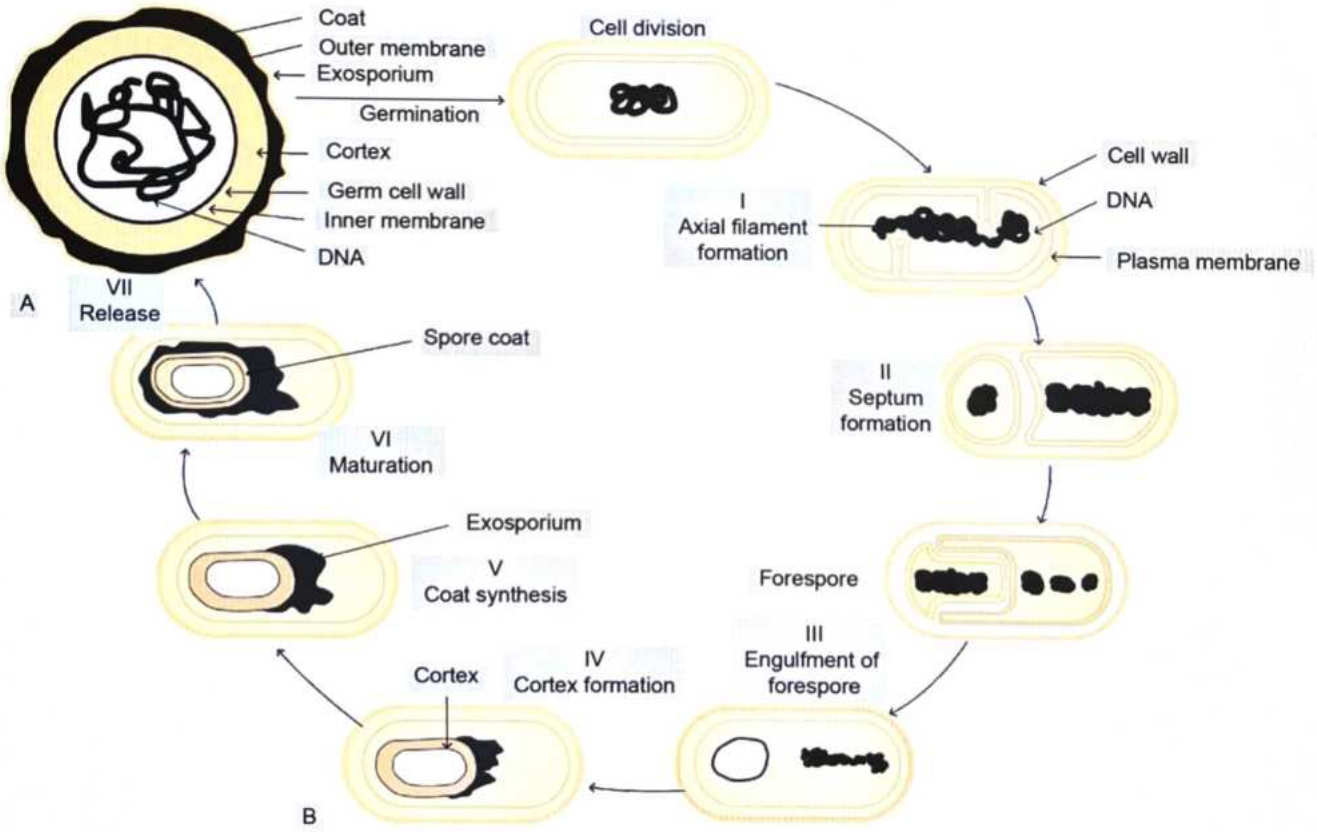
Important terms

- Autotrophs: Can synthesize all organic compounds by using Co_2
- Heterotrops: Have reduced and preformed organic molecule which they use as a carbon source
- Lithotrops: Use inorganic compounds

Metabolism of pathogenic Bacteria

- Aerobic bacteria utilize Glucose by oxidation
- Anaerobic bacteria utilize glucose by fermentation
  - Fermentation happens by
    - → Glycolysis pathway
      - Glucose is converted to Pyruvate
      - Happens in most bacteria
    - → Entner-doudoroff pathway (ED pathway)
      - Rarely seen,
      - Seen in pseudomonas
      - Glucose is converted into KDPG (Keto deoxy Phospho Gluconate)
    - → Pentose phosphate pathway
      - Seen in most of the bacteria
      - Glucose gets converted into pentose sugar
- Oxidation
  - Oxidative utilization of glucose followed by ATP synthesis Via oxidative phosphorylation pathway
  - Oxidation happen by Krebs cycle, ETC
    - → ETC happens in cell membrane of bacteria which behaves like a mitochondria

Diagram 17.1



CLINICAL QUESTIONS

Q.1 A microbiology laboratory assistant wanted to examine biophysical properties of living cell. Which is the specialized type of microscope, which enables quantitative measurement of the chemical constituents of cells?

A. Dark field microscope
B. Interference microscope
C. Phase contrast microscope
D. Fluorescence microscope

Solution:

Interference microscope enables quantitative measurement of the chemical constituents of cells.

Option	Microscope	Features
Option 1	**Dark field microscope**	• Background is dark by dark Field condensers. • Reflected light is used. • Visualise spirochaetes.

Option 2	**Interference microscope**	• Examine a living cell's biophysical properties
		  ○ Stiffness,
		  ○ Time-dependent deformability
		  ○ Mass
		• Does not require labeling with fluorescent proteins or optically active dyes.
		• Both cultured cell lines and material derived from whole organisms can be used.

Option 3	**Phase Contrast Microscope**	• Used to visualize the living cells by creating difference in contrast between the cells and water.
		• Converts slight differences in refractive index and cell density into easily detectable variations in light intensity.
		• It is useful for studying:
		  ○ Microbial motility.
		  ○ Determining the shape of living cells.
		  ○ Detecting bacterial components such as endospores and inclusion bodies.
		  ○ Reveal internal structures of living cells.

Option 4	**Fluorescence microscopy**	When UV rays absorbed on fluorescent substance and reflected, wave length of invisible light increases & becomes Luminous. **Stains**: • Auramine / Rhodamine For → MTB • Calcoflour White For → Fungus • Acridine orange For → Malarial parasite

Q.2 An electron microscope is a microscope that uses a beam of accelerated electrons as a source of illumination. It is used for the investigation of inorganic specimens including microorganisms and macromolecules. Who is the founder of Electron microscope?

A. Antonie Philips van Leeuwenhoek
B. Ernst Ruska
C. Robert Koch
D. Louis Pasteur

Solution:

- **Ernst August Friedrich Ruska** was a German physicist who won the Nobel Prize in physics in 1986 for his work on Electron optics, including the **design of the first Electron microscope.**

Electron microscope:

- Beam of electrons used instead of Light
- Beam focussed by circular magnets instead of lens
- Object is held in path of beam
- Scatters the electrons by object
- Image is formed on fluorescent viewing screen
- Maximum resolution can obtained [0.2nm-0.5nm] (100 times better than light microscope)
- Vacuum is used

18 GENERAL MICROBIOLOGY PART-2

STERILIZATION & DISINFECTION

Definitions 00:00:39

- **Sterilization**
  - Making an article free of pathogens, non pathogens including spores
- **Disinfection**
  - Removal of pathogens & non pathogens but not spores
- **Asepsis**
  - Process in which the chemical agents [antiseptic] applies to body surfaces will Kill/ inhibit pathogenic microorganisms

Resistance in Decreasing Order

- Prions > Cryptosporidium oocysts > Bacterial spores > Mycobacteria > Giardia cyst > Small non-enveloped virus > Trophozoites > Gram negative bacteria >Fungi>Large non-enveloped viruses> Gram positive bacteria > Enveloped viruses
- Most resistant: Prions
- Least resistant: Enveloped viruses

Sterilization methods

Dry heat sterilization

- Includes
  - Flaming: For Inoculating loops
  - Incineration
    - → Temperature: 1200° C
    - → Anatomical, Microbiological and hospital waste
    - → In biomedical waste management yellow bag is incinerated
  - Hot air oven
    - → It is an electrically heated chamber with a fan
    - → Temperature: 160°C x 2hrs
    - → Materials sterilized are
      - Glass wares: syringes, petridish, flask
      - Surgical instruments like scalpels and scissors
      - Dusting powder, grease and paraffin
    - → Precautions to be taken while handling Hot Air Oven
      - Overloading should be avoided
      - Equal and free circulation of air should be maintained
      - Material should be dried
      - Paper wrapping of items should be done
      - Oven should be allowed to cool before opening
    - → Controls used
      - Spores of bacillus subtilis or spores of Non pathogenic Cl. tetani
- Mechanism
  - **C** – **C**harring
  - **O** – **O**xidative damage
  - **D** – **D**enaturation
  - **E** – **E**levated electrolytes

How to remember

- CODE

Moist Heat Sterilization

- Mechanism: Coagulation and Denaturation of proteins

• <100°C	• Pasteurization • Inspissation
• =100°C	• Tyndallization • Boiling
• >100°C	• Autoclave

1. At Less than 100°C

a. Pasteurization of milk 00:17:43

- Flash method
  - 72°C For 15-20 sec and immediately bring down temperature to 13°C
  - Kills Coxiella burnetti spores also
  - Better method than Holder's
- Holder's method
  - 60°C for 30 min

b. Water bath

- Disinfection of serum, Body Fluids & vaccines can be sterilized
- Bacterial vaccines: disinfected at 60°C x 1 hr

c. Inspissation 00:19:57
- 80-85°C for 20-30 minutes for 3 days
- On 1st day: pathogenic forms get killed
- 2nd and 3rd day: spores germinating also get Killed
- Done For
  - LJ [Lowenstein Jensen] Media
  - LSS [Loffler's Serum Slope]: For Diphtheriae

2. At 100°C

a. Boiling
- 100°C for 15 minutes
- Most of vegetative Forms Killed
- Spores are not killed

b. Tyndallization
- 100°C x 20-30 min x 3 Days
- Done for Sugar& Gelatin containing Substances

3. More than 100°C

a. Autoclave 00:22:59
- Water boils when its vapour pressure equals surrounding atmospheric pressure
- As atmospheric pressure increases, Boiling temperature also increases
- At normal pressure, boiling point of water is 100°C
- Parts of autoclave
  a. Pressure chamber
  b. Lid
  c. Electrical heater
- Temperature of autoclave
  - M/c: 121°C x 15 min x 15 psi [Pounds square inch]
  - 126°C x 10 min x 20 psi
  - 133°C x 3 min x 30 psi
- Done for
  - Surgical instruments
  - All culture medias except LJ & LLS
  - Plastic tubes & pipette tips
  - Sputum samples before discard should be Autoclaved
  - Biohazard waste
  - Surgical Suturesexcept catgut
- Controls
  - Spores of Geobacillusstearothermophillus
- Chemical indicators
  - Class I: Autoclave tape (External pack control)
  - Class II: Bowie dick Test (Equipment control)
  - Class IV/V: Internal pack control
- Physical control: Temperature, pressure & time

Filtration 00:33:13
- Mainly done For Heat sensitive substances
  - Vaccines
  - Antibiotics
  - Serum
  - Toxin
  - Sugar solution
- Types of filters
  - Depth Filter: includes
    → Candle Filters
    → Seitz Filter
    → Sintered glass filter
  - Membrane Filters
    → Most widely used filters for bacterial Filtration
    → Made up of cellulose acetate, cellulose nitrate
    → Pore size
      - 0.22 µ: M/c used
      - 0.45 µ: Retains Coliforms
      - 0.8 µ: Retains air micro organisms
- Air filters
  - HEPA [High efficiency particulate air]
    → Removes 99.97% of particles having a size 0.3µ or more
  - ULPA [Ultra Low Particulate]
    → Removes 99.999% of dust, pollen, molds
- Control: Brevundimonas dimunita & serratia marcesens (produce pinkish colonies)

Radiation 00:39:49
- Types
  - Ionizing Radiation / Cold Sterilization
    → Consist of Gamma rays, X Rays, and cosmic rays
    → Breaks upon DNA without temperature rise so k/a cold sterilization
    → Used to sterilize
      - Plastic syringes
      - Catgut sutures
      - Bone and tissue grafts
  - Non Ionizing Radiation / Hot Sterilization
    → Consist of UV rays and Infrared rays
    → Temperature of surface increases so k/a Hot

sterilization

→ Used for

- Biosafety cabinets
- OTs

- Control: Bacillus pumilus

Chemical Methods of Sterilization 00:43:05

1. Alcohols

- Most widely used disinfectants & antiseptics
- Ethanol & Isopropyl alcohol (M/c used)
- 70-80% concentration acts best
- It is having a Bactericidal & Fungicidalaction but not sporicidal
- Ethyl alcohol used as Surgical spirit [70%]
- Isopropyl alcohol is used for stethoscope & thermometer x 10-15 min

2. Aldehyde

- MOA
  - Combine with nucleic acids, proteins & inactivate them by cross linking & alkylating the molecules
  - Are sporicidal [used as chemical Sterilants]

a. Formaldehyde

- Present in 2 Forms
  - Gas: used For OT Fumigation
  - Liquid [formalin]: used to preserve anatomical & pathological specimens
- Used for Duckering: done to avoid pulmonary Anthrax

b. Glutaraldehyde

- Less toxic, less irritant
- 2% Glutaraldehyde(Cidex): Used For Bronchoscopes, , endoscopes, colonoscope, proctoscope
- Available in inactive form & activated by alkalinisation before use
- Remains active only for 14 days

Previous Year's Questions

Q. Glutaraldehyde is used for all the following except? (FMGE Nov 2017)

A. Bronchoscope
B. Thermometer
C. Proctoscope
D. Endoscopic tube

Previous Year's Questions

Q. Which of following high level disinfectant? (FMGE Dec 2020)

A. Alcohol
B. Phenol
C. Lysol
D. Glutaraldehyde

c. Orthophthaldehyde

- Available in 0.55% Solution
- Can also be used for sterilizing cytoscopes & Endoscopes
- Advantages
  - Does not require activation
  - Better odour
  - More stable
  - ↑ mycobactericidal activity

3. Phenol Compounds

a. Phenol / Carbolic acid

- 1st Widely used antiseptic
- Discovered by Joseph Lister in 1867
- Derived from coal tar
- Mechanism: denature proteins & disrupts cell membranes
- Advantage
  - High mycobactericidal activity
  - Can act upon in pressure of organic waste
- Cresols, Xylenol, Lysol are various forms of phenol used as disinfectants in labs & Hospitals
- Retain activity in the presence of organic waste
- Chlorhexidine: components of Savlon [Chlorhexidine + Cetrimide]
- Chloroxylenol: component of Dettol

4. Halogens

- Includes Iodine & chlorine, have antimicrobial activity
- Exist in free state & Form salts when they combine with Sodium & most other metals

a. Iodine

- Skin antiseptic
- Oxidizing cell contents& iodinizing cell proteins
- At higher concentration, it may kill spores
- Tincture of iodine: 2% Iodine in water Ethanol Solution of Potassium Iodide
- Iodophor: Combining iodine with organic carrier like

povidone (povidone iodine/Betadine)

b. Chlorine

- Various preparations include
  - Chlorine gas, Sodium hypochlorite, Calcium hypochlorite
  - They form Hypochlorous acid which causes oxidation of cell contents
  - Organic matter interferes with its action

Previous Year's Questions

Q. Blood on OT floor is cleaned by? (AIIMS Nov 2017)

A. Phenol
B. Alcohol based components
C. Chlorine based components
D. Quaternary ammonium compounds

5. Oxidizing Agents

- Includes

a. H_2O_2

b. Peracetic Acid

c. Plasma Sterilization

a. H_2O_2

- Strong oxidizer [high level disinfectants]
- Liberates toxic free hydroxyl radicals which are active ingredients
- Attack cell membrane, DNA,& other cellular components
- Most effective concentration: 3-6%
- To kill Spores: 10% [higher concentration]

b. Peracetic Acid

- Powerful oxidizing agent
- More active than H_2O_2
- Concentration of < 1% is sporicidal

c. Plasma sterilization 01:03:33

- Plasma Refers to gaseous state consisting of ions & free electrons
- Chemical Sterilants H_2O_2& Peracetic acid used to induce Plasma state
- Sterrad: Plasma sterilizer used to producevacuum inside the chamber
- Active agents: UV photons & radicals used
- Maintain low temperature
- Used in sterilization of surgical instruments
- Control: Bacillus Stearothermophillus spores

Previous Year's Questions

Q.STERRAD is a gas plasma sterilizer used in operation theatres for disinfecting OT articles. Which of the following is the active agent used in it? (AIIMS – Nov - 2018)

A. H2O2
B. N2O
C. Ozone
D. EIO

6. Heavy metal salts

- Combine with bacterial cell proteins
- Includes
  - Silver sulfadiazine: used for Burns
  - Silver nitrate [1%]
  - Copper sulphate: used as Fungicidal
  - Mercurochrome: Known antiseptics

7. Surface Active Agents or Surfactants 01:08:57

- Lower surface tension b/w 2 liquids or b/w liquid & solid
- Classified into
  - Anionic
  - Cationic
  - Amphoteric

a. Cationic surfactants

- Quaternary Ammonium compounds are most popular cationic disinfectants
- Have positively charged quaternary nitrogen
- They disrupt the microbial membranes
- Have an Antimicrobial activity
- More active against Gram positive bacteria than Gram negative bacteria but not MTB or spores
- Eg. Cetrimide is a cationic surfactant

b. Anionic surfactants

- Have strong detergent action
- Have weak antimicrobial action
- Most active at acidic PH

c. Amphoteric (Tego compounds)

- Have detergent property of anionic compounds
- Have Antimicrobial property of cationic compounds
- Used as a antiseptic in dental practices
- Can act upon wide range of PH

8. Dyes

- Types: Aniline & Acridine
- Used as skin & wound antiseptics

a. Aniline dyes

- Includes
  - Crystal violet
  - Gentian violet
  - Malachite green
- More active against Gram positive than Gram negative
- But no activity against MTB
- Reduced activity in presence of organic waste
- Interferes with synthesis of Peptidoglycan in cell walls

b. Acridine dyes

- Includes
  - Acriflavine
  - Euflavine
  - Proflavine
  - Aminacrine
- They are affected very little by organic waste
- More active against Gram positive than Gram negative

9. Ethylene oxides (ETO)

- Most widely used gaseous chemical Sterilant
- Both microbicidal& sporicidal
- Acts by combining with cell proteins
- High penetrating power
- Used For pre packed syringes
- Highly inflammable, irritant & explosive
- Mixed with CO_2 or dichlorodifluoromethane
- 3 Factors affect ETO disinfectant power
  - ETO concentration
    - → At ETO concentration of 700 mg/l & 40-50% relative humidity at 38°C sterilization is achieved in 5-8hrs
  - Humidity
  - Temperature
- Control: Bacillus globigi spores used
- Uses: to sterilize
  - Disposable petridishes
  - Pre packed syringes
  - Heart Lung machines
  - Respirators
  - Catheters
  - Sutures
  - Dental equipments

10. Beta Propiolactone

- 0.2% is active against all micro-organisms including spores
- Used For inactivation of vaccines
- Found to be carcinogenic, so not used

Testing of Disinfectants 01:22:04

1. Phenol Coefficient Test/ Rideal Walker Test

- Determined by dilution of disinfectant
- A fixed concentration of salmonella typhi broth is used
- Phenol Coefficient Test = $\frac{\text{Highest dilution of test disinfections that kills S. typhi in given time}}{\text{Highest dilution of phenol that Kills S. typhi in given time}}$
- It doesn't assess disinfectant in presence of organic waste

2. Chick martin test

- Checks Whether Disinfectant works in presence of organic waste [feces, yeast]

3. Capacity test / Kelsey sykes test]

- Capacity of a disinfectant to act upon increasing microbiological load

4. In Use Test / Kelsey & Maurer Test

- Disinfectant is microbiologically contaminated
- Routinely performed in hospitals once in 3 months

Sporicidal Agents

- **E** - **E**thylene oxide
- **F** - **F**ormaldehyde
- **G** - **G**lutaraldehyde [2%]
- **H** - Sodium **H**ypochlorite [1% sodium hypochlorite is used for blood spills]
- Phthalic Acid
- Peracetic acid
- Autoclave
- Hot air oven
- Plasma sterilization

Chemical Disinfectants – Classification

Refer Table 18.1

Spaulding Classification

Critical devices

- Includes those Enters sterile sites like Surgical instruments, catheters
- High level disinfection needed

Semi-critical devices

- Includes those comes in contact with mucous

membranes like respiratory equipments [laryngoscope]
- High level disinfections needed

Non- critical devices
- Includes those come in contact with skin like BP cuff, ECG Electrodes
- Low/intermediate level disinfection needed

Medical equipments
- Includes Examination tables, computers
- Low level disinfections needed

Table 18.1

Germicide and their concentrations	Level of disinfectant	Bacteria and enveloped viruses	Fungi	Un-enveloped viruses	M. tuberculosis	Spore	Inactivated by organic matter
Glutaraldehyde (2%)	High/CS	+	+	+	+	+	-
Formaldehyde (3-8%)	High/CS	+	+	+	+	+	-
H_2O_2 (3-25%)	High/CS	+	+	+	+	+	+/-
Chlorine (100-1000 ppm of free chlorine)	High	+	+	+	+	+/-	+
Isopropyl alcohol (60-95%)	Intermediate	+	+	+/-	+	-	+/-
Phenol (0.4-5%)	Intermediate	+	+	+/-	+	-	-
Chlorhexidine gluconate (2-4%)	Low	+	+/-	+/-	+/-	+/- (static)	+
Iodophore (30-50 ppm of free iodine)	Intermediate	+	+	+	+/-	-	+
Quaternary ammonium compounds (0.4-1.6%)	Low	+	+/-	-	-	-	+

CLINICAL QUESTIONS

Q.1 A microbiology student was learning to disinfect and sterilize as it is needed in the laboratory to set things free from microorganisms. Which of the following sterilization/disinfection agents works by liberating free hydroxy radicals?

A. Formaldehyde
B. Hydrogen peroxide
C. Heavy metals
D. Ethylene oxide

Solution:

- **Hydrogen peroxide** works by liberating free hydroxy radicals.

Mechanism of action of aldehydes:

- **combine with nucleic acids, proteins & inactivate** them by cross linking & alkylating the molecules
- also sporicidal [used as chemical Sterilants]

Option 1

FORMALDEHYDE:

FORMS:

1. GAS → used For OT Fumigation
2. LIQUID [FORMALIN] → used to preserve anatomical & pathological specimens

Other aldehydes used are:

- **Glutaraldehyde**
- **Orthophthaldehyde**

Option 2

Hydrogen peroxide (H_2O_2)

- Strong oxidizer [high level disinfectants]: **Liberates toxic free hydroxyl radicals** which are active ingredients.
- **Attack membrane, DNA, lipid & other cellular components**
- Most effective concentration → 3-6%
- Spores might require → 10% [higher concentration]

Other OXIDIZING AGENTS:

- Peracetic Acid: concentration < 1% is sporicidal
- Plasma Sterilization

Option 3

Mode of action of Heavy metals:

- Heavy metals combine with bacterial cell proteins, often with their sulphydryl groups, and inactivate them.
- They may also **precipitate cell proteins.**

→ most heavy metals are **bacteriostatic.**

Examples:

- SILVER SULFADIAZINE [used For Buurns]
- SILVER NITRATE [1%]
- COPPER SULPHATE [Fungicidal]
- MERCURY SALTS:
  - MERCUROCHROME → Known antiseptics
  - THIOMERSAL → preservatives in vaccine & sera

Option 4

ETO:

- Ethylene oxide sterilizer is one of the widely used gaseous chemical sterilants in present days.
- It has **high penetration power**
- Has both microbicidal and sporicidal activity
- **Acts by combining with cell proteins.**
- highly inflammable, irritant & explosive
- mixed with CO_2 or dichlorodifluoromethane.

Q.2 Pasteurization is a technique in which heat is applied to liquids to kill potential agents of infection and spoilage, while at the same time retaining the liquid's flavour and food value. Which of the following is true about the holder's method of pasteurization?

A. It kills all bacteria and spores
B. It kills all bacteria except Coxiella burnetti
C. It kills 95% of microorganisms
D. All bacteria are destroyed

Solution:

- Pasteurization kills nearly 90% of bacteria in milk **except for certain bacteria like Coxiella burnetti** which is resistant to Holder's method of sterilization but **killed by Flash method** of Sterilization.

Pasteurization:

- Pasteurization is a technique in which heat is applied to liquids to kill potential agents of infection and spoilage, while at the same time retaining the liquid's flavour and food value.
- Pasteurization is an example of sterilization at a temperature **<100°C.**

Methods:

Flash method:

- 72°C For 15-20 sec → bring down to 13°C
- Kills Coxiella burnetti spores also
- Better method than Holder's method.

Holder's method → 60°C For 30 min

Q.3 A microbiologist wants to sterilize heat sensitive liquids like serum, vaccines, antisera, enzymes, antibiotic solutions. Which of the following is best to sterilize heat labile solutions?

A. Dry heat
B. Autoclave
C. Membrane filtration
D. Pasteurization

Solution:

- Heat sensitive liquids like serum, vaccines, antisera, enzymes, antibiotic solutions and urea solutions can be sterilized by using **membrane filtration.** The filtration can be aided by using either positive or negative pressure.

2 TYPES OF FILTERS:

1. **DEPTH FILTERS** – POROUS FILTERS [metal polymer with random mats]

Examples:

- Candle Filters
- Unglazed porcelain Filters
- Seitz Filter
- Sintered glass filter

2. **MEMBRANE FILTERS**

- Most widely used filters For bacterial Filtration
- Made up of cellulose acetate, cellulose nitrate, polycarbonate
- Pore size:
  - 0.22 μ → most common used
  - 0.45 μ → retains coliforms
  - 0.8 μ → retains airbone micro organisms

USE → to sterilize sugar, sera & antibiotic solutions

AIR FILTERS:

1. **HEPA** [High efficiency particulate air] FILTER → Removes 99.97% of particles having a size 0.3μ or more.
2. **ULPA** [Ultra Low Particulate/Penetration] → Removes 99.999% of dust, pollen, molds.

CONTROL → Brevundimonas diminuta & Serratia marcesens

19 GENERAL MICROBIOLOGY PART-3

BACTERIAL GENETICS

- **DNA:** Made of
  - De oxy ribose
  - Phosphate groups
  - 4 nitrogenous bases
    - → 2 purines: Adenine & Guanine
    - → 2 pyrimidines: Thymine & cytosine
  - Strands held together by hydrogen bonds

- **RNA**
  - Made up of
    - → Ribose sugar
    - → Uracil replaces the Thymine
  - Types
    - → mRNA [messenger RNA]
    - → tRNA [Transfer RNA]
    - → rRNA [Ribosomal RNA]

- **Gene:** Segment of DNA that specifies for a particular polypeptide
- **Codon**: Genetic information stored in DNA
- **Anticodon**: Binds to codon
- **Non sense codon:** UAA, UAG, UGA
  - Don't code for any amino acid
  - Terminate the synthesis of polypeptide
  - Aka stop codons

Extra Chromosomal Genetic Materials (Plasmids) 00:05:19

- Present free in cytoplasm of bacteria
- When they integrate with host cell its K/a Episome
- Not essential for the Survival of bacteria
- Plasmid Encode genetic information for
  - Resistance to antibiotics
  - Bacteriocins
  - Enterotoxin production
  - Enhanced pathogenicity
  - Degrading complex organic molecules

Bacterial Variations 00:08:44

Phenotypic	Genotypic
• Physical expression in environment	• Sum total of genes or Genetic Apparatus of the cell
• Examples ○ Typhoid Bacilli → Normally flagellated → In phenol agar flagella is lost ○ Lactose Fermentation (E.coli, Klebsiella) by β galactosidase	• Examples ○ Mutations ○ Transfer of genes by → Conjugation → Transformation → Transductions

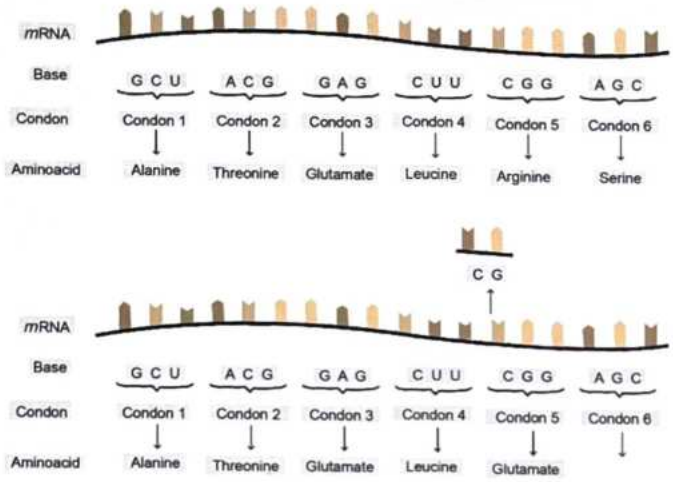
Mutation 00:13:07

- Random, undirected, heritable variation
- Alternation of nucleotide sequences happens, by
  - Addition
  - Deletion
  - Substitution [one or more bases]
- Bacteria undergo mutationsat 10^{-2} to 10^{-10} per bacterium per division
- Mutagenic agents
  - UV Rays
  - Alkylating agents
  - Acridine dyes
- Lethal mutation: Harmful, Destroys the vital function
- Conditional lethal mutants
  - Example: Temperature sensitive mutants
    - → Can lives at 35°C
    - → But dies at 39°C [Restrictive temperature mutants]
- Types
  - Point mutation
    - → Base pair substitution
    - → Frame shift mutation
  - Multisite Mutations: can happen due to some Gain, Loss or Duplication can occur

Point mutation 00:19:57

Transition	Transversion
• Mc type • Purine is replaced by purine • Pyramidine is replaced by pyramidine	• Purine can be replaced by • Pyramidine &Vice Versa

- Frame Shift mutation
  - Deletion or addition of purine or pyramidine causing change in entire frame



Multi-Site Mutations 00:23:55

- Happens by
  - Addition/ Gain
  - Deletion/ Loss
  - Duplication
  - Inversion

Missense Mutations

- Triplet code is altered, different amino acid is being coded

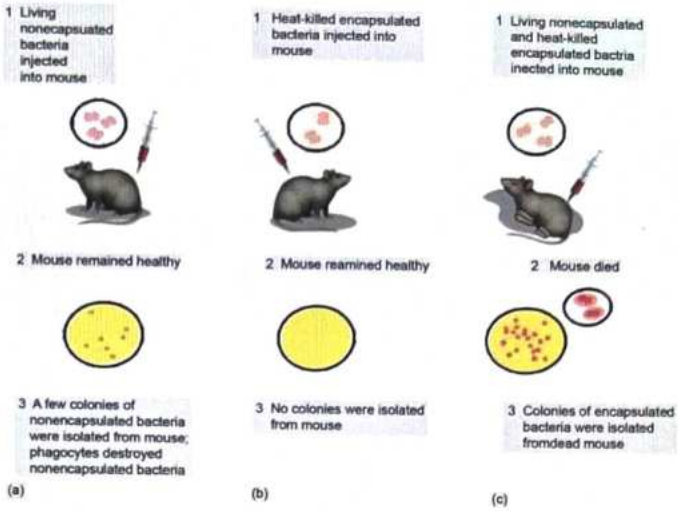
Nonsense mutations

- Premature polypeptide chain termination by stop codon

Gene Transfers 00:26:35

1. Transformation

- Discovered by Griffith
- Rediscovered by Avery, McLeod, and Mc Carty
- Experiment of Griffith

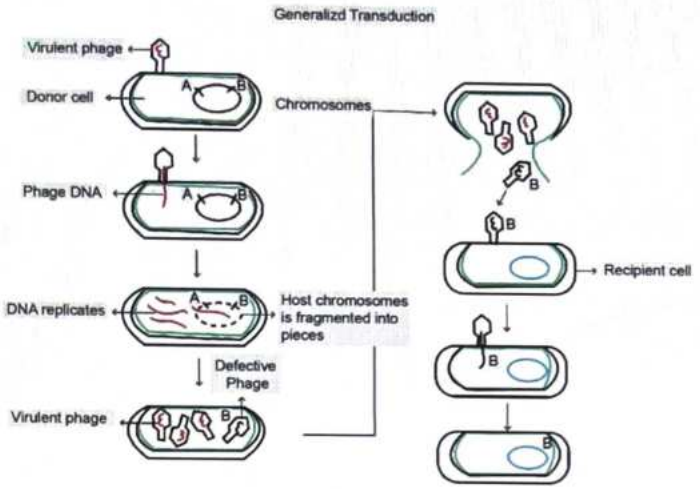


- Case A
  - → Living capsulated pneumococcus when injected into mouse → mouse dies
- Case B
  - → Heat Killed encapsulated pneumococcus when injected into mouse → No effect
- Case C
  - → Heat Killed capsulated pneumococcus + Live rough strains of Pneumococcus when injected into mouse → Mouse dies
  - → On Autopsy Colonies of living capsulated pneumococcus are isolated from dead mouse [through agency of free DNA]

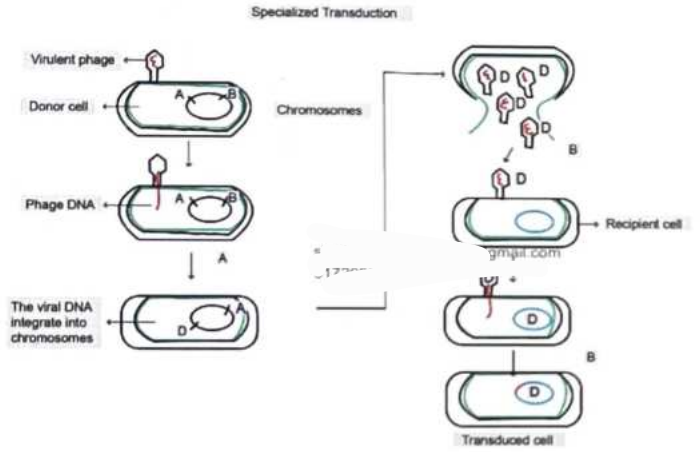
2. Transduction 00:31:22

- Bacteriophage mediated transfer DNA b/w cells
- Types
  - Lytic or Virulent Cycle
    - → Bacteriophage multiplies in host cytoplasm & produce large no. of daughter phages
    - → Subsequently gets released, causing death & lysis of host cycle
  - Lysogenic or Temperate Cycle
    - → Host bacterium unharmed
    - → Phage DNA integrate with bacterial chromosome as prophage that multiplies Synchronously with bacterial DNA
    - → When phage DNA tries to come out it is disintegrated From host chromosome, comes out in cytoplasm & behaves like a lytic phage;
    - → It replicates to produce daughter phages which subsequently released by host cell lysis

- Generalised transduction
  - Involves transfer of any part of donor bacterial genome to recipient bacteria
  - Occurs as a result of defective assembly during the lytic/ virulent cycle & sometimes temperature phages



- Specialized / restricted transduction
  - Part adjacent to integration of phage DNA with host cell DNA

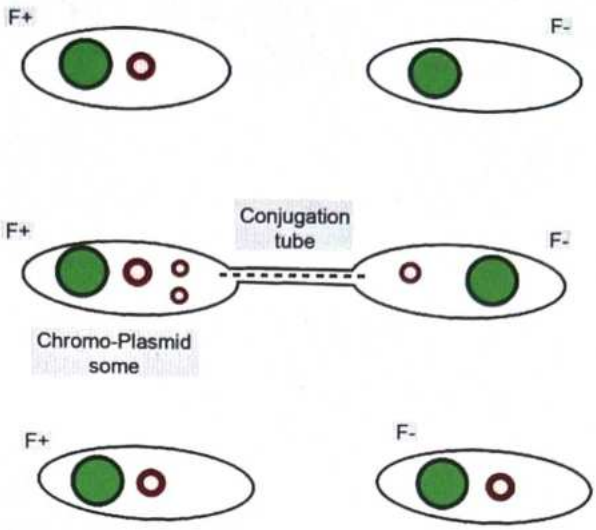


- Bacterial toxins that are coded by Lysogenic phages
  - A - A & C Pyrogenic toxins of Strepto Pyogenes
  - B - Botulinum toxin C & D
  - C - Cholera toxin
  - D - Diphtheria toxin
  - E - EHEC [Entero Haemorrhagic E. coli] aka Verotoxin]

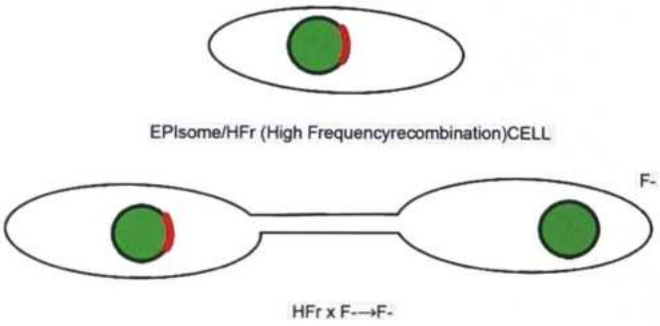
3. Conjugation

00:47:24

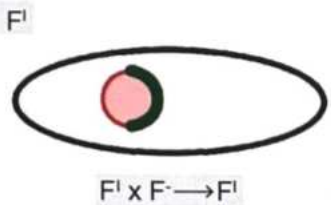
- MC method of resistance transfer in bacteria
- Possibility 1



- Possibility 2
  - Episome / HFR [High Frequency Recombination] cell



- Possibility 3



- Possibilities

F^+	x	F^-	→	F^+
HFr	x	F^-	→	F^-
$F^'$	x	F^-	→	$F^'$

Transferable drug resistance	Mutational drug resistance

• More dangerous as it can lead to MDR
• Resistance = RTF + r
  ○ RTF – Resistance transfer Factor
  ○ r – Resistant determinant [main Factor]
• Difficult to treat
	• Single point mutation leading to single drug resistance
	• Easy to treat

- In MTB, mc method of resistance is Mutational drug resistance

Transposons/Jumping Genes 01:00:13

- They are responsible for intracellular transfer
- Transfer happens b/w
  - Chromosome → chromosome
  - Chromosome → Plasmid
  - Plasmid → Plasmid
- Work in a cut & paste manner
- Have inverted repeat sequences & insertion sequence
- Method is Known as Transposition
- Resistance is transferred mainly with transposons
- Eg. VRE (Vancomycin resistant Enterococci) produces Transposons due to which there is transfer of resistance to the staph Aureus as a result these Staph Aureus change into VRSA

CLINICAL QUESTIONS

Q.1 Transduction is the most common method of resistance transfer in Staphylococcus aureus. Which of the following is involved in the process of bacterial transduction?

A. Plasmids
B. Sex pili
C. Bacteriophage
D. Uptake of genetic material by other bacteria.

Solution:

- **Bacteriophage** is involved in the process of bacterial transduction.
- **Transduction** – Transduction is defined as transmission of a portion of DNA from one bacterium to another by a bacteriophage.

LYTIC OR VIRULENT CYCLE:

- Bacteriophage multiplies in host cytoplasm & produce large no. of daughter phages.
- Subsequently gets released, causing death & lysis of host cycle.

LYSOGENIC OR TEMPERATE CYCLE:

- Host bacterium unharmed.
- Phage DNA integrate with bacterial chromosome as prophage that multiplies synchronously with bacterial DNA.
- When phage DNA tries to come out it is disintegrated from host chromosome, comes out in cytoplasm & behaves like a lytic phage.
- It replicates to produce daughter phages which subsequently released by host cell lysis.

Q.2 An 86-year-old male who is a known case of DM x 3 years presented to ER with a complaint of a non-healing foot ulcer. The patient also complained that he is been taking different antibiotics for a long time for his foot ulcer but the medications had no results. The doctor suspecteddrug resistance. Which of the following statements is false regarding drug resistance?

A. In mutational drug resistance one drug resistance at a time is seen
B. Virulence is decreased in transferable resistance
C. Drug combinations can prevent Mutational drug resistance
D. High degree of resistance is seen in transferable drug resistance

Solution:

- Virulence is **increased** in transferable resistance

Mutational drug resistance	Transferable drug resistance
One drug resistance at a time	Multiple drug resistance
Low degree resistance	High degree resistance
Can be overcome by high drug dose	High dose ineffective
Drug combinations can prevent	Combinations cannot prevent
Resistance does not spread	Spreads to same or different species
Mutants may be defective	Not defective
Virulence may be low	Virulence not decreased

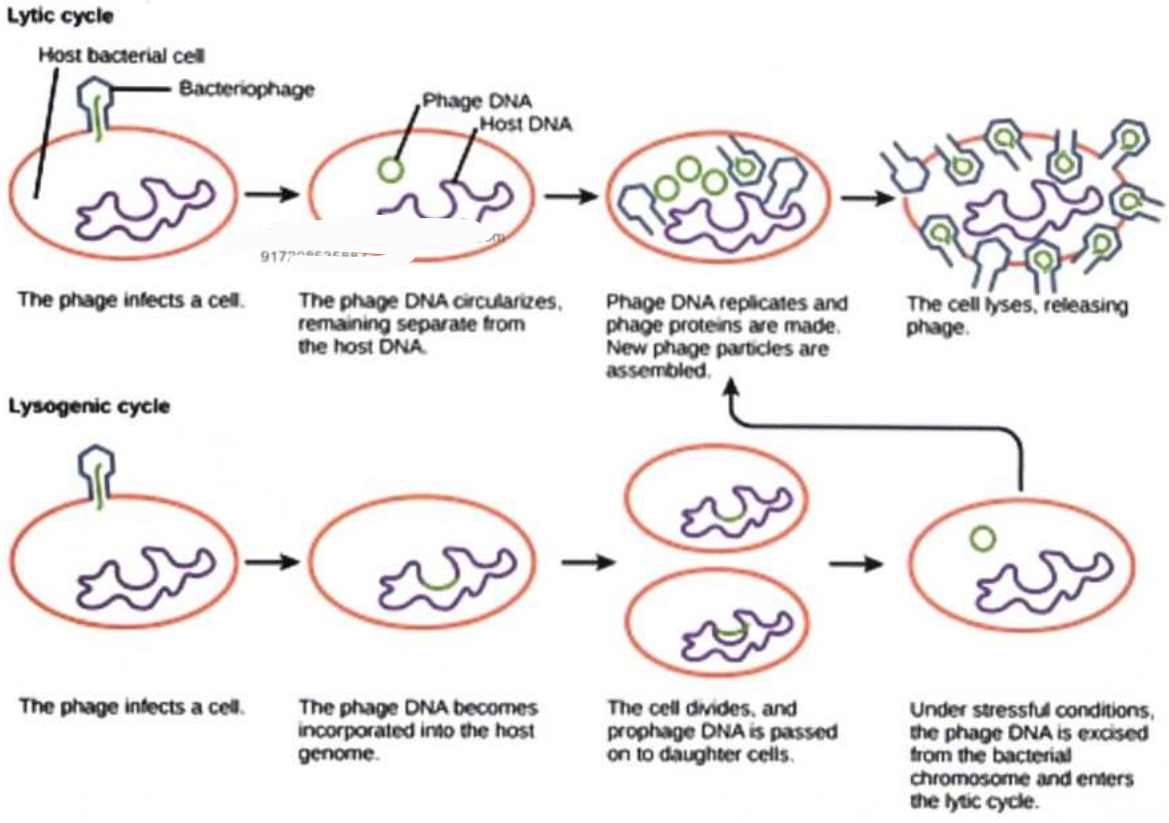
Transferable drug resistance:

- Example of conjugational transfer
- R factor (or the resistance factor) is a plasmid which has two components.
  - Resistance transfer factor (RTF) is the plasmid responsible for conjugational transfer(similar to F factor)
  - Resistance determinant (r): Codes for resistance to one drug. An R factor can have several r determinants.

→ RTF + r = RESISTANCE

→ Leads to MDR [multidrug resistance] → difficult to treat

Mutational drug resistance → SINGLE DRUG RESISTANCE → EASY TO Rx



TYPES OF TRANSDUCTION:

Generalized transduction:

- Involves transfer of any part of donor bacterial genome to recipient bacteria.
- Occurs as a result of defective assembly during the lytic/virulent cycle & sometimes temperature phages.

Specialized transduction: part adjacent to integration of phage DNA with host cell DNA.

LEARNING OBJECTIVES

UNIT 8: CLINICAL MICROBIOLOGY

- Normal microbial flora
  - Introduction, normal flora, medically important members of normal flora

- Bone and joint infection
  - Osteomyelitis, infectious septic arthritis, viral or immune complex mediated arthritis, reactive arthritis, rheumatic fever

- GIT infections
  - Esophagitis, Gastritis, Diarrhea, Appendicitis
  - Diverticulitis, Enteric fever

- Urinary tract infections
  - Predisposing Factors of UTI, Lowe UTI, Upper UTI, Treatment of UTI

- Pelvic infections
  - Genital Ulcer, Prevention strategies
  - Vaginitis, Cervicitis, Pelvic Inflammatory Disease
  - Urethritis, Prostatitis

- Cardiac infections
  - Endocarditis, Myocarditis, Pericarditis: pathophysiology, diagnosis and treatment

- Upper respiratory tract infections
  - Otitis media, Sinusitis
  - Pharyngitis, common cold, Croup
  - Laryngitis
  - Epiglottitis

- Lower respiratory tract infections
  - Bronchitis
  - Bronchiolitis
  - Pneumonia
  - Lung abscess

- CNS infections
  - Meningitis, Encephalitis
  - Brain abscess
  - Subdural empyema and Epidural empyema

- Blood stream infections
  - [illegible]ological agents, Types, Clinical manifestations

- Fever of unknown origin
  - Criteria
  - Investigation
  - Systemic Bacterial Infections, Viral infection, Parasitic Infections and Fungal Infections a/w FUO

- Anemia causing organisms
  - Iron Deficiency, Hemolytic, Megaloblastic and Aplastic Anemia

- Eye and ear infections
- Skin lesions
  - Primary skin lesions
  - Hair follicle infections
  - Agent causing burn wound infections
  - Agents causing surgical site infections

- Random skin lesions
- Nosocomial infections
  - Catheter associated UTI
  - Catheter related blood stream infection
  - Ventilator associated Pneumonia
  - Surgical Site Infections

- Hand hygiene
- Organisms transmitted by droplet and aerosols
- Biosafety levels
- Occupational exposure (HIV and HBV)
- BMW management
  - BMW guidelines 1998
  - BMW Treatment Modalities

- Bioterrorist agents

20 NORMAL MICROBIAL FLORA

Introduction

00:00:13

- Normal flora is the term used to describe the various bacteria and fungi that are permanent residents of certain body sites, especially the skin, oropharynx, colon, and vagina.
- Viruses and parasites, which are the other major groups of microorganisms, are usually not considered members of the normal flora.
- Other names: commensals, human microbiome, indigenous microbiota
- The members of the normal flora vary in both number and kind from one site to another

- The skin and mucous membranes always harbor a variety of microorganism that can be arranged into two groups
  1. **Resident microbiota**: It consists of relatively fixed type of microorganism regularly found in a given area at a given age; if disturbed, it promptly reestablishes itself for example E.coli in intestine.
  2. **Transient microbiota**: It consists of nonpathogenic or potentially pathogenic microorganisms that inhabit the skin or mucous membranes for hours, days, or weeks for example MRSA.

- Normal flora depends upon various factors
  - Temperature
  - Moisture
  - pH
  - Environment (community or hospital)
  - Immune status
  - Anatomical sites
  - O_2

Role of Normal Flora

00:04:06

- Beneficial effects
  - Prevent colonization of pathogenic bacteria by competing for attachment sites and nutrients
  - Synthesize vitamins (B,K) which are absorbed as nutrient
  - Inhibits the growth of pathogenic bacteria by secreting waste substances (fatty acids, peroxidase, lactic acid, bacteriocin etc.)
  - Immune stimulation
  - Prevent allergic diseases (Hygiene hypothesis)
  - Complement activation by endotoxin of normal flor
- Harmful effects
  - Agent of disease
  - Transfer to susceptible host
  - Bacterial synergism
  - Contribute in spread of drug resistance
  - Competition for host's nutrients

Medically Important Members of the Normal Flora

00:09:54

Location	Important Organisms	Less Important Organisms
• Skin	• S. Epidermidis	• S. aureus, • Corynebacterium • Various Streptococci • Anaerobes • Yeasts
• Nose	• S. aureus[3]	• S. epidermidis • Corynebacterium • Various streptococci
• Mouth	• V. streptococci	• Various streptococci • Eikenella corrodens
• Dental plaque	• Strept. mutans	• Prevotella intermedia • Porphyro-monasgingivalis
• Gingival crevices	• Various anaerobes (e.g., Bacteroides, Fusobacterium, Streptococci., Actinomyces)	
• Throat	• V. streptococci	• Various streptococci • Neisseria species • H. influenzae • S. epidermidis

• Colon	• Bacteroides fragilis, E. coli	• Bifidobacterium • Eubacterium • Fusobacterium • Lactobacillus • Various aerobes • Gram –ve Rods • Enterococcus faecalis • Various strept.
• Vagina	• Lactobacillus, E. coli[3], group B strept.[3]	• Various strept. • Various gram –ve rods • Corynebacterium • C. albicans
• Urethra		• S. epidermidis • Corynebacterium • Various strept. • Gram –ve rods (E.coli[3])

- These organisms are not part of the normal flora

Probiotics 00:19:00

- Probiotics are live, nonpathogenic bacteria (or yeasts) that may be effective in the treatment or prevention of certain human diseases
- They are useful in the condition where the normal intestinal flora is suppressed.
- Examples: Bacillus coagulans, Bifidobacterium longum, Lactobacillus acidophilus, Saccharomyces boulardii.

Prebiotics 00:20:20

- In contrast to probiotics, prebiotics are the dietary non-digestible fibres which when administered, stimulated the growth and activity of normal flora.

PRACTICE QUESTIONS

Q1. The colon is the site of the largest number of normal flora bacteria. Which one of the following bacteria is found in the greatest number in the colon? 00:20:56

A. Bacteroides fragilis
B. Clostridium perfringens
C. Enterococcus faecalis
D. Escherichia coli

Q2. Certain microorganisms are never considered to be members of the normal flora. They are always considered to be pathogens. Which one of the following organisms fits intothat category? 00:21:28

A. Streptococcus pneumoniae
B. Escherichia coli
C. Mycobacterium tuberculosis
D. Staphyloccus aureus

Q3. Your patient is a 30-year-old woman with a previous history of rheumatic fever who has had fever for the past 2 weeks. On examination, you find a new heart murmur. Your suspect endocarditis and do a blood culture, which grows a viridians group streptococcus later identified as S. sanguis. Using your knowledge of normal flora, what is the most likely sources of this organism? 00:22:00

A. Duodenum
B. Skin
C. Throat
D. Urethra

Q4. A 76-year-old woman with a prosthetic (artificial) hip comes to you complaining of fever and pain in that joint. You are concerned about an infection by S. epidermidis. Using your knowledge flora, what is the most likely source of this organism? 00:23:25

A. Dental plaque
B. Mouth
C. Skin
D. Stomach

Q5. Antimicrobial therapy can decrease the amount of susceptible bowel flora and follow proliferation of relatively resistant colonic bacteria. Which one of the following species can proliferate and produce a toxin that causes diarrhoea? 00:24:35

A. Enterococcus species
B. S. epidermidis
C. Pseudomonas aeruginosa
D. Clostridium difficile

21 BONE AND JOINT INFECTIONS

OSTEOMYELITIS

- Osteo → Bone
- Myelo → Bone Marrow
- Types
  - Acute osteomyelitis
  - Chronic osteomyelitis

Acute Osteomyelitis

00:03:46

- Mainly happens after
  I. Skin infection (boils) gets hematogenous access
  II. After Trauma → Direct contamination of bone
  - ...ute: Hematogenous route
- MCC of acute osteomyelitis – Staph Aureus

Chronic Osteomyelitis

00:05:44

- Predisposing condition: Diabetes mellitus
  - In diabetes mellitus there is vascular insufficiency due to which they develop osteomyelitis

Clinical manifestations of osteomyelitis

00:06:58

- Bone pain & tenderness
- Fever, night sweats, fatigue
- Vertebral osteomyelitis
  - Lumber (60%) >Cervical or Thoracic
  - Back pain
- Symptoms of Acute osteomyelitis
  - Abrupt in onset
  - Progress rapidly
- Symptoms of chronic osteomyelitis
  - Indolent
  - Necrosis of bone occurs causing formation of sequestrum (An avascular piece of the Infected Bone)

Pathogens associated with osteomyelitis

00:10:28

Predisposing Factor	Common Organisms
• Neonates	• Streptococcus agalactiae (group B Streptococcus)
• Children and adults	• S. aureus
• Adults with vertebral osteomyelitis	• S. aureus, MTB
• Intravenous drug users	• S. aureus, Pseudomonas aeruginosa, Serratia marcesens, Candida albicans
• Puncture wounds of foot	• P. aeruginosa
• Cat bite	• Pasturella multocida
• Sickle cell anemia	• Salmonella species
• Exposure in endemic area	• Coccidioides immitis, Histoplasma capsulatum

Treatment

00:16:59

- Empirical Rx against S. aureus
  - Vancomycin (most of organisms are MRSA)
  - Nafcillin or other cephalosporins
- In cases
  - Vancomycin Intermediate Sensitive S. aureus (VISA): Daptomycin/ Linezolid
  - Vancomycin Resistant S. aureus (VRSA): Daptomycin/ Linezolid
- Surgical debridement must be done in cases of Chronic Osteomyelitis

INFECTIOUS / SEPTIC ARTHRITIS

00:19:42

- Main organism involved: S. Aureus
- Monoarticular involvement of large weight bearing joint like hip / knee joint

Diagnosis

- Analysis of Synovial Fluid

Refer Table 21.1

Clinical Manifestations

00:23:36

- Typical feature: Acute onset of inflamed joint (Hip or knee joint)
- Fever
- On physical examination
  - Joint is Red, warm, swollen with joint effusion
  - In children there is reluctance in moving the joint → Joint immobility d/t pain

Pathogens 00:25:28

- MCC of infectious arthritis: S. aureus
- In young sexually active individual with multiple partners person develops joint pain: Gonococci
- In patient with hip & knee Prosthesis: Staph. Epidermidis
- IV drug abusers: S. aureus & Pseudomonas Aeruginosa
- Borrelia Burgdorferi: Lyme's Disease (Transmitted via Tick)
- Arthritis in Lyme's disease is immune mediated therefore isolation of Bacteria is not possible

Lab Diagnosis 00:30:38

- Gram Stain of joint fluid
- Blood cultures are positive only in <30% cases
  - 10ml blood with 100ml Glucose Booth
- Radiological features-MRI
  - Soft tissue swellings
  - Destruction of joints occur & Reduced mobility if untreated

Treatment

- Empirical treatment for Staph. Aureus by
  i. Vancomycin
  ii. Nafacillin
- DOC is Ceftriaxone for Gonococci infection
- Remove joint fluid through Arthrocentesis & provide culture specific Antibiotic

VIRAL / IMMUNE COMPLEX MEDIATED ARTHRITIS 00:34:17

- Immunecomplex gets deposited on joints causing Arthritis / Arthralgia inducing inflammatory response

Clinical features

- Arthralgia: Painful joint without visible inflammation
- Short duration which goes off with time
- Small joints of hands are involved mainly
- Rubellain its course of infection or after immunization can cause viral arthritis
- Parvo virus B_{19} a/w Arthralgia mimics RA
- Arthralgia & Arthritis also a/w prodromal period of Hep B
- Hep C infection also a/w Arthritis like features which resembles RA
- Dengue & Chikungunya a/w severe Arthralgia

Treatment

- Symptomatic treatment

REACTIVE ARTHRITIS 00:43:57

- After GI Infection / Genito Urinary Infection: Develops joint pain
- It is d/t immune response to bacterial infection
- More seen in HLAB27 positive individuals as they are more prone to reactive arthritis
- Organisms a/w Reactive arthritis
  - Campylobacter
  - Chlamydia
  - Yersinia
  - Shigella
  - Salmonella

Clinical features

- Assymetric Arthritis of knee & ankle and accompanied by fever
- Resolves within days / weeks
- Recurrence are common
- Culture of synovial fluid always negative
- Reiter Syndrome
  - Conjunctivitis
  - Urethritis
  - Reactive Arthritis

Treatment

- NSAIDS (mainstay)
- Antibiotics have no role

RHEUMATIC FEVER 00:49:48

- Immune mediated post streptococcal disease (streptococcus pyogenes) → Group A
- Affects: Joints, Heart, Brain, Skin
- Age: 5-15yr
- It begins with migratory Polyarthritis involving the large joints after pharyngitis
- Carditis often occurs in this cases & mainly Pancarditis (Endocarditis, Myocarditis, Pericarditis) which results in CHF
- Mitral valve → MC involved
- Chorea → Involuntary movements
- Skin → Erythema marginatum & subcutaneous nodule

Jones criteria 00:55:26

Refer Table 21.2

- Low risk population
  1. Incidence is <1/1000
  2. School children <2/1 Lakh
- In addition lab evidence of prior infection by S.pyogenes is needed
- It consists of either
  - Positive throat culture or positive rapid streptococcal Ag test
  - Rising Anti-streptolysin O antibody Titer
    → ASO >200 todd units

Treatment

- DOC: Aspirin to reduce inflammation
- Antibiotics such as Penicillin G have no effect on course of the disease but can be given to reduce carriage of streptococci in the pharynx
- Prevention of RF involves prompt diagnosis & treatment of Streptococcus throat with penicillin G or oral penicillin V
- In patient with residual heart disease, prevention of additional damage to heart valves by preventing subsequent episodes of streptococcal [illegible] is very important
- This is achieved by monthly administration of Benzathine penicillin G, a depot preparation
- This should continue until the patient is atleast 20yr old or for 10yr after last attack

Table 21.1

Disease	Appearance	Cell Number (per µL)	Glucose (Fluid / blood ratio)
• Normal	• Clear	• <200 neutrophils	• Approx 1.0
• Infectious (septic)	• Cloudy	• >20,000 neutrophils	• <0.25
• Rheumatoid arthritis	• Opalescent	• 2000-20,000 neutrophils	• 0.5-0.8
• Trauma	• Clear	• 200-2000 neutrophils	• 1.0

Table 21.2

A. For all Patient Populations with Evidence of preceding Group A Streptococcal Infection

- Diagnosis: initial ARF
  - 2 major or 1 major manifestations + 2 minor manifestations
- Diagnosis: recurrent ARF
  - 2 major or 1 major &2 minor or 3 minor

B. Major Criteria

- Low risk populations
  - **C – C**arditis
    - → Clinical and / or subclinical
  - **A – A**rthritis
    - → Polyarthritis only
  - **R - R**heumatic chorea
  - **E – E**rythema marginatum
  - **S – S**ubcutaneous nodules
- Moderate and high risk populations
  - Carditis
    - → Clinical and /or subclinical
  - Arthritis
    - → Monoarthritis or Polyarthritis
    - → Polyarthralgia
  - Chorea
  - Erythema marginatum
  - Subcutaneous nodules

C. Minor criteria

- Low risk populations
  - Polyarthralgia
  - Fever (≥38.5°c)
  - ESR ≥60mm in the first hour and / or CRP ≥3.0mg/dL
  - Prolonged PR interval, after accounting for age variability (unless Carditis is a major criterion)
- Moderate and high risk populations
  - Monoarthralgia
  - Fever (≥38°c)
  - ESR ≥30mm/hr and / or CRP ≥3.0mg/dL
  - Prolonged PR interval, after accounting for age variability (unless carditis is a major criterion)

22 GIT INFECTIONS

Introduction 00:00:16

- Infections with a variety of agents can occur in any part of the gastrointestinal (GI) tract from the mouth to the anal canal
- Infections can range in severity from self limited to life-threatening, particularly if infection spreads from the gut to other parts of the body
- Infections are typically caused by the ingestion of exogenous pathogens in sufficient quantities to evade host defenses
- And the cause disease by multiplication, toxin production, or invasion through the gastrointestinal mucosa to reach the bloodstream and other tissues.
- In other cases, members of the normal flora of the GI tract can cause disease

ESOPHAGITIS 00:02:17

- Esophagitis is an inflammatory process that can damage the esophagus
- Inflammation caused by infection, typically by fungi such as Candida or viruses such as herpes simplex virus, CMV causes the symptoms of esophagitis.
- Most cases occur in immunocompromised patients, especially those with reduced cell-mediated immunity. The extent of damage to the esophagus is typically related to the severity of symptoms
- Clinical Manifestations
  - Odynophagia and dysphagia are the key clinical manifestations of esophagitis.
- Pathogens
  - Candida is the most common etiology, particularly among human immunodeficiency virus (HIV)-infected patients and other immunocompromised hosts.
  - Less common pathogens include herpes viruses such as cytomegalovirus and herpes simplex virus
  - Noninfectious causes also occur, such as acid reflux from the stomach and medication-induced disease (e.g., Doxycyclin)
- Diagnosis
  - May be empiric after a trial of Fluconazole results in improvement for presumed Candida esophagitis.
  - If an empiric course of Fluconazole does not work, then endoscopy for visualization and biopsy could be helpful, particularly in immunocompromised hosts.
  - Biopsy samples should be analyzed by using pathologic and microbiologic tests

GASTRITIS 00:06:39

- Gastritis refers to inflammation of the mucosa of the stomach
- It may be erosive or non-erosive, depending on histologic and endoscopic findings
- A break in the gastric and adjacent duodenal mucosa defines peptic ulcer disease

Pathophysiology

- The mechanism by which one of the main pathogens, Helicobacter pylori causes peptic ulcer disease has been largely elucidated. Following attachment to the gastric mucosa
- H. pylori causes direct mucosal damage by the combination of ammonia production (from the action of the organism's urease on urea) and the host inflammatory response

Clinical Manifestations of Gastritis 00:08:39

- Patients with gastritis typically complain of dyspepsia (epigastric pain, burning), nausea, and vomiting. In the case of peptic ulcer disease, epigastric pain is the primary symptom
- Some patients may report alleviation of pain with food, particularly with Duodenal ulcer
- Gastrointestinal bleeding is a complication of peptic ulcer disease. Some patients with gastritis may be asymptomatic.

Pathogens

- Infectious and noninfectious etiologies are possible. Among infectious causes, H. pylori is the most important
- Viruses such as cytomegalovirus and fungi such as Mucor may rarely cause ulcer disease as well. Particularly among immunocompromised patients
- Following ingestion of raw fish, larvae of Anisakis species may become embedded in the gastric mucosa and cause severe abdominal pain
- Mycobacteria (tuberculosis and nontuberculosis mycobacterial), Giardia, and Strongyloides may also cause gastritis
- Noninfectious causes such as alcohol and medications (e.g., non steroidalanti-inflammatory drugs) are also implicated

Diagnosis

- Upper endoscopy with gastric biopsy [illegible] diagnostic strategy. If abnormal findings are detected, pathologic analysis and further directed testing may be performed
- For the H. pylori-associated ulcers
  - Confirmed using a Rapid urease test on the biopsy specimen
  - Stained by Warthin starry stain

Treatment

- For H. pylori
  - Combination therapy with two antibiotics, such as Ampicillin and Clarithromycin, plus a proton pump inhibitor. such as omeprazole, or Bismuth is used with varying success

DIARRHEA

00:13:43

- Acute diarrhea classified as non-inflammatory (watery. Non-bloody) or inflammatory (bloody).
- Bloody diarrhea is also known as dysentery. For example, bloody diarrheacaused by &Shigella is often called bacillary dysentery.
- Diarrhea must be calibrated against the patient's normal bowel movements but is usually considered to be greater than three to five bowel movements per day.
- Most of the infectious agents that cause diarrhea act at the small intestine (where the majority of fluid normally gets absorbed) or the colon

Infectious Agents of Acute Diarrhea and the underlying mechanism

Refer Table 22.1

Infectious Agents of Food Poisoning 00:23:53

Refer Table 22.2

Agents Causing Traveller's Diarrhea

- **Vi** - **Vi**brio cholera
- **P** - **P**lesiomonas
- **E** - **E**ntamoeba
- **C** - **C**ryptosporidium
- **G** - **G**iardia
- **S** - **S**almonella & **S**higella
- **C** - **C**ampylobacter
- **A** - **A**eromonas
- **N** - **N**orovirus →a/w cruise ships
- **E** – **E**TEC (most imp. cause), EAEC
- **R** - **R**otavirus

How to remember

- ViP ECG S²CANER

Identification Features/ Detection Methods of Common Organisms Causing Acute Diarrhea or Dysentery

Refer Table 22.3

APPENDICITIS

00:48:12

- Appendicitis is inflammation of the vestigial vermiform appendix. It is one of the most common causes of acute abdomen requiring surgical exploration.

Pathophysiology

- Obstruction of the appendix by one of a variety of causes (e.g., fecoliths, infection such as parasites, tumor) leads to an increase in luminal and intramural pressure.

Clinical Manifestations

- Clinical manifestations include abdominal (especially periumbilical migrating to right lower quadrant) pain, anorexia, nausea, and vomiting.
- Initial symptoms may be missed because they may be nonspecific (e.g., indigestion). A standard abdominal computed tomography (CT) scan with contrast is often used when appendicitis is suspected.
- Early in the course of the disease the predominant organisms are anaerobic. In late disease, mixed organisms predominate. E. coli, Peptostreptococcus, Bacteroides fragilis, and Pseudomonas are commonly isolated.
- Yersinia. Campylobacter, and Salmonella can cause an acute ileitis and mesenteric adenitis that can mimic appendicitis
  - Yersinia pseudoTB / Yersinia Enterocolitica can cause Pseudo appendicitis

Diagnosis

- Clinical manifestations combined with imaging are typically used to make a decision as to whether a patient should be taken to the operating room.

Treatment

- Surgery is the definitive treatment for appendicitis, usually in concert with perioperative antibiotics.

DIVERTICULITIS

00:52:29

- Diverticulitis is inflammation of a sac-like protrusion of the colonic wall, usually in the sigmoid colon.

- Perforation of the diverticulum with consequent abscess formation or peritonitis may occur

Pathophysiology
- Colonic diverticula may occur following years of a diet deficient in fiber.

Clinical Manifestations
- There is a range of symptoms depending on the degree of perforation.
- Patients usually present with dull, aching, left lower quadrant abdominal Pain. This is often accompanied by a low-grade fever, leukocytosis nausea, and vomiting. Diarrhea or constipation may be present.
- If perforation occurs, patients can present with generalized peritoneal signs with diffuse abdominal pain and shock.

Pathogens
- Bowel flora such as anaerobes, i.e B. fragilis (may lead to shock and DIC) and those in the Enterobacteriaceae family, such as E. coli, are typically involved.

Diagnosis
- Abdominal CT will show colonic diverticula and associated wall thickening, fat infiltration, abscesses, and extra luminal air or contrast medium

ENTERIC FEVER

00:55:59

- Enteric fever is a clinical syndrome comprised of constitutional symptoms, such as fever, headache, nausea, vomiting and abdominal pain caused by the Salmonella typhi.
- Following the consumption of contaminated food. Salmonella bacteria enter through the intestinal mucosal epithelium by transcytosis.
- The microbes then replicate in the macrophages of Peyer patches, mesenteric lymph nodes, and the spleen.
- Bacteremia then occurs with dissemination to lungs, gallbladder, kidneys, or central nervous system.
- Humans are the only reservoir for S. typhi, so contamination of food or water by human fecesshould be suspected

Clinical Manifestations
- A prodromal phase is characterized by constitutional symptoms such as malaise, together with abdominal pain, constipation, and headache.
- Fever increases over the next several days. Diarrhea is uncommon
- During the second week of disease, a typical transient rash of pink Maculopapular lesions (rose spots) may be seen.
- Splenomegaly occurs more commonly than hepatomegaly, but both may occur. Relative bradycardia and leukopenia are often observed
- The chronic carrier state occurs in approximately 3% of patients with typhoid fever.
- The organisms typically reside in the gallbladder and are excreted in the stool, serving as a source of infection for others.

Pathogens
- S. typhi and other Salmonella species, such as Salmonella paratyphi A and S. paratyphi B. cause Enteric fever

Diagnosis
- A history of travel to endemic areas, together with a compatible clinical presentation, is often used initially.
- Any fever in a returning traveler should promptblood culture and a clinical suspicion for enteric fever.
  - **B** – **B**lood culture/ Bone marrow culture (1st week)
  - **A** – **A**gglutination (2nd week)
  - **S** – **S**tool culture (3rd week)
  - **U** – **U**rine culture (4th week)

How to remember

- BASU

- Enrichment media: Selenite-f-broth

Treatment

01:02:24

- Intravenous Ceftriaxone is effective treatment modality.
- Cholecystectomy should be considered for those chronic carriers who do not respond to antimicrobial therapy.
- Prevention Hygienic measures to protect the food and water supply from Human fecal contamination are an important public health intervention.
- Immunization may not always be effective but can be considered in epidemic outbreaks, for travelers to endemic countries and for household contacts of typhoid carriers.

Vaccine
- Two vaccines against typhoid fever are available in the United States, both providing approximately 50% to 80% protection.
- The vaccine containing the Vi capsular polysacch[illegible] typhi has the advantage of being administered once IM
- The other vaccine contains live attenuated S. typhi organisms and is administered orally. It has the advantage of stimulating gut immunity thereby interrupting transmission

Table 22.1

Non-inflammatory	**Location**
	• Proximal small bowel
	
	Illness
	• Watery diarrhea
	
	Stool findings
	• No fecal leukocytes
	• Fecal Lactoferrin is not increased
		Bacteria:(Mostly Enterotoxin mediated)
		• Vibrio Cholerae
		• Escherichia coli
		  ○ Enteropathogenic
		  ○ Enterotoxigenic
		  ○ Enteroaggregative
		• Clostridium Perfringens
		• Bacillus cereus
		• Staphylococcus aureus
		• Aeromonas hydrophila
		• Plesiomonasshigelloides
			Viruses
			• Rotavirus
			• Norovirus
			• Enteric adenoviruses
			
			Parasites
			• Giardia lamblia
			• Cryptosporidium species
			• Cyclospora species
			• Cystoisopora species
			• Microsporidia

Inflammatory (invasion or cytotoxin)	**Location** • Colon or distal small bowel **Illness** • Dysentery • Inflammatory diarrhea **Stool findings** • Fecal pus cells (Polymorphonuclear leukocytes)- increased • Fecal Lactoferrin-increased	**Predominantly dysentery** • Shigella species • Campylobacter Jejuni • Campylobacter E. coli • Enteroinvasive E.coli • Vibrio parahemolyticus **Predominantly inflammatory diarrhea** • Salmonella species • Yersinia enterocolitica • Listeria monocytogenes • Clostridium Difficile • Aeromonas hydrophila • Plesiomonas shigelloides	**Parasite:**(Predominantly dysentery) • Entamoeba histolytica • Balantidium coli
Penetrating	**Location** • Distal small bowel **Illness** • Enteric fever **Stool findings** • Fecal mononuclear leukocytes (↑)	• Salmonella typhi (enteric fever) • Yersinia enterocolitica (typhoid like illness)	

Table 22.2

Incubation period, organism	Symptoms	Common food sources
1-6 hr		
• S. aureus	• Nausea, vomiting, diarrhea	• Ham, poultry, potato or egg salad, mayonnaise, pastries
• Bacillus cereus	• Nausea, vomiting, diarrhea	• Fried rice
• Clostridium botulinum	• Nausea, vomiting, constipation	• Canned food
8-16 hr		
• Clostridium perfringens	• Abdominal cramps, diarrhea (vomiting rare)	• Beef, poultry, legumes, gravies
• B. cereus	• Abdominal cramps, diarrhea (vomiting rare)	• Meats, vegetables, dried beans, cereals
> 16 hr		
• Vibrio cholerae	• Watery diarrhea	• Shellfish, water
• Enterotoxigenic E.coli	• Watery diarrhea	• Salads, cheese, meat, water
• Enterohemorrhagic E.coli	• Bloody diarrhea	• Ground beef, salami, raw milk, raw vegetables, apple juice
• Salmonella species	• Inflammatory diarrhea	• Beef, poultry, eggs, dairy products
• Campylobacter Jejuni	• Inflammatory diarrhea	• Poultry, raw milk
• Shigella species	• Dysentery	• Potato or egg salad, lettuce, raw vegetables
• Vibrio parahaemolyticus	• Dysentery	• Mollusks, crustaceans

Table 23.3

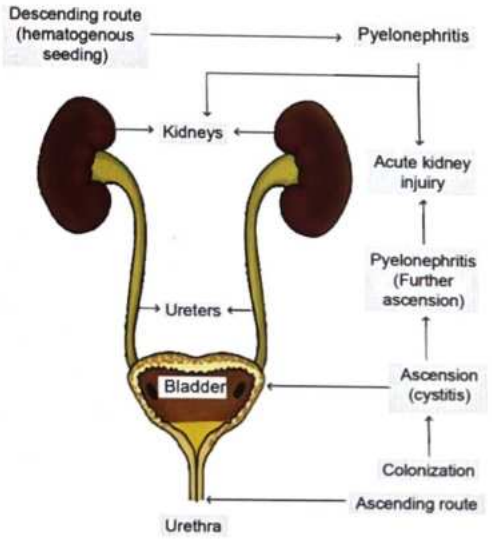
	Presentation	Identification features
Vibrio cholerae	Watery diarrhea	• Darting motility • Coma-shaped Gram –negative bacilli in culture smear • Catalase and oxidase positive • TCBS agar: sucrose fermenting yellow colored colonies • Agglutinates with Vibrio cholerae O1 antisera and Ogawa antisera
Shigella	Dysentery	• Gram –ve bacilli, non-motile • Catalase +ve, oxidase –ve • MAC or DCA: NLF translucent • XLD: red colonies without black center • Agglutinates with Shigella polyvalent antisera and specific monovalent antisera
Group B Salmonella	Inflammatory diarrhea	• Gram –ve bacilli, motile • Catalase positive, oxidase –ve • MAC: non-lactose fermenting translucent colonies • DCA: non-lactose fermenting colonies with black center • XLD: red colonies with black center • Agglutinates with Salmonella poly-O antisera and serotype (O_4) specific
Viral agents	Diarrhea	**Agents**: Rotavirus, Norovirus, Adenovirus 40, 41 etc • Detection of viral particles in stool specimen by electron microscopy • Detection of viral antigen by ELISA or • Detection of nucleic acid (RNA of DNA) by PCR in stool specimen
Intestinal parasites		**Stool microscopy detects**
Entamoeba histolytica	Dysentery	• Trophozoites and / or quadrinucleated round cyst • Detection of specific antigen (e.g. lectin)/ specific genes in stool
Giardia intestinalis	Fatty diarrhea	• Trophozoites (tear drop-shaped binucleate) with four pairs of flagella and / or • Tetra-nucleated oval cyst with a central axoneme
Trichuris	Dysentery	• Barrel-shaped eggs with mucus plugs at both ends, bile stained
Enterobius vermicular	Nocturnal and pruritus	• Plano-concave egg containing larva, non-bile stained
Ascaris lumbricoides	Malabsorption	• Fertilized egg: round oval, thick albumin coat, floats in saturated saline, bile stained • Unfertilized egg: Elongated, thin albumin coat, does not float in saturated saline, bile stained
Hookworm	Diarrhea, anaemia	• Egg: Oval, contains segmented four blastomeres, clear space between blastomere and egg, shell, non-bile stained
Strongyloides	Diarrhea	• Detection of rhabditiform larva in stool microscopy

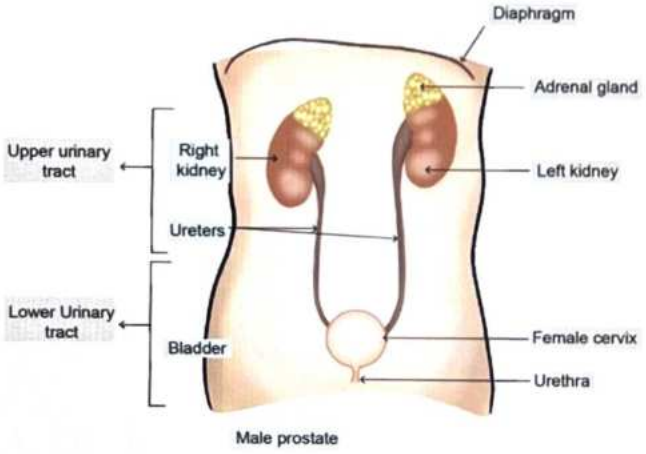
23 URINARY TRACT INFECTION

- Disease of Urinary Tract in which there is microbial invasion of Urinary Tract

Difference between upper UTI and Lower UTI 00:01:31

	Lower UTI	Upper UTI
• Site	• Urethra & Bladder	• Kidney & Ureter
• Symptoms	• Dysuria, Urgency, Frequency	• Fever, Vomiting, Abdominal/ flank pain
• Route of spread	• Ascending	• Can be Ascending or Descending





Male prostate

Predisposing Factors of UTI 00:06:55

- Gender: F >M
  - Short urethra
  - Close proximity of urethral meatus to anus
- Age: Incidence increase with age
- Pregnancy
  - Anatomical &Hormonal changes predispose to UTI
  - Asymptomatic bacteriuria needs to be treated
- Structural &Functional Abnormality
  - Obstruction d/t
    - → Renal & ureteric stone
    - → Prostate can cause obstruction in males in chronic stage
    - → Any tumors
  - Neurogenic Bladder d/t
    - → Spinal cord injury
    - → Multiple sclerosis
- Virulence of Bacteria
  - Express Pili or Fimbriae which helps in adhesion to uro-epithelium
  - P. Fimbriae are mannose resistant & leads to more destruction
- Vesicoureteric Reflex (VUR)
  - Valve mechanism is defective
  - Allow urine from bladder into ureter allowing Retrograde movement
- Genetic factors

Etiology 00:17:50

- Uropathogenic E. coli: MCC of UTI
  - MC Nosocomial infection: UTI
- Enterobacteriaceae family
- Candida albicans
- Certain staph like Staph Saprophyticus, cause UTI in young female associated with Honeymoon Cystitis
- Enterococcus
- Parasites
  - T. vaginalis
  - Schistosoma Haematobium
- Viruses
  - HSV
  - Adeno virus
  - CMV

Lowe UTI

- Invasion of renal parenchyma through hematogenous spread occurs d/t Bacteria
- Cause by
  - S. aureus
  - Salmonella
  - MTB
  - Leptospira
  - Candida

- Cystitis 00:23:56
  - Inflammation of Bladder
  - Clinical Features
    - → Dysuria, Frequency, urgency
    - → Urine become cloudy
    - → No Hematuria
    - → No associated systemic manifestation

- Acute Urethral Syndrome 00:25:36
  - Lower UTI
  - Mainly in young sexually active female
  - Bacterial count = $10^2 - 10^5$ FU/ml
  - Pyuria is present
  - Etiology: Chlamydia, Gonococcus, HSV

Upper UTI 00:27:27

- Inflammation of
  - Kidney parenchyma
  - Calyces
  - Renal pelvis
- Systemic Manifestation: Fever, Flank pain, Vomiting (Absent in Lower UTI)

Lab Diagnosis 00:29:11

- Urine specimen Collection
  1. MSU Clean catch
  2. Suprapubic aspirate
  3. Catheterized patient: Taken from catheter tube after clamping

- Transportation: In Urine
  - Add boric acid
  - Refrigeration if time is there for urine processing

- In lab
  - Direct examination
    - → Wet mount examination: Pus cells present
  - Leukocyte Esterase Test: Detects esterase enzyme activity
  - Nitrate Reduction test aka Griess Test: Converts Nitrate to nitrite
  - Catalase test
  - Gram staining
  - Culture on
    - → MacConkey agar or CLED
      - CLED is better media because
      - It is a non selective media
      - Inhibits the growth of proteus
    - → Both medias are Differential and indicator
    - → Get colony count and Multiply with dilution factor (10^3): If final count is
      - $>10^5$: Significant Bacteriuria. Even if patient have no symptom it should be treated for sure
      - $>10^4$ + signs & symptoms of UTI: Case of UTI
      - 1 bacteria/ml in Suprapubic Aspirate: UTI

Treatment 00:38:54

- Norfloxacin
- Nitrofurantoin
- Antibiotic sensitive testing should be done on MHA by Kirby Baur Disc Diffusion Test→ then prescribe antibiotics
- Cephalosporin
- Aminoglycoside
- In case of Nosocomial infections (UTI) treat according to the causative organism:
  - Piperacillin + Tazobactam
  - Fosfomycin
  - Carbapenems: if more resistant strains are present

Q1. A 30 yr old female with frequency, urgency, Dysuria since 2 days 00:41:32

Ans: Lower UTI

Q2. A 20 yr old female with High grade fever along with vomiting, flank pain & increase in frequency of urination x 3 days

Ans: Upper UTI

24 PELVIC INFECTIONS

Introduction 00:00:13

- Infections in the pelvic organs and surrounding structures comprises a heterogenous group of diseases
- Primarily affect sexually active Women & Men
- Most of the pathogens implicated are sexually transmitted, so an important facet of treatment is Partner notification and treatment, as well as patient education regarding safe sexual practices

Genital Ulcer 00:01:21

- Manifests as a breach in the skin or mucosa of the genitalia
- Usually caused by a sexually transmitted infection.
- Causative organisms
  - HSV-2 (MC) followed by Syphilis and Chancroid
  - Non infectious cause: Behcet's disease

Pathophysiology

- There are different mechanism of injury depending on the pathogen
- In Chancroid: A cytotoxin secreted by Haemophilus ducreyi may be important in epithelial cell injury

Clinical manifestation

- Although lesions have a characteristic appearance, it is important to note the local epidemiology because lesions may appear in an atypical fashion
- Common infections etiologies of genital ulcer disease include
  - HSV-2 (Genital Herpes)
  - Treponema pallidum (Primary Syphilis)
  - H.ducreyi (Chancroid)
  - Chlamydia trachomatis serovars L_1-L_3 (Causing Lympho granuloma venereum)
  - Klebsiella granulomatis (Causing Granuloma inguinale / Donovanosis)

Refer Table 24.1 00:04:53

Chancre Syphilis

Genital Herpes

LGV

Chancroid

Donovanosis

Diagnosis

- Thorough sexual and medical history f/b physical examination
- Clinical characteristics are very helpful but there can be overlap in presentation, and there may also be multiple syndromes co-presenting
- Diagnostic testing is highly recommended, testing for other sexually transmitted diseases including HIV is also important because there is often co-transmission of multiple pathogens

Treatment

- DOC for Genital Herpes: Acyclovir or one of its derivatives, Famciclovir or Valacyclovir

- Primary and secondary syphilis: Long acting penicillin (Benzathine penicillin G)
- DOC for Chancroid: Azithromycin
- DOC for Lymphogranuloma venereum: Doxycycline

Prevention strategies

- Consistent use of condoms is an important measure that can prevent genital ulcers
- Partner notification & treatment are important prevention strategies as well

Vaginitis 00:29:30

- Inflammation of Vagina that can result in discharge, itching and pain

- Common causes of vaginitis are
  - Candidiasis
  - Trichomoniasis
  - Bacterial vaginosis

- Non infectious causes
  - Lichen Planus
  - Certain medications E.g. Oral contraceptives

Pathophysiology

- Use of Antibiotics that inhibit the normal flora of the vagina (especially lactobacilli), predisposes to Candida vaginitis. Candida is a member of Normal flora of many women
- Bacterial vaginosis: Not a sexually transmitted disease.
- Trichomoniasis, on the other hand, is a Sexually transmitted disease

- The patients are usually prompted to seek medical attention because of an abnormal vaginal discharge. This may be accompanied by Pruritus, pain (including dyspareunia), and symptoms of vaginal irritation

- Appearances
  - White "cottage cheese" appearance: Vaginal Candidiasis
  - Strawberry cervix: Trichomoniasis.

- Strawberry cervix: there are red, punctuate lesions on the cervix, and frothy (greenish, Greyish white) exudates can be seen at the cervical os

- Bacterial vaginosis: Discharge is thin, grayish with unpleasant odor, often described as Fishy (Whiff test +ve)

Refer Table 24.2

Candida Vulvovaginitis

Trichomoniasis

Clue cells

Cervicitis 00:50:43

- Cervicitis is Inflammation of the Uterine cervix.
- Acute cervicitis is usually due to a sexually transmitted infection caused by either C. trachomatis or Neisseria gonorrhoeae (MC) or both

Cervicitis

Clinical Manifestations

- Asymptomatic in most of cases.
- Detected on speculum examination and following routine screening to C.trachomatis and N. gonorrhoeae
- Women with concomitant urethral infection may have dysuria
- Physical examination: Friability of the cervical tissue
- Pathogens: C. trachomatis (Serovars D-K), N. gonorrhoeae, HSV and T. vaginalis
- Diagnosis
  - Clinical diagnosis →Increased friability of the cervix, with or without mucopurulent discharge.
  - Laboratory diagnosis
    - → NAAT for C. trachomatis and N. gonorrhoeae
    - → Gram stain, Culture (If NAAT testing not available)
- Treatment (for both C. trachomatis and N. gonorrhoeae)
  - Ceftriaxone intramuscularly + Azithomycin orally
  - Treat the partner too
- Prevention
  - Consistent use of Condoms
  - Partner notification and treatment

Pelvic Inflammatory Disease 00:54:36

- Polymicrobial infection of upper genital tract structures (Uterus, Fallopian tubes and Ovaries)
- Endocervical canal barrier compromised→ Vaginal bacteria ascend into normally sterile space of the upper genital tract

- STD's affecting the Cervix can initiate the process, permitting the anaerobic bacteria of the vagina to ascend
- Having multiple sex partner increase the risk of PID
- Multiple episodes of PID lead to scarring of the fallopian tubes and an increased risk of ectopic pregnancy and sterility

Clinical Manifestation

- Symptoms
  - Lower back pain
  - Fever, chills, lower abdominal pain
  - Cervical and adnexal tenderness
  - On physical examination, pain and tenderness on motion of the cervix are important diagnostic signs

- Pathogens: PID is primarily associated with N. gonorrhoeae and C. trachomatis, together with enteric gram-negative rods and anaerobes

- Diagnosis
  - Uterine, adnexal, or cervical motion tenderness
  - Fever, presence of leukocytes on cervical / vaginal discharge, elevated C-reactive protein
  - Laboratory evidence of cervical infection with N. gonorrhoeae or C. trachomatis

Treatment

- Mild disease
  - Cefoxitin / Ceftriaxone (one dose) + doxycycline (14 days) ± Metronidazole (OPD based treatment)

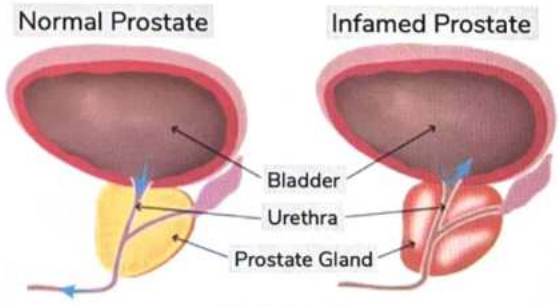
- Moderate to severe disease
  - IV Cefoxitin / Cefotetan + Doxycycline or Clindamycin + gentamicin (IPD based treatment)
  - This is f/b Oral antibiotics only after 24 hours of improvement of the patient

Urethritis 01:02:02

- Definition→Inflammation of the Urethra, usually caused by STD, particularly in sexually active men.
- Noninfectious causes: Reiter's syndrome
  - Reiter's syndrome includeUrethritis, Conjunctivitis, and Reactive arthritis
- Clinical manifestations:
  - Dysuria: common presenting complaint
  - Discharge from Urethra
  - Pruritis and burning sensation
- Pathogens
  - N. gonorrhoeae and C. trachomatis (MC),
  - Other organisms: Mycoplasma genitalium and T. vaginalis.
- Diagnosis : NAAT for C trachomatis and N. gonorrhoeae

Prostatitis 01:03:48

- Acute bacterial prostatitis
  - Characterized by typical Irritative voiding symptoms (urinary frequency, hesitancy, feeling of incomplete voiding, dribbling), fever, Pyuria, and positive urine cultures
- Chronic bacterial prostatitis
  - Characterized by the same voiding symptoms, but fever and pyuria are typically absent
- Pathophysiology
  - Bacteria ascend the urethra, and then reflux into the prostatic ducts where infection occurs
- Clinical manifestations
  - Acute Prostatitis
    - → Patients appear ill with fever, chills, irritative voiding symptoms, and pelvic or perineal pain.
    - → Physical examination reveals a very tender and enlarged prostate
  - Chronic Prostatitis
    - → Symptoms are more subtle.
    - → Patients may present with Recurrent UTI
    - → Prolonged treatment of prostatitis cures the disease
- Pathogens
  - Enterobacteriaceae (eg. Escherichia coli, Klebsiella and Proteus species), Pseudomonas
  - In sexually active men, N. gonorrhoeae and C trachomatis cause prostatitis, especially in a/w urethritis and epididymitis
- Diagnosis
  - Edematous and tender prostate
  - Urine culture: Done to determine causative organisms
  - Prostatic fluid culture: Not done in Acute Prostatitis because prostatic massage should not be done during the acute phase
  - Prostatic massage may be useful in Chronic Prostatitis
- Treatment
  - Trimethoprim-Sulfamethoxazole or Ciprofloxacin can be used as empiric therapy until culture results return
  - These agents exhibit good penetration into the prostate. Therapy is prolonged (4 to 6 weeks)



Prostatitis

Table 24.1

	Syphilis	Herpes	Chancroid	LGV	Donovanosis
• I.P.	• 9-90 days	• 2-7 days	• 1-14 days	• 3 days -6 weeks	• 1-4weeks
• Primary lesions	• Papule	• Vesicle	• Pustule	• Papule, Pustule, Vesicles	• Papule
• No. of lesions	• 1	• Multiple	• Multiple	• 1	• Variable
• Diameter	• 5-15mm	• 1-2mm	• Variable	• 2-10mm	• Variable
• Edges	• Sharply demarcated	• Erythematous	• Undermined edges	• Elevated	• Elevated, Irregular
• Depth	• Superficial or deep	• Superficial	• Excavated	• Sup. / deep	• Elevated
• Base	• Smooth, non purulent	• Serous red	• Purulent, bleeds easily	• Variable, non vascular	• Red velvety
• Indurations	• Firm	• None	• Soft	• Firm	• Firm
• Pain	• Painless • (uncommon)	• Tender	• Very tender	• Variable	• Uncommon
• LAP	• Firm, non-tender B/L	• Firm tender B/L	• Tender	• Tender	• Pseudo-buboes (non-tender)

Table 24.2

Feature	Normal findings	Candidiasis	Trichomoniasis	Bacterial vaginosis
• Symptoms	• Nothing	• Vulval itching	• Produce discharge, itching	• Bad odour, increased discharge
• Discharge feature	• Variable / scanty / clear	• Scanty white clumps	• Profuse, white / yellow or green coloured discharge	• White / grey
• Vaginal epithelium	• None	• Redness of vaginal epithelium • Vulvar dermatitis	• Redness of vaginal epithelium • Colpitis macularis (Strawberry cervix)	• None
• PH of vaginal fluid	• <4.5	• <4.5	• >5	• >4.5 (Amsel's criteria)
• Microscopy	• N. epithelial cells, lactobacilli	• Leucocytes, pseudo-hyphae	• Leucocytes, Trichomonads in Urine Microscopy	• Clue cells (Nugent score)
• Investigation	• NAAT (Nucleic Acid Amplification Test)			• NAAT
• Treatment		• Clotrimazole 100mg vaginal tab OD x 7days or Fluconazole 150mg orally (single dose)	• Metronidazole 500mg BD x 7 days (orally)	• Metronidazole 500mg BD x 7 days (orally)

25 CARDIAC INFECTIONS

Introduction 00:00:26

- Cardiac infections are severe, life threatening infections in many cases. The heart valves endocardium, myocardium, and pericardium can all be infected
- In addition, infection of cardiac devices (pacemakers, defibrillators) is becoming more frequently diagnosed with their increase in use.

ENDOCARDITIS 00:01:17

- Endocarditis is an infection of the valves of the heart.

Pathophysiology

- Infection of the heart valves is thought to result from the colonization of damaged valvular endothelium by circulating pathogens.
- Endothelial damage may result from
  - Turbulent blood flow around the valve (Because of congenital or rheumatic heart disease)
  - Direct injury from foreign bodies (e.g., intravenous catheters)
  - Repeated intravenous injections of particles in intravenous drug users.
- Organisms enter the bloodstream most often at the site of dental surgery, indwelling intravenous catheters, or intravenous drug use.
- Adhesion of bacteria to the damaged endothelium is enhanced by their ability to produce a glycocalyx
- Once the infection has begun, a combination of organisms and thrombus organize to form a vegetation
- Destruction of the valve occurs at different rates depending on the virulence of the organism. As the valve is destroyed, symptoms of valvular regurgitation can develop.
- Organisms can spread to surrounding myocardium. resulting in abscess formation and destruction of the electrical conduction system
- As the vegetation on the valve enlarges, fragmen[t] can spread via the blood stream (emboli), resulting in catastrophic effects, such as CVA and metastatic infections
- Prolonged infection as seen in subacute endocarditis can result in antigen-antibody complex formation. Deposition of these complexes can result in other clinical manifestations.
- Artificial materials within the heart, such as prosthetic heart valves, pacemakers, and defibrillators, severe as potential sites for infection

Refer Table 25.1

HACEK Organisms

- Haemophilus aphrophilus and Haemophilus paraphrophilus, now called Aggregatibacter aphrophilus and Aggregatibacter paraphrophilus
- Actinobacillusactinomycetemcomitans, (now called as Aggregatibacter actinomycetemcomitans)
- Cardiobacterium hominis
- Eikenella corrodens
- Kingella kingae
- DOC: Ceftriaxone

- Pathogens that do not grow in routine blood culture media and require specialized testing for diagnosis include:
  - Bartonella species, Coxiella burnetii, Brucella species, and Tropheryma whipplei. These are cited as pathogens that cause "culture-negative" endocarditis.
  - The most frequent cause of "culture-negative endocarditis" is the use of antimicrobials prior to obtaining blood cultures

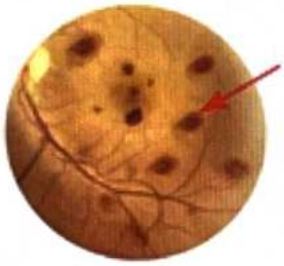
Clinical Manifestations

- Depending on the virulence of the infecting pathogen. the time course of illness may be days
- Acute endocarditis: caused byStaphylococcus aureus
- Subacute endocarditis: caused byviridans group streptococci
- Constitutional symptoms: fever (>80% cases). Chills, night sweats, anorexia
- Consequences of destruction of heart valves and associated structures leads to formation of new murmur, heart failure, atrioventricular (AV) block (PR prolongation seen on ECG)
- Embolic phenomenon: Left-sided endocarditis: CVAs or brain abscess (new focal neurologic deficits),splenic or renal infarcts (abdominal or flank pain), and emboli to others sites manifesting as splinter hemorrhages , Janeway lesions , retinal hemorrhages, and conjunctival hemorrhages
- Right-sided endocarditis: septic pulmonary emboli (cough, shortness of breath, chest pain, hemoptysis)
- Antigen—antibody deposition from uncontrolled infection leads to formation of

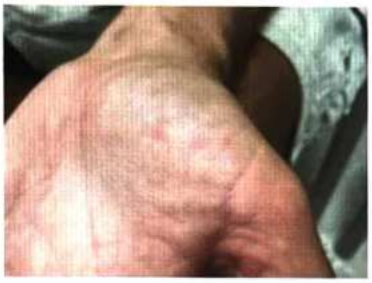
- R - Roth's spots
- O - Osler's nodes
- G - Glomerulonephritis (hematuria), and/or arthritis

How to remember

- ROG



Roth's spots (Litten's Sign)



Janeway Lesion

Osler's nodes (Painful, red, raised lesion)

Splinter Haemorrage

Clinical and Laboratory Features of Infective Endocarditis

Frequency	Frequency (%)
• Fever	80-90
• Chills and sweats	40-75
• Anorexia, weight loss, malaise	25-50
• Myalgias, arthralgias	15-30
• Back pain	7-15
• Heart murmur	80-85
• New/worsened regurgitant murmur	20-50
• Arterial emboli	20-50
• Splenomegaly	15-50
• Clubbing	10-20
• Neurologic manifestations	20-40
• Peripheral manifestations (Osler's nodes, subungual hemorrhages, Janeway lesions, Roth's spots)	2-15
• Petechiae	10-40
• Laboratory manifestations	
○ Anemia	70-90
○ Leukocytosis	20-30
○ Microscopic hematuria	30-50
○ Elevated ESR	60-90
○ Elevated CRP	>90
○ Rheumatoid factor	50
○ Circulating immune complexes	65-100
○ Decreased serum complement	5-40

Modified Duke criteria for the clinical diagnosis of infective endocarditis

Major criteria

1. Positive blood culture: Any one of the following:

a. Typical IE organism isolated from two separate sets of blood cultures (Viridans Streptococci, S. gallolyticus, HACEK group, S. aureus or Enterococci)

b. Persistently positive blood culture with agents other than typical IE organisms
- Blood culture sets drawn > 12 h apart; or
- All of 3 or majority of > 4 separate sets of blood culture, with first and last drawn at least 1 h apart

c. Single positive blood culture for Coxiella burnetii or phase 1 IgG antibody titre of >1:800

2. Evidence of endocardial involvement: Anyone

a. Positive echocardiogram
- Oscillating intracardiac mass on valve or
- Abscess or
- New partial dehiscence of prosthetic valve

b. New valvular regurgitation (increase or change in the preexisting murmur not sufficient)

Minor criteria

1. Predisposition: predisposing heart condition or IV drug use

2. Fever >38.0°C (>100.4° F)
3. Vascular phenomena
   a. Major arterial emboli
   b. Septic pulmonary infarcts
   c. Mycotic aneurysm
   d. Intracranial hemorrhage
   e. Conjunctival hemorrhages
   f. Janeway lesions
4. Immunologic phenomena
   a. Glomerulonephritis
   b. Osler's nodes
   c. Roth's spots or rheumatoid factor
5. Microbiologic evidence: positive blood culture but not meeting major criterion as noted previously or serological evidence of active infection with organism consistent with infective endocarditis

Definite endocarditis if the following are present

- Two major criteria or
- One majorcriterion and three minor criteria or
- Five minor criteria

Endocarditis- Valve Vegetation on Echocardiography

- Evaluation of valves for infection is best accomplished through echocardiography
- A transthoracic echocardiogram (TTE) has reduced sensitivity when compared with a transesophageal echocardiogram
- TEE to assess for vegetations and myocardial abscesses but is a less invasive test. Not only can echocardiogram identify new vegetations on valves, which are evidence of infection, but it can also assess the degree of valvular damage and complications such as perivalvular abscesses
- ECG can be used to detect damage to the conducting system. The MC finding is PR prolongation in patients with aortic valve endocarditis and associated perivalvular abscess.

There Are Two Types of Echocardiograms

- Transthoracic echocardiogram (TTE), where the probe is placed on the chest wall, and a TEE, where the probe is inserted into the esophagus.
- The TEE often produces higher-quality images, particularly of aortic and mitral valves, since the TEE probe is closer to the heart itself.

Treatment

- Without treatment, endocarditis is always fatal, so prompt effective therapy is essential. Bactericidal drugs should be used.
- The treatment for endocarditis always includes antimicrobial therapy, and in some cases, surgical removal of the infected valve is indicated as well.
- Empiric therapy for endocarditis is recommended in cases where the patient has hemodynamic instability, severe disease, evidence of embolic disease, or large vegetations.
- Empiric antimicrobial coverage should be active against MRSA, Viridansgroup streptococci, Enterococci and HACEK organisms.
- Common empiric regimens include
  - Vancomycin + Ceftriaxone or Gentamicin
  - Specific antimicrobial therapy should be instituted when the results of blood cultures and antibiotic susceptibility tests are known.
  - Antimicrobial therapy for endocarditis is usually required for 4 to 6 weeks
- Surgical therapy is either indicated or should be strongly considered inpatients with severe congestive heart failure perivalvular abscesses, infections refractory to medical management, and embolic events with large vegetations.

Prevention

- In patients with prior endocarditis a prosthetic heart valve, or select types of CHD, antibiotic prophylaxis is recommend prior to certain procedures.
- Guideline supports giving antibiotics, such as amoxicillin, to these high risk patients at the time of invasive dental procedures, surgery involving respiratory mucosa, or surgery involving infected tissues

MYOCARDITIS

00:43:40

- Myocarditis is infection of the heart muscle.

PATHOPHYSIOLOGY

- Infection of the myocardium most frequently occurs following
  - Hematogenous spread
  - Direct spread from adjacent structures can occur

- Infection and inflammation of myocardium may result in cardiac dysfunction, leading to heart failure.
- Patients with myocarditis present with signs and symptoms of heart failure.
- Depending on the pathogen, the pace of disease progression may be over days or weeks.
- Patients may have signs and symptoms of a systemic infection as well (fever, constitutional symptoms). Those with associated pericarditis often have chest pain

- Related Microbes
  - Viral pathogens are thought to be the predominant cause of infectious myocarditis, although many cases are idiopathic.
  - Include
    - → Coxsackie virus: MCC
    - → CMV
    - → EBV
    - → Parvovirus B19
    - → Influenza virus
  - Other pathogens include
    - → Trypanosoma cruzi: the agent of Chagas disease
    - → Trichinella spiralis: it is viviparous

Diagnosis

- A definitive diagnosis requires cardiac muscle biopsy revealing myocardial inflammation and necrosis.
- However, most cases are presumptively diagnosed in a patient presenting with heart failure, who has (often global) cardiac dysfunction on ECG and elevated cardiac enzymes.
- The ECG may be abnormal and may show ST changes mimicking an acute myocardial infarction

Treatment

- There is no known treatment for most causes of myocarditis, and supportive care is most often given. Patients may ultimately require heart transplant

PERICARDITIS

00:49:56

- Pericarditis refers to inflammation of the pericardium, which can be due to infection, autoimmune diseases, trauma, or malignancy

Pathophysiology

- Pathogens reach the pericardium by either hematogenous spread through the blood or direct spread from adjacent intrathoracic structures or, rarely, directly from infected myocardium.
- Inflammation of the pericardium can result in the formation of pericardial effusion.
- Pericardial effusions can result in cardiac tamponade. Inflammation can also result in a constrictive physiology.
- Certain infections causing pericarditis may also be associated with a concomitant myocarditis

Constrictive Pericarditis

- Chest pain is the MC manifestation of pericarditis. Pain often worsens with inspiration or coughing. Sitting up and leaning forward often improve the pain associated with pericarditis.
- Patients may have fever and constitutional symptoms.
- On examination a friction rub (often consisting of three phases) may be heard when performing auscultation of the heart. This exam finding is very specific for pericarditis.
- Severe infection may result in cardiac tamponade or constrictive cardiac physiology. These patients present with acute or subacute/chronic onset of symptoms of heart failure, respectively.

Pathogens

- Viruses, bacteria, mycobacteria and fungi have all been reported to cause pericarditis.
- Among viral infections: Coxsackie virus and echovirus are MC, although HIV and CMV can cause pericarditis as well.
- Among bacteria: S. aureus and S. Pneumoniae are MC.
- MCC worldwide: MTB
- Clinical presentation is often subacute and may result in a constrictive pattern.
- Several fungi such as Histoplasma capsulatum and Coccidioides immitis can cause pericarditis, which clinically presents similarly to tuberculous pericarditis
- Culture of pericardial fluid or pericardial tissue may reveal causative bacteria. Viruses are rarely isolated
- Additional diagnostic tests can help make the diagnosis include ECG that reveals changes in the PR and ST segments
- If a significant pericardial effusion is present, the ECG may have reduced amplitude in all leads
- An ECG or cardiac magnetic resonance imaging will often reveal a pericardial effusion and/or pericardial thickening.

- In addition, chest X-ray may show an enlarged cardiac silhouette, and cardiac enzymes can be elevated.
- Recovery of a pathogen often requires a pericardiocentesis or pericardial biopsy
- Treatment for infectious pericarditis is dependent on the pathogen.
- Most viral etiologies are treated with symptomatic management and supportive care, whereas bacterial. mycobacterial, and fungal infections will require directed antimicrobial therapy.
- In patients with constrictive pericarditis and tamponade, pericardiocentesis can be life-saving. Untreated bacterial pericarditis is rapidly fatal

Table 25.1

Organism(s)	Percentage of cases								
	Native Valve Endocarditis		Prosthetic Valve endocarditis at indicated time of onset (months) after valve surgery			Endocarditis in IV drug users			
	Community acquired	Health care associated	< 2	2-12	> 12	Right sided	Left sided	Total	CIED
Streptococci	40	13	1	9	31	5	15	12	2
Pneumococci	2	-	-	-	-	-	-	-	-
Enterococci	9	16	8	12	11	2	24	9	4
Staphylococcus aureus	28	52	22	12	18	77	23	57	36
Coagulase –ve staphylococci	5	11	33	32	11	-	-	-	41
Fastidious gram –vecocco bacilli (HACEK group)	3	-	-	-	6	-	-	-	-
Gram –ve bacilli	1	1	13	3	6	5	13	7	6
Candida spp.	<1	1	8	12	1	-	12	4	2
Polymicrobial	3	3	3	6	5	8	10	7	2
Diphtheroids	-	<1	6	-	3	-	-	0.1	1
Culture –ve	9	3	5	6	8	3	3	3	6

26 UPPER RESPIRATORY TRACT INFECTIONS

OTITIS MEDIA

00:00:18

- Otitis media is an Infection of the middle ear caused by either viruses or bacteria.
- Otitis media can be either acute or chronic.
- Any process that leads to eustachian tube obstruction can result in fluid retention and concomitant infection of the middle ear.
- The most common predisposing factors are upper respiratory tract infections and seasonal allergic rhinitis.
- Otitis media is very common in children under the age of 3 years because they have a shorter and narrower eustachian tube that is easily blocked by the inflammation caused by a viral infection or an allergic response

Clinical Manifestations

- Patients present with Ear pain and pressure, often accompanied by an upper respiratory tract infection. In infants, the ear pain may manifest as ear pulling.
- Patients may also complain of decreased hearing and fever.
- On examination, the tympanic membrane is erythematous with a loss of the light reflex and decreased mobility.
- In some cases, the tympanic membrane may bulge.

Pathogens

- Both Bacteria and Viruses cause otitis media
- Among Bacteria, Streptococcus pneumoniae the most common cause. Non-typeable strains of Haemophilus influenzae and Moraxella catarrhalis are also common causes.
- Among viruses, Respiratory syncytial virus, Coronaviruses and Rhinoviruses are commonly involved.

Diagnosis

- Otitis media is usually diagnosed clinically. If the membrane ruptures, a sample of the exudate can be analyzed by Gram stain and culture.
- If indicated, tympanocentesis can be done to relieve pressure before the drum ruptures and to obtain a specimen for culture

Treatment

- Amoxicillin orally is usually the drug of choice together with nasal decongestants to open the eustachian tube.
- In cases of bacterial resistance, Amoxicillin-clavulanate (Augmentin) may be used.
- Ventilating tubes may be inserted as a strategy to prevent recurrent infections.
- The conjugate pneumococcal [illegible] is effective in preventing invasive pneumococcal disease but is less effective in preventing otitis media

SINUSITIS

00:07:00

- Sinusitis is inflammation of the paranasal sinuses. It can be either acute or chronic.
- Acute infections are considered those with symptoms lasting less than 4 weeks. The information here refers to acute sinusitis.

Pathophysiology

- Impaired mucociliary clearance caused by viral infection or allergic rhinitis can obstruct the orifice of the sinus.
- Mucus then accumulates in the sinus cavity. Stasis can lead to bacterial overgrowth and super infection.
- Sinusitis frequently involves the maxillary sinus because the ostium of that sinus is located superior to most of the sinus and drainage of mucus has to occur against gravity
- Drainage of the other sinuses is aided by gravity

Clinical manifestations

- Purulent nasal discharge.
- Nasal congestion
- Facial or anus pain,
- Decreased sense of smell and fever.
- Headache and malodorous breath may be present.

Pathogens

- Many cases begin with viral upper respiratory tract infection and later bacterial super infection can occur.
- In the case of acute bacterial sinusitis, common organisms are
  - S. pneumoniae.
  - H. influenzae, and M. catarrhalis, as in the case of acute otitis media
  - Staphylococcus aureus also causes sinusitis but less commonly.
- In immunocompromised patients and diabetics, sinusitis caused by fungi such as Aspergillus or Mucor may occur

Diagnosis

- Sinusitis is often diagnosed based on a typical constellation of symptoms and clinical findings.
- Computed tomography scan of the sinuses is very sensitive modality for indicating inflammatory processes of the sinus.
- However, in the absence of bony destruction, these are nonspecific findings for diagnosing clinically significant sinusitis requiring antibiotic therapy

Treatment

- If symptoms are severe, antibiotics are given in concert with intranasal corticosteroids, as well as nasal decongestants.
- Amoxicillin is the drug of choice, but if resistance is a concern, then amoxicillin-clavulanate (Augmentin) is used.
- In mild cases, antibiotics are not normally used unless then symptoms have lasted for longer than 10 to 14 days.

PHARYNGITIS

00:12:27

- Pharyngitis is inflammation of the throat caused primarily by viruses.
- Approximately 10% of cases of pharyngitis are caused by S. pyogenes
- Streptococcal pharyngitis (strep throat) is important because poststreptococcal immune sequelae, such as rheumatic fever, may occur.

Clinical Manifestations

- Sore throat worsens on swallowing.
- Fever may also be present.
- Typical symptoms a/w an upper respiratory tract infection rhinorrhea, sinus tenderness, ear pain, cough may accompany the sore throat
- On examination
  - An inflamed pharynx, Tonsils, and Palate are typically seen
  - A grayish exudate is often present on the tonsils
  - Tender, anterior cervical lymphadenopathy
  - Petechiae on the palate may also be a diagnostic clue for Group-A Strepto
- S. Pyogenes: most important bacterial cause.
- Group C and G streptococci also associated
- Pharyngitis caused by Neisseria gonorrhoeae is likely to be the result of sexual activity and if it occurs in children, is considered as a sign of child abuse.
- Mycoplasma pneumoniae, Chlamydia pneumoniae, and Arcanobacteriumhaemolyticum also cause pharyngitis.
- In certain countries where the diphtheriae vaccine is not widely used, Corynebacterium diphtheriae is a significant cause of pharyngitis, often accompanied by a pseudomembrane.
- Fusobacterium necrophorum, a gram –negative anaerobe can cause pharyngitis accompanied by septic thrombophlebitis (Lemierre's syndrome)
- Note that although S. pneumoniae and H. influenza colonize the oropharynx, they do not cause pharyngitis.

Viruses

- Most cases of pharyngitis are caused by respiratory viruses, such as Adenovirus, Influenza A & B viruses, Parainfluenza viruses, Rhinovirus and Coronavirus.
- Other viral causes include: Coxsackie virus (herpangina), Epstein-Barr virus (infectious mononucleosis), and herpes simplex virus (especially [illegible])
- HIV causes and acute retroviral syndrome that includes pharyngitis as one of its components

Diagnosis

- Throat swab
- Tonsillar exudates
- Tender anterior cervical Lymphadenopathy
- Fever and absence of cough.
- Rapid antigen detection tests for GAS
- A throat culture is the most reliable method of determining whether S. pyogenes is the cause

Treatment

- If GAS is diagnosed: Penicillin G, penicillin V or amoxicillin
- In penicillin allergic patients: Erythromycin or cephalexin can be used.

Prevention

- Vaccine against C. diphtheriae and Influenza virus
- Long-term carriers of GAS should not be treated
- Note that children who have RHD should receive penicillin orally for many years to prevent infection by S. pyogenes. Which could cause a flare of their RHD

COMMON COLD

00:21:11

- Viral infection of the upper respiratory tract, including some or all of the following structures: the Nose, Throat, Sinuses, Eustachian tubes, Trachea, and larynx.

Pathophysiology

- The viruses that cause the common cold are transmitted primarily by aerosols generated by sneezing or by direct contact.
- Direct contact involves either hand-to hand contact or hand-to-surface contact Eg. Rhinovirus & Adenovirus

Clinical manifestations

- Nasal congestion
- Decreased sense of smell

- Rhinorrhea (watery nasal discharge without purulence).
- Sneezing
- Malaise and sore throat.
- Headache

Pathogens

- Rhinovirus (> 100 serotypes) are the most common etiology (up to 50%).
- Coronaviruses
- Adenoviruses
- Enteroviruses: Coxsackie viruses
- Para influenza virus and respiratory syncytial virus are also possible causes but they primarily causes other diseases such as croup and broncheolitis, respectively

Diagnosis

- The common cold is usually diagnosed clinically.
- Erythematous and edematous nasal mucosa is seen on physical examination.
- Conjunctival and pharyngeal infection

Treatment

- Generally Symptomatic
- Zing acetate in doses > 75 mg/day may reduce the duration of symptoms
- Other strategies: oral decongestants and buffered hypertonic saline nasal irrigation.
- If used for more than a few - days, nasal sprays can be used.

CROUP

00:25:35

- Croup is an inflammation of the larynx, trachea, and large bronchi (laryngotracheobronchitis).

Clinical Manifestations

- Inspiratory stridor is the key finding, together with a barking cough and a hoarse voice
- Symptoms may begin in a subtle fashion with nasal irritation and congestion and then rapidly progress to stridor over a day.

Pathogens

- Parainfluenza viruses, especially type 1 are the most common cause.
- Respiratory syncytial virus
- Influenza virus

} 1% to 10% of cases

Diagnosis

- The diagnosis is usually made clinically. Plain radiographs may show a 'steeple sign"
- Subglottic tracheal narrowing results in an inverted "V" shape

Treatment

- Patients with moderate to severe symptoms may be given corticosteroids (Dexamethasone), with or without epinephrine
- Thereis no antiviral drug therapy

LARYNGITIS

00:27:26

- Laryngitis is inflammation of the vocal folds of the larynx.

Clinical manifestations

- Hoarseness
- Inability to speak (aphonia)
- Laryngitis may be accompanied or preceded by an upper respiratory infection.
- Etiology
  - MCC: Parainfluenza viruses and Rhinoviruses
  - Other causes
    - → Influenza virus
    - → Adenovirus
    - → Coronavirus
  - Bacteria such as
    - → S. pyogenes
    - → M. catarrhalis.
    - → H. influenzae

Diagnosis

- Done Clinically

Treatment

- Hydration and voice test
- Antibiotics are not needed

EPIGLOTTITIS

00:28:39

- Epiglottitis is an inflammation of the epiglottis.

Clinical Manifestations

- Rapidly worsening sore throat
- Odynophagia (pain on swallowing)
- Dysphasia (difficulty in swallowing)
- Pain may be out of proportion to physical examination findings.
- Airway obstruction can occur in severe cases

Treatment

- Epiglottitis in young children should be treated as a medical emergency

Pathogens

- MCC H. influenzae typeB
- Less common pathogens include other H. influenzae, S. pneumoniae, S. pyogenes and S. aureus

Diagnosis

- Is made by visualization of the epiglottis.
- If indirect laryngoscopy (done primarily in children) is performed, a swollen and erythematous "cherry-red" epiglottis may be visualized.
- On lateral plain X-rays an enlarged epiglottis may be seen as a "thumb" sign

Microorganismscausing URTI and associated symptoms

00:31:00

Rhinitis common cold	Pharyngitis (sore throat), and tonsillitis	Laryngitis	Laryngotracheobronchitis
MC caused by viruses • Rhinovirus • Coronavirus • Adenovirus • Influenza virus • Human metapneumovirus • Respiratory syncytial virus	**Symptoms** • Pharynx and/or tonsils become inflamed, red, swollen, and show exudates, and sometimes a membrane is formed **Viruses** (MC causes) • Influenza virus • Parainfluenza virus • Coxsackievirus A • Rhinovirus • Coronavirus • Epstein-Barr virus • Adenoviruses	**Symptoms** • Hoarseness of voice • Lowering & deepening of voice **Mostly viral agents** • Influenzae virus • Parainfluenzae virus • Rhinovirus • Adenovirus • Coronavirus • Human metapneumovirus	• Age- Children, < 3 years age **Symptoms** • Inspiratory stridor (high-pitched breath sounds resulting from turbulent air flow in the larynx) • Hoarseness • Fever • Cough (harsh, barking non-productive) **Agents** • Parainfluenza virus (MC) • Influenza virus • Respiratory syncytial virus • adenoviruses
Sinusitis • Inflammation of the sinuses (d/t allergy or infections) **Symptoms** • headache/ facial pain • thick nasal mucus	**Bacteria** • streptococcus pyogenes (MC bacterial cause) • streptococcus group C & G • mycoplasma pneumonia • Vincent angina: Borrelia Vincentii Leptotrichiabuccalis **Fungal** • Candida albicans	**If membrane or exudates present** • Streptococcus pyogenes • C. diphtheriae • Epstein-Barr virus	**Epiglottitis** • Edema and inflammation of epiglottis & soft tissue above vocal cords • Age: Children 2-6 years **Symptoms:** • Fever • Difficulty in swallowing • Inspiratory stridor **MC agent:** • Haemophilus influenza type b

27 LOWER RESPIRATORY TRACT INFECTIONS

Introduction 00:00:19

- Lower respiratory tract infections are an important cause of morbidity and mortality worldwide in children and in adults.
- Anatomic approach to lower respiratory tract infections, moving from the large bronchi (bronchitis) down to the very small bronchioles (bronchiolitis) and then into the alveoli where pneumonia occurs

BRONCHITIS 00:01:09

- It self-limited inflammation of the bronchi
- Acute bronchitis must be distinguished from chronic bronchitis in which patients have a cough for more than 3 months.
- The coughing so characteristic of bronchitis is an attempt to clear the mucus produced by the inflammatory response to viral infection.
- Bronchitis occurs more often in the winter months than in the summer.
- Smoking predisposes to bronchitis (and pneumonia) by damaging the cilia in the bronchi, leading to an inability to clear mucus from the respiratory tract
- Cough is the most prominent symptom of bronchitis.initially, bronchitis presents with the symptoms of an upper respiratory infection, namely, nasal congestion, scratchy sore throat and perhaps a low-grade fever.
- Physical examination typically reveals expiratory wheezes However, if cough persists for > 5 days & pneumonia has been ruled out, acute bronchitis should be suspected
- Bronchitis is self-limited & usually resolves in 1 to 2 wks. However, cough may persist for several more wks d/t airway hyperreactivity
- Respiratory viruses are the MC pathogens (Influenza A & B, parainfluenza virus, coronavirus, rhiovirus, respiratory syncytial virus [RSV], and human metapneumovirus).
- Bacterial pathogens are not thought to play a significant role in acute bronchitis
- The diagnosis is primarily made clinically.
- Cough, with or without sputum production, which may persist for > 5 days, is the typical presentation.
- Patients are usually afebrile but may have a low-grade fever. Suptum cultures are typically not done.
- In patients with chronic cardiorespiratory disease, a rapid antigen test for influenza virus may be useful because oseltamivir can shorten the duration & intensity of symptoms
- Because treatment of both upper respiratory infections & acute bronchitis is largely supportive, these distinctions may have less clinical significance.
- What may be more important clinically is to distinguish acute bronchitis (usually viral) from pneumonia (mainly bacterial), which does require antimicrobial therapy.
- A chest radiograph may be performed to determine whether pneumonia is present
- Treatment involves reassurance and symptom relief with agents such as nonsteroidal anti-inflammatory drugs
- Influenza vaccine can prevent bronchitis and pneumonia caused by influenza A and B viruse.
- The neuraminidase inhibitor oseltamivir (Tamiflu) should be given to unimmunized individuals with chronic cardiorespiratory disease
- Handwashing is recommended to reduce the carriage of respiratory viruses

BRONCHIOLITIS 00:08:30

- Bronchiolitis is inflammation of the bronchioles – the small airways < 2 mm in diameter
- Bronchiolitis among infants & young children is primarily infectious
- Particularly among children under 2 yrs of age viruses can directly damage the epithelial cells of the terminal bronchioles, causing inflammation & obstruction of the small airways. Prematurity is an important predisposing factor
- Usually children initially have symptoms consistent with an upper respiratory tract infection and then are noticed to have increased respiratory distress
- Children under 2 yrs old in particular may have tachypnea, wheezing, nasal flaring, and chest retractions.
- In severe cases, hypoxia, apnea and respiratory failure may ensue. In most cases, recovery occurs in 1 to 2 wks
- RSV is the MC pathogen. Other etiologies include influenza virus, parainfluenza virus, adenovirus, coronavirus, rhinovirus and human metapneumovirus
- In children, viruses are the main eitiolgyof broncheolitis. Bacteria are not through to be involved. In adults, the causes are more varied and range from viruses, to inhaled toxic chemicals in the work place, to idiopathic causes

- Bronchiolitis caused by RSV occurs primarily in the winter months
- The diagnosis is primarily clinical. Upper respiratory tract infection symptoms followed by lower respiratory tract symptoms and signs (e.g., nasal flaring, wheezing) in a young child during the winter would be very suggestive of bronchiolitis.
- Chest radiograph typically shows hyperinflation of the lungs.
- An enzyme immunoassay (EIA) for RSV antigen in respiratory secretions is available for diagnosis in hospitalized patients.
- A PCR assay that detects the RNA of RSV is also available.
- Self-limited disease in most cases, general supportive measures are adequate in most cases
- Patients with moderate or severe respiratory distress will require hospitalization.
- Ribavirin, delivered by aerosol into the lungs is approved for severe disease caused by RSV, but it's use is limited to hospitalized infants. Inhaled bronchodilators may be useful
- Hand washing to minimize transmission of pathogens is an important strategy
- Palivizumab is a humanized monoclonal antibody against the RSV F (fusion) envelope protein that may be used in certain populations to decrease the risk of diseasecausedby RSV (population include children with bronchopulmonary dysplasia and congenital heart disease and prematurely born infants

PNEUMONIA

00:16:58

- Pneumonia is an inflammation of the lung affecting the alveoli.
- Pneumonia is community acquired Vs hospital acquired to help us determine the spectrum of potential pathogens that differs based on setting
- Empiric therapy is often given in pneumonia because of this reason
- Hospital-acquired pneumonia, also known as nosocomial pneumonia, is pneumonia that occurs 48 hours or more after admission to the hospital and was not present at the time of admission.
- The alveoli of the lungs are continually exposed to microbes from the environment via the upper respiratory tract.
- Our host defenses usually keep these potential pathogens in check. However, disease can occur when there is a particularly virulent organism, when there is large burden of organisms inhaled from the environment or aspirated from the oropharynx, or when there is a defect in host immunity.

Predisposing factors to pneumonia include

- Extremes of age
- Chronic obstructive pulmonary disease (COPD)
- Chronic bronchitis
- Diabetes mellitus
- Cystic fibrosis
- Congestive heart failure
- IV drugs users, Alcoholics, and Seizure disorders have a high risk of pneumonia because they can aspirate organisms into the lung when unconscious
- People exposed to water aerosols, especially from air conditioners, are at risk for pneumonia caused by Legionella
- Hospitalized patients in the ICU are at risk for ventilator-associated pneumonia by Gram –ve rods such asE. coli, Pseudomonas & Acinetobacter

ClinicalManifestations

- Symptoms include
  - Cough with sputum
  - Fever with chills
  - Chest pain
  - Shortness of Breath
  - Rusty sputum
  - Currant Jelly sputum seen in pneumonia caused by Klebsiella

Physical examination findings

- Tachypnea
- Rales
- Rhonchi
- Dullness on percussion: if lung is consolidated
- Patients who areintubated and who acquire a nosocomial pneumonia may only havefever as a presenting sign
- Pneumonia may be complicated by an infected pleural effusion or a pleural empyema
- A pleural empyema is walled-off collection of pus in the pleural space

Pathogens

- MCC of community-acquired pneumonia Streptococcus pneumoniae
- Other common bacterial pathogens include Klebsiella pneumoniae and H. influenzae.

Important Information

- Non-typeable strains of H. influenzae that cause pneumonia in elderly patients with COPD rather than type B strain

- Other pathogens include Mycoplasma pneumoniae, Legionella species, and Chlamydophila pneumoniae
- Infection with MTB can also manifest as pneumonia.

Important Information

- Approximately 30% of adults with community acquired pneumonia, no pathogen, neither bacteria nor virus is isolated

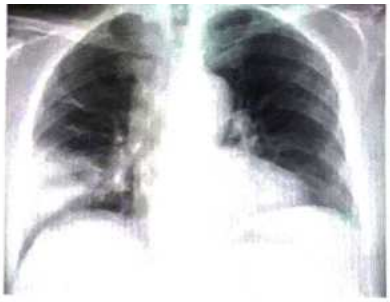
- Causes of pneumonia in a neonate are those acquired during passage through the birth canal.
- The main cause of pneumonia in an infant, Chlamydia trachomatis, is also acquired during passage through the birth canal but is a less aggressive pathogen so its onset in delayed.
- M. pneumoniae is the MCC in young adults

Age	Bacteria	Viruses
• Neonates	• Group B streptococci • E. coli	• Respiratory syncytial virus (RSV)
• Infants	• C. trachomatis • S. pneumonia	• RSV • Parainfluenza virus
• Children	• S. pneumoniae • Haemophilua influenzae	• RSV • Parainfluenza virus
• Young adults	• M. pneumoniae • C.pneumoniae • S. pneumoniae	• Various respiratory viruses (eg. adenovirus)
• Older adults	• S. pneumonia • H. influenzae • Legionella pneumophile	• Influenza virus

Predisposingfactors	Typical Pathogens
• Alcoholism	• Klebsiella pneumonia, oral anaerobes
• Bird exposure, especially psittacine birds such as parrots (psittacosis)	• Chlamydophila psittaci
• COPD including smoking related	• H. influenzae
• Cystic fibrosis	• Pseudomonas aeruginosa
• Imported wool, spores in wool (wool-sorter's disease)	• B. anthrax
• Influenzae virus infection	• S. aureus
• Intubation, postsurgery and ICU	• Coliforms: P. aeruginosa, S. aureus
• Mouse dropping exposure, especially in southwestern states	• Hantavirus
• Sheep exposure, especially placentaltissue (Q fever)	• C. burnetii
• Travel to or reside in central valley of California, Arizona, or New Mexico	• Coccidioides immitis
• Travel to or reside in Ohio or Mississippi river valleys	• Histoplasma capsulatum
• Ventilator-associated, especially in ICU	• Acinetobacter species
• Water aerosols, especially from air conditioners	• Legionella pneumonia

Diagnosis

- The "gold standard" for a diagnosis of pneumonia is an infiltrate on a plain chest radiograph
- Clinical data may help, but ultimately the chest radiograph is the most important diagnostic tool
- Sputum analysis for Gram stain and culture and blood cultures may be helpful in the hospitalized patient but are only optional in an outpatient setting because therapy is largely empiric for community acquired pneumonia
- In pneumonia caused by one of the encapsulated pyogenic bacteria such as S. pneumoniae
- White blood cell count is frequently elevated and the number of neutrophils is often increased



Lobar pneumonia caused by S. pneumoniae
Area of consolidation in right lung

Curb-65 Score 00:42:33

- The prediction of prognosis of CAP in an adult is done by CURB-65 score
  - C (Confusion) = 1 point
  - U (blood urea nitrogen > 19 mg/dL) = 1 point
  - R (respiratory rate > 30 mint) = 1 point
  - B (BP < 90/60) = 1 point
  - 65 (Age >65 yrs) = 1 point
  - Higher the score, greater is the mortality
  - If the score < 1, outpatient therapy is indicated
  - If the score > 1, patient should be hospitalized

Clinical Pulmonary Infection Score (CPIS) used for ventilator associated pneumonia (VAP) 00:45:35

Refer Table 27.1

- It is important that sputum (not saliva) be sent to the lab for Gram stain and culture.
- If the specimen contains many neutrophils and few epithelial cells, then the specimen is likely to be sputum and will be analyzed
- If, however, the specimen contains many epithelial cells and few neutrophils, then the specimen is saliva and will be rejected by the lab

Refer Table 27.2

Identification features/ detection methods of common organisms causing pneumonia

- Treatment for community-acquired pneumonia is largely empiric because microbiologic diagnostic strategies are generally insensitive.
- Outpatients are generally treated with
  - Macrolides → Azithromycin
  - Tetracycline → Doxycycline
  - Respiratory Quinolone → Levofloxacin
- Inpatients: Ceftriaxone + Macrolide/Respiratory Quinolone Monothrerapy
- Patients with suspected hospital-acquired pneumonia may be given broader spectrum agents such as a Carbapenem depending on the local epidemiology, given that many hospital-acquired infections are multidrug resistant
- Prompt initiation of antibiotics is important because morbidity and mortality increase after a delay of more than 8 hours
- Drainage of an empyema or infected pleural fluid should be performed
- The influenza vaccine is effective in decreasing the likelihood of pneumonia.
- The Pneumococcal Polysaccharide (nonconjugate) Vaccine available for older adults is important in decreasing bacteremia associated with pneumonia and mortality but not the likelihood of pneumonia itself.
- Because it is the Non-typeable strains of H. influenzae, rather than the type B strain, that are the MCC of pneumonia, the conjugate vaccine against type B is not an important source of protection.
- Smoking cessation and treatment of alcohol abuse may also decrease pneumonia risk

Organism Causing Lower Respiratory Tract Infections (LRTI) 01:05:40

Refer Table 27.3

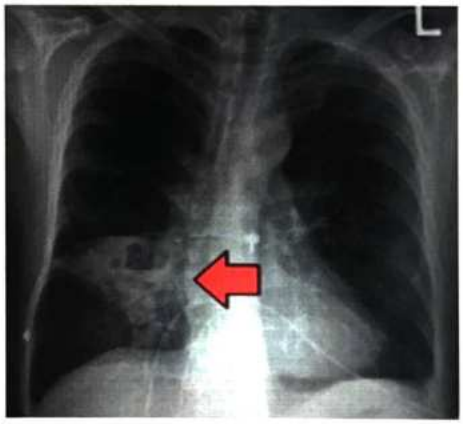
LUNG ABSCESS 01:15:02

- Lung abscess in a necrotic process within the lung parenchyma that frequently results in a cavity with an air-fluid level
- Patients may aspirate oropharyngeal bacteria into the lower airways and alveoli. This usually occurs when the patient is in the recumbent position and cannot clear secretions.
- For example, aspiration can occur when a person is unconscious from drug overdose, excess alcohol intake, or the anesthesia that accompanies surgery
- Poor oral hygiene is a common predisposing factor. A pneumonitis may first occur, but this can progress to necrosis in a week or so.
- A lung abscess caused by S. aureus may infect the lung via the bloodstream from a distant site of infection such as right-sided endocarditis in an intravenous drug user
- Patients present with symptoms typical of pneumonia with fever and productive cough.
- The sputum is often foul smelling, indicating the presence of anaerobes
- These symptoms may be indolent and progress over a period of wks. Systemic symptoms such as night sweats, fatigue and weight loss may also be present
- The MC organisms are anaerobes or mixed aerobes and anaerobes that are part of the oral flora.

- Anaeorbes commonly involved include Peptostreptococcus species, Prevotella species, and Fusobacterium nucleatum.
- Aerobes include Streptococcus milleri and S. aureus. (Clinicians often use the term "aerobe" rather than facultative to describe bacteria that are not anaerobic.)

Diagnosis

- A chest radiograph shows a pulmonary infiltrate with a cavity, often with an air-fluid level
- An air-fluid level occurs when the abscess erodes a bronchus and some of the pus in the abscess is coughed up and replaced by air
- Pleural fluid, if present, and blood culture may provide microbiologic data, but anaerobes may be difficult to identify



- Image shows Lung Abscess
- Arrow points to air-fluid interface within the abscess.

Treatment

- Clindamycin or ampicillin- sulbactam are typical treatment options. Duration of therapy is usually 4-6 weeks
- Patients who do not respond to antibiotics will required surgical drainage

Case scenario

Q. An 8 yr old girl admitted with complaints of productive cough chest pain, shortness of breath since 3 days. On clinical examination dullness over left sixth Inter costal space on percussion, Crepitation & Rales over sixth ICS on auscultation. On Chest X ray: Homogeneous Ground Glass opacities in the left lower lobe. Sputum is collected & subjected to microscopy & Culture. What is the Diagnosis?

Diagnosis: Typical Case of Pneumonia

- Productive cough, Ground glass opacities, Crepitations & rales indicate LRTI

Table 27.1

Parameter (s)	Score 0	Score 1	Score 2
• Temperature (°C)	• 36.5°C &38.4°C	• 38.5°C &38.9°C	• 39°C &36.4°C
• Leukocytosis	• 4000 &11,000	• < 4000 &> 12,000	
• Tracheal aspirate	• None	• Non-purulent	• Purulent
• Oxygenation (PaO_2/FIO_2 mmHg)	• > 250 or ARDS		• 250 & no ARDS
• Chest radiograph	• No opacity	• Diffuse (patchy) opacity	• Localized opacity
• Progressive radiological progression	• No radiological progression		• Radiological progression
• Culture of tracheal aspirate	• Pathogenic bacteria light or no growth	• Pathogenic bacteria moderate of heavy growth	
• Culture – Gram stain	• Different morphology	• Same morphology as Gram	

Table 27.2

Agents of pneumonia	Direct demonstration in sputum	Culture identification
Lobar pneumonia		
Streptococcus pneumonia	Pus cells > 25/ LPF and epithelial cell < 5/ LPF Gram +ve cocci in pair, lanceolate shaped	• Alpha haemolytic, draughts man-shaped colonies on blood agar, sensitive to optochin • Bile soluble, ferments insulin
Haemophilus influenzae	Pleomorphic Gram –ve bacilli	Satellitism on blood agar with S. aureus streak line
S. aureus	Gram +ve cocci in clusters	BA: golden yellow haemolytic colonies catalase +ve, Coagulase +ve
Gram –ve bacilli: E. coli, Klebsiella, Pseudomonas	Gram –ve bacilli	Identification is based on • Growth on MacConkey agar (LF or NLF colonies) • Biochemical reactions (ICUT: Indole, Citrate, Urease, TSI)
Interstitial or atypical pneumonia		
Chlamydophila pneumoniae	Direct immunofluorescence test Antigen detection by enzyme immunoassay Nucleic and amplification test (NAAT) detecting specific genes	Serology-antibody detection by • CFT using LPS antigen • ELISA using recombinant LPS antigen • Micro- IF test using outer membrane protein antigen
Mycoplasma pneumoniae	Direct immunofluorescence test Capture ELISA – detecting antigen (P1 adhesin) PCR targeting P1 adhesin gene	Cuture- fried egg colonies on PPLO agar Antibody detection • Non-specific test (cold agglutination test) • Specific test (e.g. ELISA)
Legionella pneumophila	Pus cells > 25/LPF & epithelial cells < 5/LPF Detection of specific antigen in sputum, urine	Growth on BCYE medium
Viral pneumonia	Detection of specific viral antigen in sputum Detection of specific viral genes in sputum (PCR)	

Table 27.3

Pneumonia		Bronchitis

Community acquired
 	
No co-morbidity
• Streptococcus pneumoniae (MC)
• Atypical pathogens
  ○ Chlamydophila pneumoniae and C. psittaci
  ○ Legionella and Mycoplasma
  ○ Coxiella burnetti (Q fever)
  ○ Viral pneumonia (influenza, adenovirus, parainfluenza, RSV)
 	
Co-morbidity
• Alcoholism: S. pneumoniae, H. influenzae
• COPD: H. influenzae, M. catarrhalis, S. pneumoniae
• Post – CVA-aspiration: S. pneumonia
	Hospital acquired
	
	Bacteria agents
	• Gram –ve bacilli (MC)
	  ○ MDR non-fermenters(Pseudomonas & Acinetobacter)
	  ○ MDR Enterobacteriaceae E. coli, Klebsiella & Enterobacter
	• Staphylococcus aureus (both MRSA & MSSA)
	• S. pneumoniae (rarely, in early stage)
	
	Viral agents
	• Influenza, adenovirus, parainfluenza RSV
		Bacterial agents
		• B. pertussis
		• B. parapertusis
		• M. pneumoniae
		• Chlamydophila
		
		Viral agents
		• Influenza virus
		• Adeno viruses
		• Rhino viruses
		• Corona viruses
		
		Bronchiolitis
		
		Viral agents
		• Respiratory syncytial viruses
		• Parainfluenza virus
		• Rhino virus
		• Influenza virus
		• Adeno virus
		• Entero virus
		• Human metapneumo virus

- Post-obstruction of bronchi: S. pneumoniae, anaerobes
- Post-influenza: S. pneumoniae, S. aureus
- MCC of CAP → S. pneumoniae
- MCC of Atypical Pneumonia → M pneumoniae

28 CENTRAL NERVOUS SYSTEM INFECTIONS

Introduction 00:00:13

- CNS infections are often life-threatening and can have severe sequelae
- These infection leads to inflammation and edema resulting in damage to brain tissue & loss of function
- MCC: Bacteria, Virus
- Others
  - Fungi
  - Protozoa
  - Helminthes

MENINGITIS 00:02:00

- Meningitis is an infection of the Meninges, the membranes that line the brain and spinal cord
- Types (Depending on speed of the initial presentation and the rate of progression of the illness)
  - Acute
  - Subacute
  - Chronic

Acute Meningitis 00:03:05

- Is caused by either
  - Pyogenic bacteria such as: S. pneumoniae and N. meningitides
  - Virus such as: Coxsackie virus and HSV type 2
- Viral Meningitis is also known as aseptic meningitis because routine culture for bacterial pathogens are negative

Subacute Meningitis

- Is caused by
  - MTB
  - Fungi such as Cryptococcus
- The causative organism are often found in the spinal fluid located in the subarachnoid space

Pathophysiology

- Hematogenous spread (MC)
- Direct spread via adjacent infections
- Via neurosurgery: CSF shunt
- Via trauma
- The importance of hematogenous spread is emphasized by the success of the conjugate vaccines against S. pneumoniae, N. meningitidis, Haemophilus influenzae type B that induce circulating IgG antibodies which neutralize the bacteria in the blood
- Acute bacterial Meningitis begins with Nasopharyngeal colonization f/b Hematogenous spread
- This is followed by an inflammatory response that causes many of the clinical manifestations, especially the edema resulting in increased intracranial pressure leading to headache
- Cerebral vasculitis and Cerebral infarction can also occur

Pathogenesis of Bacterial Meningitis

Refer Diagram 28.1

Clinical Manifestations

- Early symptoms include Fever, headache, stiff neck & photophobia
- If Untreated meningitis may progress to Vomiting, seizures, focal neurologic deficits& altered mental status
- N. meningitidis infection can be associated with
  - Rash
  - Acute meningococcemia a/w purpura fulminans

Purulent Meningitis

Purpura Fulminans

- MCC of pyogenic meningitis: S. pneumoniae
- Other causes
  - Meningococci (25%)
  - S. agalactiae (15%)
  - Listeria (10%)

- H. influenzae (<10%)
- Neonates
  - S. agalactiae
  - GNB such as E.coli, Klebsiella and Listeria
- Elderly (>60 yrs)
  - Pneumococcus
  - S. agalactiae
  - Listeria monocytogenes
- Less common pathogens: Borrelia Burgdorferi (Lyme's disease) and Treponema pallidum (syphilis)

Acute Viral pathogens 00:21:14

- MCC: Enterovirus (Coxsackie virus & Echovirus)
- Enteroviral meningitis occurs primarily in young children and the peak incidence is inthe summer seasons
- HSV type 2 is also a common cause of meningitis
- Note that HSV-2 typically causes meningitis, whereas HSV-1 causes encephalitis
- Primary genital infections with HSV-2 are more likely to result in meningitis than Recurrent HSV-2 infections
- Primary reactivation of Varicella zoster virus (VZV) infection can also be associated with meningitis
- Although Arbovirus typically cause encephalitis, Arboviruses such as West Nile virus (WNV) and St. Louis encephalitis virus can also cause meningitis
- Mumps virus used to be a common cause of meningitis

Subacute and Chronic Meningitis 00:25:05

- MCC of subacute and chronic meningitis are MTB and Fungi such as Cryptococcus, Coccidioides & Histoplasma
- Cryptococcal meningitis occurs most commonly in immunocompromised patients, such as those with acquired immunodeficiency syndrome (AIDS)

Normal CSF Findings

- CSF pressure : 50-150 mm of water
- Total leukocyte count : 0-5/mm^3
- Predominant Cell Type : Lymphocytes
- Glucose : 40-70mg%
- Total proteins : 15-45 mg%

CSF analysis in pyogenic meningitis

Opening pressure	>180 mmH_2O
White blood cells	10/µL to 10,000/µL; neutrophils predominate
Red blood cells	Absent in nontraumatic tap
Glucose	<2.2 mmol/L (<40 mg/dL)
CSF/ serum glucose	<0.4
Protein	>0.45 g/L (>45 mg/dL)
Gram's stain	Positive in > 60%
Culture	Positive in > 80%
PCR	Detects bacterial DNA
Latex agglutination	+ve in patients with meningitis due to S. pneumoniae, N. meningitides, H. influenzae type b, E. coli, group B streptococci
Limulus lysate	+ve in cases of gram –ve meningitis

Identification features/ detection methods of common organisms causing meningitis

Refer Table 28.1

- Lumbar puncture should not be performed if there are increased signs of ICT eg. Papilledema or focal neurologic signs, as can lead to herniation of brainstem & death may occur
- A CT scan should be performed prior to the lumbar puncture

Treatment 00:49:12

- Empiric therapy for acute bacterial meningitis
- Drugs with excellent penetration to the CSF & that are bactericidal are used
  - Ceftriaxone/ cefotaxime + vancomycin (older children & adults)
  - Ampicillin should be added if Listeria is a likely cause
- Empiric therapy for neonatal bacterial meningitis
  - Ampicillin + ceftriaxone or cefotaxime
- Acyclovir is used for the treatment of HSV and VZV infection

Prevention

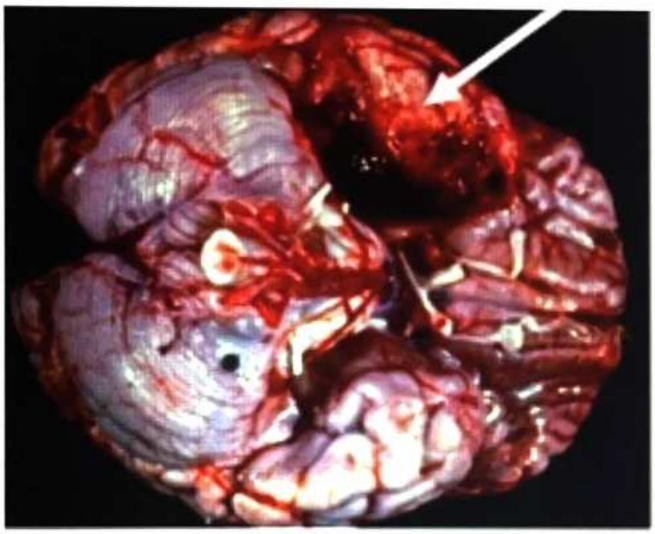
- Immunization
- Pre exposure and post exposure chemoprophylaxis
- Vaccines: Conjugate vaccines such as
  - Pneumococcal vaccine (Protects against 13

serotypes)
  - Meningococcal vaccine (Protects against 4 serotypes)
  - H. influenzae vaccine (Protects against only type B serotype)
- Pre exposure chemoprophylaxis against S. agalactiae to reduce vaginal carriage in mother
- If vaginal or rectal cultures are positive at 35 to 37 weeks of gestation then Ampicillin should be given
- Post exposure chemoprophylaxis aimed at reducing nasopharyngeal carriage of N. meningitidis and H. influenzae type B.
- Close contacts of patients with meningitis should receive ciprofloxacin for Neisseria or Rifampicin for Haemophilus

ENCEPHALITIS

00:54:07

- Encephalitis is an infection of the brain parenchyma predominantly caused by viruses
- Meningoencephalitis: when both brain and meninges are involved



Viral Encephalitis

Clinical Manifestations

- Most characteristic clinical manifestation of encephalitis include Fever, Headache, and Altered mental status as well as Seizures and Focal neurological deficits
- Rabies encephalitis has two clinical manifestations
  - Most cases of rabies (80%) presents with Hyperactivity, Agitation, Delirium, Hydrophobia, and Seizures (called as furious rabies)
  - Dumb rabies (20%): presents with Paralytic symptoms in which an Ascending paralysis without hyperactivity
- Coma and death are common in both forms

Pathogens

- Viruses are main cause of Encephalitis
  - Approximately 15% are caused by HSV-1
  - Arboviruses (5%) such as west nile viruses, Rabies virus is not so rare cause
  - VZV, CMV and EBV also cause encephalitis
- WNV is the most common arboviral cause of encephalitis is the US
  - 80% WNV are asymptomatic.
  - 20% WNV develop flu-like illness
  - < 1% develop CNS disease, of which half have Encephalitis
- Other Arboviruses that causes encephalitis with some frequency are St. Louis encephalitis virus, the LaCrosse strain of California encephalitis virus, and Eastern and Western equine encephalitis viruses (EEE and WEE respectively)
- They are all transmitted by either Culex or Aedes mosquitoes. Post infection encephalitis follows immunization or infection caused most often by VZV, measles, and influenza

Diagnosis

- Mild elevation of CSF lymphocytes
- Elevation of proteins
- Glucose normal
- PCR &ELISA is done

- Dx of Rabies
  - Direct fluorescent Ab staining (biopsy of skin from nape of neck)
  - PCR Assay (CSF, Saliva or Tissue)
    → Identifies animal reservoir & geographical location of virus
- Radiological findings can be usefull as well
  - On MRI: Temporal lobe abnormalities can be seen in HSV encephalitis

Treatment

- Rx of choice for HSV-1, HSV-2 & VZV: IV Acyclovir
- No Antiviral therapy for Arbovirus & Rabies Encephalitis

Prevention

- Prevention of rabies include both pre exposure and post exposure prophylaxis
- Pre exposure prophylaxis vaccine should be given to veterinarians and others at risk of exposure
- Post exposure prophylaxis consists of both the vaccineand the hyperimmune globulins (20 units /Kg)

that contain a high titer of anti-rabies virus antibodies. They are inoculated at different sites so the antibodies do not neutralize the virus in the vaccine. This is an important example of passive- active immunization

- There is no vaccine for HSV-1, HSV-2, and WNV
- To reduce the transmission of HSV-2 to neonates, pregnant women with active lesions late in pregnancy should receive acyclovir and should be considered for cesarean section

BRAIN ABSCESS

01:08:17

- A brain abscess is a localized, walled-off collection of pus surrounded by a fibrous capsule

Etiology

- MCC: bacteria
- Fungi and protozoa also involved
- Viruses do not cause brain abscess

Pathophysiology

- Brain abscess is a recognized Complication of head and neck pyogenic infections, such as sinusitis, otitis media, and dental infections
  - Sinusitis predisposes to lesions in the frontal lobe
  - Otitis media predisposes to lesions in the temporal lobe
- Hematogenous spread from an infected site (infective endocarditis) can also occur
- Increased use of immunosuppressive drugs, IV catheters, and hyperalimentation are a/w fungal brain abscesses
- Immunocompromised patients, especially those with AIDS, also have brain abscesses caused by Toxoplasma gondii

Clinical Manifestations

- ↑ICT: Headache, vomiting (in early course of disease)
- Focal neurologic deficits and seizures (as the lesion expands)

Pathogens

- Bacteria
  - Streptococci: S. anginosus and Viridans group
  - Non-sporing anaerobes
    - → Prevotella
    - → Fusobacterium
    - → Bacteroides
- Fungi
  - A. fumigatus: in Neutropenic patients
  - Mucor and Rhizopus species (Rhinocerebralmu cormycosis): In diabetic patients with ketoacidosis
  - Cryptococcus: in patients with HIV/AIDS
  - Candida
- Protozoa
  - T. gondii is the main protozoal cause of brain abscess
  - A/w immunocompromised patients or with those receiving immunosuppressive drugs
  - MOT
    - → Solid organ transplant (especially heart transplants)
    - → Ingestion of raw meat containing cysts
    - → Exposure to cat feces containing oocyte
    - → Transplacental transmission: intracranial calcifications in fetus

Diagnosis

- MRI: ring-enhancing lesion
- Culture of pus from abscess for bacteria and fungi
- Gram staining reveals several types of bacteria indicating a mixed infection

Brain abscess

- MRI showing a small ring-enhancing lesion with mild surrounding edema adjacent to the ventricular catheter and ventricular dilatation

Treatment

- Empiric antimicrobial therapy for bacteria
  - Ceftriaxone or cefotaxime + metronidazole
- Treatment of bacterial and fungal brain abscess may require a surgical drainage of pus
- Treatment of Toxoplasma brain abscess includes a combination of pyrimethamine and sulfadiazine

SUBDURAL AND EPIDURAL EMPYEMA

- Subdural empyema is a collection of pus on the inner surface of the dura matter
- Whereas epidural empyema is a collection of pus on the

outer- surface

- They can occur adjacent to dura of either the brain or spinal cord
- Sinusitis and otitis media are common predisposing factors, and the bacteria causing these empyemas are those that cause sinusitis and otitis media
- Mixed infections are common

Clinical manifestations

- Clinical features include fever plus symptoms of increased intracranial pressure such as headache, vomiting, focal neurologic deficits, and altered mental status
- MRI with gadolinium [illegible]eals a mass adjacent to the dura[illegible]agnosis involves aspirating pus from the lesion and performing a Gram stain and culture

Treatment

- Involves surgical drainage of the pus combined with antibiotics appropriate for bacteria isolated from the aspirated pus

Diagram 28.1

CNS infections

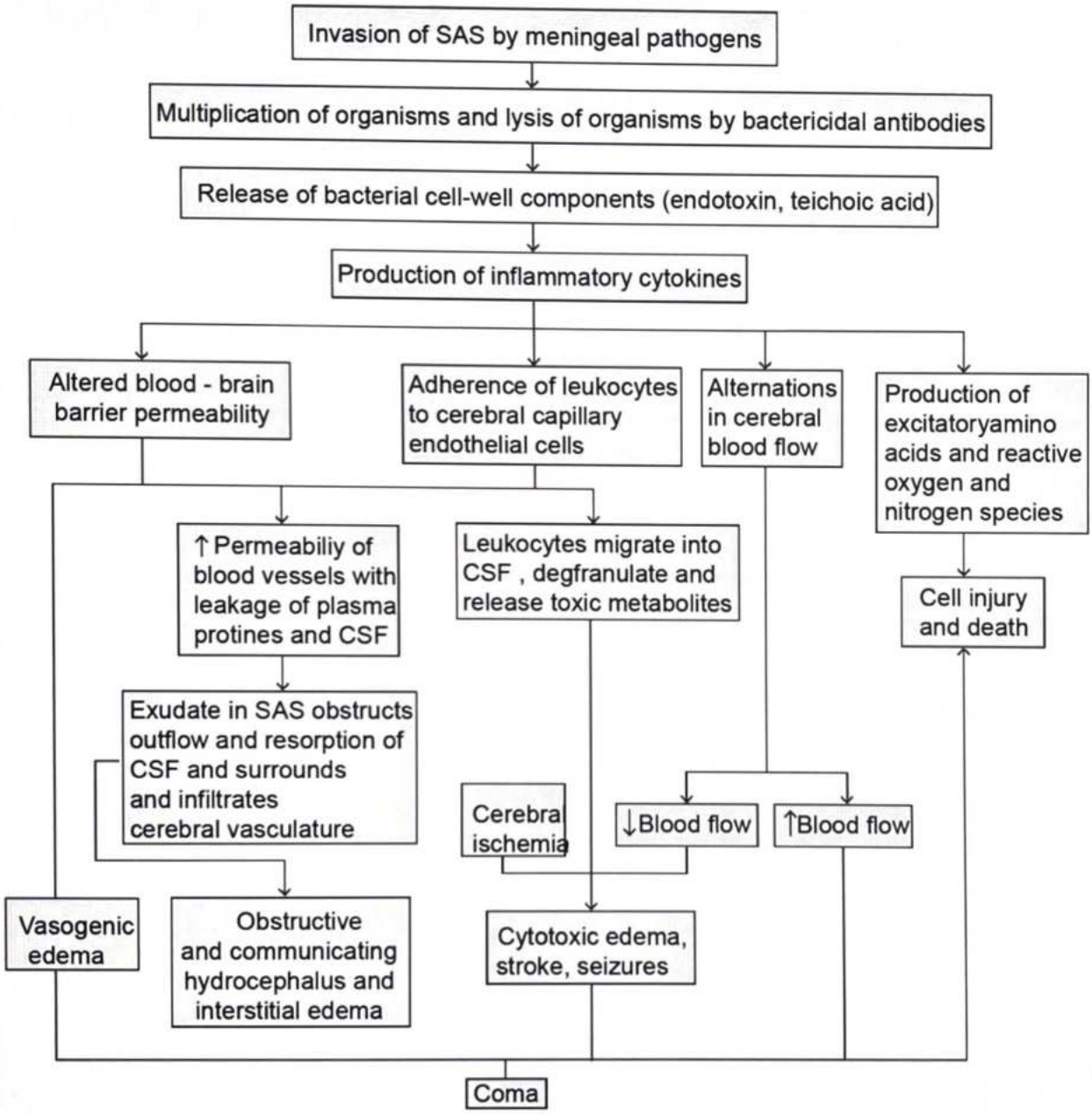


Table 28.1

Agents of meningitis	Biochemical analysis	Direct demonstration	Culture identification
S. pneumoniae	**Pyogenic meningitis** • CSF pressure: highly elevated >180 mm of water • TLC: highly elevated, neutrophilic (100-10,000/mm^3) • Glucose: ↓ed to absent <40mg/dL • Total proteins: >45mg/dL	• Gram +ve cocci in pair, lanceolate shaped	• Alpha hemolytic draughtsman-shaped colony on BA • Sensitive to optochin • Bile soluble • Ferments insulin
S. agalactiae		• Beta hemolytic pin point colony on BA • CAMP test +ve, resistant to bacitracin	
Neisseria meningitis		• Oxidase +ve, catalase +ve growth on chocolate agar	
H. influenzae		• Pleomorphic gram –ve bacilli	• Satellitism on BA with S. aureus streak line, growth surrounding disk containing combined X and V factors
E. coli or other Gram –ve bacilli		• Identification is based on colony grown on MacConkey agar and biochemical reactions	
Cryptococcus neoformans True yeast	• CSF pressure: slightly elevated/ normal • Leukocyte count: slightly elevated & lymphocytic • Glucose: normal • Total proteins: normal or slightly elevated	• India ink stain shows budding yeast cells • Latex agglutination test detects capsular Ag in CSF	• BA or CA or SDA shows mucoid ..... colonies
Viral meningitis		• Detection of DNA or RNA in CSF by PCR or RT PCR	
Tubercular meningitis	• CSF pressure: moderately elevated, cobweb coagulum seen • Leukocyte count: moderately elevated & lymphocytic • Glucose: slightly ↓ed • Total proteins: moderate to markedly ↑ed	• ZN stain of CSF shows acid fast bacilli • Detection of specific genes in CSF by PCR or GeneXpert	• Growth on LJ medium → rough, buff, tough colonies • Growth on MGIT

29 BLOOD STREAM INFECTIONS

Introduction

00:00:15

- Bacteremia: Presence of bacteria in blood without any multiplication
- Septicemia: Bacteria circulating & actively multiplying in blood stream & produce toxins
- Transient Bacteremia: Occurs spontaneously or after some event like brushing teeth, chewing food, instrumentation
- Continuous Bacteremia
  - Bacteria are in blood stream at a constant rate
  - Seen in Septic shock, Endocarditis
- Intermittent Bacteremia
  - Bacteria released in blood intermittently
  - Eg. In an undrained abscess, bacteria are released approximately 45 minutes before a febrile episode
  - Also seen in Meningitis, Pneumonia, Osteomyelitis

Etiological Agents of BSI

00:07:46

- Etiological agents include bacteria, fungi, viruses, parasites
- Bacterial agents include
  - Salmonella typhi
  - Brucella
  - Spirochetes
    - → Leptospira
    - → Borrelia
  - HACEK (Endocarditis group)
  - Rickettsiae
    - → Infect vascular endothelium
    - → Doesn't grow in blood culture
    - → Cultured in cell lines
  - GPC
    - → Staphylococcus
    - → β Hemolytic Streptococci
    - → Enterococcus
    - → Pneumococcus
  - GNC
    - → Meningococci
  - GPB
    - → B. anthracis
    - → Listeria
  - GNB
    - → E. coli
    - → Klebsiella
    - → Enterobacter
    - → Pseudomonas
    - → Acinetobacter
    - → H. influenzae
  - Anaerobes
    - → Bacteroides
- Viral agents include
  - Retro viruses
  - Hemorrhagic fever viruses
    - → Marburg
    - → Ebola
    - → Dengue
    - → Chikungunya
    - → Yellow fever
  - Herpes family
    - → EBV
    - → CMV
- Parasites causing BSI include
  - Plasmodium
  - Babesia microti
  - T. gondii
  - Leishmania
  - Trypanosoma
  - Filarial parasites
- Fungal agents
  - Candida albicans & Non Candida albicans (NCA)
  - Dimorphic fungus
    - → Histoplasma capsulatum
    - → Blastomyces
    - → Coccidoides
    - → Paracoccidiodes

Types of BSI

00:15:10

- Types include
  - Intravascular
  - Extravascular

- Pre disposing factors for BSI
  - Immunosuppression
  - Use of Broad spectrum antibiotics for a long time
  - Invasive procedures (like Central line)
  - Extensive surgeries
- Intravascular BSI
  - Infection of blood vessels
  - Includes
    - → Endocarditis
    - → Myocarditis
    - → Pericarditis
- Extravascular BSI
  - Organisms multiply at primary site such as lung (in case of Pneumonia)

↓

Drained by lymphatics

↓

Reaches blood stream

↓

Move to various organs

  - Organisms ultimately reach Reticuloendothelial system (RES)

↓

Escape various mechanisms of RES & multiply

↓

Cause Septicemia

  - Portal of entry
    - → Genito Urinary Tract (GUT) (25%) → MC
    - → Respiratory tract (20%)

Clinical Manifestations of BSI 00:20:57

- BSI have Bacteremia stage f/b Septicaemia stage
- Clinical manifestations evident only in Septicemic stage
- Stages of BSI based on severity & extent of organ failure
  - Sepsis
  - Septic shock

Definition of sepsis and the assessment of severity and organ failure 00:21:56

Sepsis

- Defined as life threatening organ dysfunction caused by a dysregulated host response to infection

SOFA score

- Sepsis is diagnosed by SOFA (Sepsis related Organ Failure Assessment) Score
- It depends on six parameters

1. Respiratory system: PaO_2, FiO_2
2. Coagulation system: Platelet count
3. Liver: Serum bilirubin
4. Cardiovascular system: Mean Arterial Pressure (MAP)
5. CNS: Glasgow Coma Scale (GCS)
6. Renal system: Serum creatinine & Urine output

- Organ damage can be identified as an acute change in the total SOFA score ≥2 points following the infection

qSOFA (Quick SOFA) Score

- Determination of SOFA score takes considerable time as it depends upon a number of laboratory parameters. However, before the result of SOFA score is available, sepsis can promptly be identified at bedside with qSOFA score
  1. Respiratory rate ≥22/min
  2. Altered mental status
  3. Systolic BP ≤100 mmHg

Septic shock

- Defined as subset of Sepsis in which underlying circulatory & cellular / metabolic abnormalities are profound
- Patient with Sepsis shock are identified by clinical construct of Sepsis with
  - Persisting Hypotension requiring Vasopressors to maintain MAP (Mean Arterial Pressure) ≥65 mmHg
  - Serum Lactate level > 2 mmol/L (18mg/dL) despite adequate volume resuscitation
- Patients with Septic shock have a mortality of >40% in contrast to 10% for Sepsis cases

Lab Diagnosis 00:28:01

Specimen Collection for Blood culture

- Site: Blood for Culture should always be collected in pairs; from 2 separate Venipuncture& 2 separate skin decontamination processes
- Preparation of the site: To avoid contamination with skin flora, blood should be collected under strict aseptic conditions using sterile disposable syringe
- Skin Decontamination

- Timing of collection: Blood collected before starting Antimicrobial therapy
- Blood volume
  - Adult: 8-10 mL of blood per bottle
  - Pediatric: 1-3 mL of blood per bottle
  - Ratio of Blood : Blood culture fluid - 1:10
- Number of blood cultures
  - Atleast 2-3 blood Culture sets are required
  - Each set consists of 2 bottles: 1 Aerobic & 1 Anaerobic
- Dispensing
  - Collected blood is directly dispensed into Blood Culture bottle at the bedside

Steps of Collection of Blood for Blood Culture

Refer Diagram 1.1

1. Perform hand hygiene & use sterile gloves
2. Apply tourniquet, palpate the vein & mark the area
3. Use 70% Alcohol to disinfect the site upside down. Wait for 30 sec (allow the skin to dry)
4. Use Chlorhexidine / Povidone iodine to disinfect site concentric inside out. Wait for 1 min (allow the skin to dry)
5. Alcohol wipe to clean the blood culture bottle top
6. Collect blood aseptically & inject into blood culture bottle without changing the needle

Conventional Culture Medium

- Types of Media
  - Monophasic Medium: 50 – 100 ml of brain heart infusion (BHI)broth
  - Castaneda's Biphasic Medium: BHI agar slope and BHI broth, BHI agar

Monophasic Medium

Castaneda's Biphasic Medium

- Dilution
  - Blood is inoculated in the medium at a dilution of 1:5 (1:10) so that the antibacterial components in the blood, if any, will get diluted
- Sodium polyanethol sulfonate (SPS)
  - SPS is added to the medium as an anticoagulant.
  - It also counteracts the bactericidal action of blood
- Incubation
  - Bottles should be directly incubated in upright position at 37°C for 7 days
  - Initial report comes in 3 days
- Repeat subcultures are made from the BHI broth onto Blood agar & Mac Conkey agar

Automated Culture Media

- BACTEC & Bact / ALERT
- Bact / ALERT Virtuo- Most advanced system
- Growth is continuously monitored and reading is recorded every 15-20 mins
- When growth is detected, the system gives a positive signal
- Then the bottle is removed and processed similarly as done for Conventional bottles

Bactec BacT / Alert

Identification

- The isolated organisms is identified by colony morphology, Gram staining, followed by either conventional biochemical reactions or Automated identification system such as MALDI-TOF or VITEK

Maldi-TOF

Vitek

Antimicrobial Susceptibility Test (AST)

- AST is carried out for the guiding institution of appropriate therapy
- MIC based method (Eg. VITEK) is preferred over disc diffusion
- It is ideal for Endocarditis isolates, especially while reporting susceptibility of Penicillin

Treatment

- D/t high prevalence of Multi Drug Resistant Organisms (MDRO) & higher mortality in Sepsis, Antibiotics are started at earliest as soon as Sepsis is clinically suspected

30 FEVER OF UNKNOWN ORIGIN

FUO
00:00:13

- Despite doing intensive investigations not able to find out the real cause

Criteria
00:00:46

- Fever: ≥38.3° C on atleast 2 occasions
- Duration of illness: ≥3 wks
- No know immunocompromised state
- Diagnosis: uncertain diagnosis after History taking, Physical examination & various investigations

Investigation

- ESR & CRP
- Blood count (TLC, DLC, Hb, Platelets)
- Electrolytes, total proteins
- Enzymes
- ANCA & RF test
- Urine analysis
- Blood Culture (3 –ve Blood Cultures)
- Chest X ray, CT scan & MRI

Etiology
00:07:20

- Infections (36%)
- Neoplasms (19%)
- Non inflammatory diseases →SLE, RA
- Granulomatous disease
- Undiagnosed cases (7%)

Systemic Bacterial Infections a/w FUO
00:09:15

- Mycobacterial infection
- Typhoid fever
- Rickettsial infection
- Melioidosis (B. pseudomallei)
- Listeriosis
- Bartonella
- Actinomycosis & Nocardiosis
- Syphilis
- Spirochete infection
  - Lyme's disease
  - Relapsing fever
- Leptospirosis

Viral Causes a/w FUO
00:13:52

- CMV & EBV
- Coxsackie virus (Group B infection)
- Viral hepatitis
- HIV infection
- Dengue
- Colorado tick fever

Parasitic Infectionsa/w FUO
00:15:25

- Malaria
- Amoebiasis
- Leishmaniasis
- Chagas disease
- Toxoplasmosis
- Echinococcus infection
- Schistosoma
- Toxocaracanis / catis

Fungal Infections a/w FUO
00:16:49

- Dimorphic fungus
- Aspergillosis
- Mucormycosis
- Candida & Cryptococcus
- GMS
  - PCP
  - PJP

31 ANEMIA CAUSING ORGANISMS

Iron Deficiency Anemia 00:00:23

- **T** - **T**richuris trichiura
- **S** - **S**chistosoma species
- **H** - **H**ookworm
  - Ancylostoma
  - NecatorAmericanus

How to remember

- T S H

Hemolytic Anemia 00:01:49

- **B** - **B**abesia
- **B** - **Bartonella** bacilliformis
- **C** - **C**lostridium perferingens
- **England** - EBV infection (Infectious mononucleosis)
- **Main** - Malaria
- **Hai** - **H**ep A

How to remember

- B B C England Main Hai

Megaloblastic Anemia 00:03:47

- Diphyllobothrium latum

Aplastic Anemia 00:04:18

- **A**nemia
- **P** - **P**arvo B_{19}
- **L** - **L**. donovani
- **A**
- **S**
- **T** - **T**B
- **I** - **I**nfectious mononucleosis (EBV)
- **C** - Hep **C**
- **Hai** - **H**HV_3, **H**HV_4, **H**HV_5, **H**HV_6 & **H**IV

How to remember

- A P L A S T I C Hai

32 EYE & EAR INFECTIONS

EYE INFECTIONS

00:00:13

External structures of Eye involved	Causative organisms
• Eyelid (Blepharitis)	• Staph aureus
• Conjunctiva (conjunctivitis)	• H. Influenzae • Pneumococcus • Chlamydia trachomatis • Gonococcus • Moraxella (Angular conjunctivitis) • Adenovirus • HSV
• Cornea (Keratitis)	• S. aureus • Pneumococcus • Fusarium • Candida • Acanthmoeba
• Sclera (Scleritis)	• S. aureus

Internal structures of eye involved

- Retina (Retinitis)
- Uvea (Uveitis)
- Aqueous humor or Vitreous Humor (Endopthalmitis)

Causative organisms

- Chorioretinitis & Uveitis
  - MTB
  - T. pallidum
  - Borrelia burgdorferi
  - CMV
  - Toxoplasma gondii
- Endopthalmitis
  - S. aureus
  - Pnemococcus
  - Pseudomonas aeruginosa
  - Other Gram Negative Bacteria (GNB)
  - HSV
  - Candida

EAR INFECTIONS

00:09:03

Otitis Externa / Swimmer's Ear

1. Acute Otitis externa

- Causative organisms
  - S. aureus (MC)
  - S. pyogenes
  - Pseudomonas
    → Common in DM
    → Causes Malignant Otitis externa
  - Aspergillus
  - Candida

2. Chronic Otitis externa

- Causative organisms
  - Anaerobes (MC)
  - Pseudomonas

Otitis Media

Acute Otitis media	Chronic Otitis media

• Causative organisms
  ○ Pneumococcus (33%) (MC)
  ○ H. influenzae type B
  ○ Moraxella catarrhalis
  ○ S. Pyogenes
  ○ RSV
	• Causative organisms
	  ○ Anaerobes (MC)

33 SKIN LESIONS

Primary Skin Lesions 00:00:20

1. Macule
- Flat, non palpable discoloration of skin (<5mm in size)
- Size >5mm: Patch
- Caused by
  - Dermatophytes
  - T. pallidum (secondary syphilis)
  - Viral rashes (Enterovirus)

2. Papule
- Elevated, palpable, sold lesion
- Usually <5mm in size
- Seen in
  - Molluscum contagiosum
  - Scabies
  - Warts (HPV)

3. Nodule
- Elevated, palpable, solid lesion
- Usually >5mm in size
- Caused by
  - C. diptheriae
  - Sporothrix schenkii
  - PKDL
  - M. marinum
  - Nocardia
- Size >2cm: Plaque

4. Vesicles
- Fluid filled blister ≤5mm size
- Caused by
  - HSV
  - VZV
  - M. contagiosum
  - Hand, Foot & Mouth disease (Coxsackie A_{16})

5. Bulla
- Fluid filled blister >5mm size
- Caused by
  - C. perfringens (Gas Gangrene)
  - S. aureus (Bullous Impetigo)
  - Vibrio Vulnificus (Halophillic bacteria)

6. Pustule
- Pus filled lesion ≤5mm size
- Caused by
  - Candida
  - S. aureus (Folliculitis)
  - S. pyogenes (Impetigo)
  - Dermatophyte

7. Abscess
- Pus filled lesion >5mm size
- Caused by
  - S. aureus
  - Strep. Pyogenes

8. Petechiae
- Blood extravasated into skin
- Size <2mm
- Caused by
  - HIV
  - Hep C
  - EBV
  - Rubella
  - CMV
  - Rocky mountain spotty fever (RMSF)

9. Purpura
- Blood extravasated into skin
- Size: >2mm to 1cm
- Seen in
  - CMV
  - Endocarditis
  - Meningococcemia
  - Mononucleosis like syndrome
  - RMSF
  - Scarlet fever
  - Sepsis
  - Viral hemorrhagic fever

10. Ecchymosis
- Blood extravasated into skin
- Size >1cm
- Caused by
  - Ebola virus
  - Marburg virus

- Both ebola and Marburg causes viral hemorrhagic fevers

Other important skin lesions

1. Scale
- Excess dead epidermal layer
- Caused by Dermatophytes

2. Ulcer
- Break in epithelial lining extending upto epidermis / dermis
- Caused by
  - Bacillus anthracis
  - M. leprae (Decubitus ulcers of leprosy)

3. Erysipelas
- Painful, red, indurated swollen lesion involving the dermis
- A/w fever & lymphadenopathy
- Caused by: S. pyogenes

4. Cellulitis
- Diffuse spreading infection involving deeper layers of dermis
- Caused by
  - S. pyogenes
  - S. aureus

5. Hidradenitis
- Chronic infection of obstructed sweat glands
- Caused by
  - S. aureus
  - Streptococcus anginosus

Hair Follicle Infections 00:19:59

1. Folliculitis
- Superficial infection of single hair follicle
- Mostly presents like Pustule
- Caused by: S. aureus

2. Furuncle
- Deeper infection of hair follicles
- Presents as Abscess
- Spreads deeply into the dermis & subcutaneous tissue
- Caused by: S. aureus

3. Carbuncle
- Coalescence of various furuncles
- Caused by: S. aureus

Infection of Fascia & Muscles 00:24:06

1. Necrotizing fascitis
- Rapidly spreading infection
- Caused by: S. pyogenes (Flesh eating bacteria)

2. Pyomyositis
- Pus formation in muscle layer
- Caused by
  - S. aureus
  - S. pyogenes

3. Myonecrosis
- Extensive necrosis of muscle layer with gangrene formation (Clostridial myonecrosis)
- Caused by
  - C. perferingens
  - C. novyi
  - C. septicum
  - Other anaerobes

Agent Causing Burn Wound Infections 00:27:31

1. S. aureus
2. P. aeruginosa
3. CONS (S. epidermidis)
4. Candida albicans

Agents Causing Surgical Site Infections

1. Clean wounds
- S. aureus
- CONS
- Enterococcus
- Candida albicans

2. If Bowel integrity is compromised
- Gram negative bacteria: E.coli
- Anaerobic infections: Bacteroides, Prevotella

Lab Diagnosis 00:32:21
- Appropriate sample is take
  - Pus taken up from wound by: Sterile swabs
  - Pus taken up from abscess by: Incision & drainage
  - Vesicles & Bulla fluid by: Needle aspiration
  - Subcutaneous infections: Sample from base of lesion
  - Dermatophytic infections
    - → Skin scrapings
    - → Nail clippings
    - → Plucked hairs

- Microscopy
  1. Gram staining: Gives initial idea about Morphology of organism
  2. 10% KOH mount for most of fungal infections
  3. Tzanck smear of vesicle fluid for HSV_1, HSV_2, HHV_3

- Culture
  - Aerobic organisms
    - → Inoculate samples on to
      - Blood Agar
      - Mac Conkey agar
    - → Incubate overnight at 37°C
  - Atypical Mycobacteria: LJ medium
  - Dermatophytes
    - → DIM
    - → SDA

- Anaerobic organisms: Bacteria incubated anaerobically on
  - → RCM (Robertson Cooked Meat Media)
  - → BHI agar (Brain Heart Infusion Agar)

- Culture growth: go for AST (Antibiotic Sensitivity Testing)
  - System used for identifying organisms along with AST patterns → Vitek
  - Vitek also tells about MIC

Vitek

34 RANDOM SKIN LESIONS

Tinea infection

Tinea infection

- Types of tinea include ring worm, athletes foot and Jock itch

Impetigo

- Honey colured crust Typical of impentigo done by Streptococcus pyogenes and staph aureus

Psoriasis plaque

Psoriasis Plaque

- Plaques are lesions >2cm

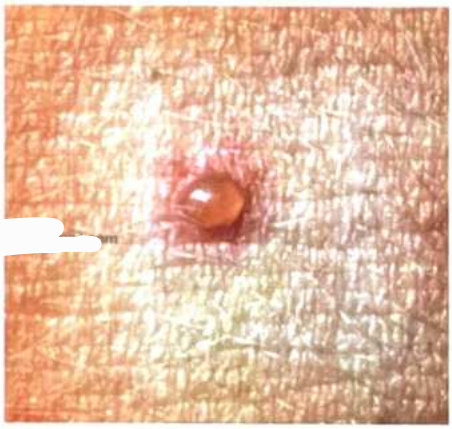
Molluscum contagiosum

Pearly white Umblicated nodule

- Seen in molluscum contagiosum
- Organism: MCV1 and 2
- MCV 2 mostly relates with HIV +ve

Chicken pox

Chicken Pox



- Dew drop on rose petal appearance
- Pleomorphic lesion

Herpes labialis

Herpes labialis

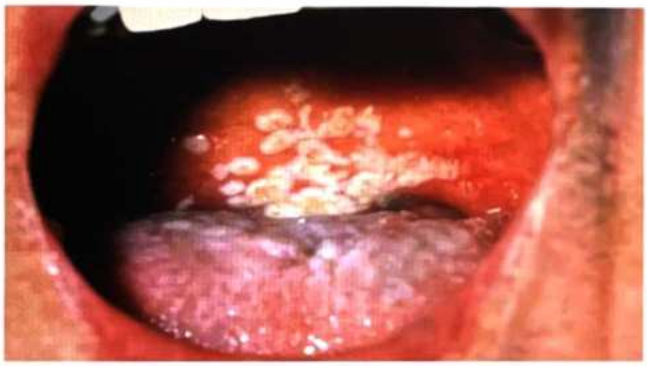
- Small painfull blister

Measles

Measles

- Small red spots over faces
- Rash goes down the arms and trunk and later to the thighs, lower leg and feet
- Infectivity period: 4 days before and 5 days after rash

Candidiasis



- Typical of oral candidiasis
- Treated using Azoles

Oral hairy Leukoplakia

Oral Hairy Leukoplakia

- Whitish patches on tongue typical of oral hairy leukoplakia
- Organism: EBV

Congenital rubella

- Congenital rubella
- Skin manifestations include
  - Skin had multiple well defined violaccous, non-blanchable macules
  - Barely palpable plaques to nodules ranging is size from 0.3 x 0.3cm^2 to 0.5 x 0.5 cm^2, present all over the body including palms and soles predominantly face

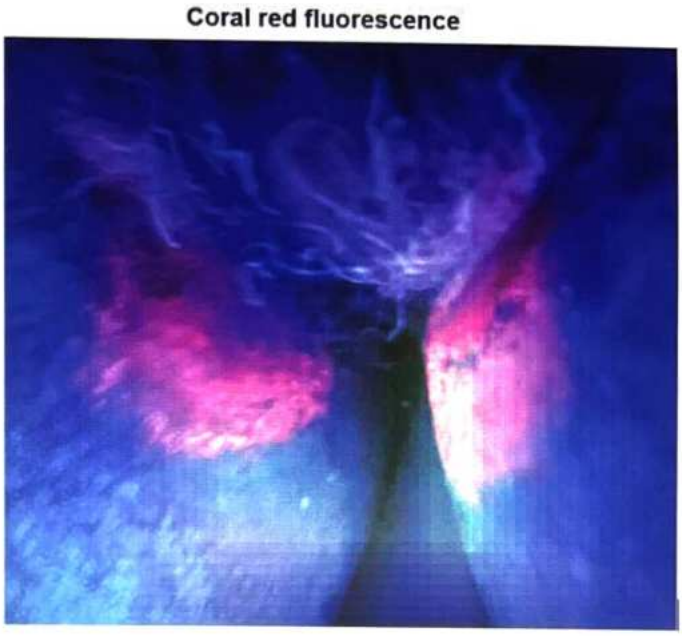
Warts

- Verruca vulgaris and verruca plana (HPV-3)
- Common warts are mostly done by HPV 2 and 4
- Butchers wart is done by HPV 7
- Laryngeal warts and Anogenital warts done by HPV 6 and 11

Morbilliform rash

- Morbilliform rash is defined as a Rose-red flat (macular) or slightly elevated (maculopapular) eruption, showing circular or elliptical lesions
- Varying in diameter from 1 to 3 mm, with healthy-looking skin intervening
- Observed in
  - Measles
  - Rubella

Coral red fluorescence



- Observed in Erythrasma
- Organism: Corynebacterium Minutissimum

Papule

- Images shows raised lesions ≤5mm

Kerion

- Organism: Trichophyton verrucosum and mentagrophytes
- Painful Boggy swelling

Jock itch

- Organism: Trichophyton , Epidermophyton

Herpes Zoster

- Blisters on torso is a common presentation
- Mainly painful

35 NOSOCOMIAL INFECTIONS

- Definition: Infection occurring >2days of Hospital admission

Classification 00:00:34

- CAUTI: Catheter associated UTI
- CRBSI: Catheter related blood stream infection
- VAP: Ventilator associated Pneumonia
- SSI: Surgical Site Infections

CAUTI 00:02:17

- MC HAI worldwide
- Short term catheterization
  - Causative organisms
    - → E.Coli (MC)
    - → Klebsiella
    - → Pseudomonas
    - → Enterococci (GPC)
- Long term catheterization
  - Usually Polymicrobial infection occurs
  - Causative organisms
    - → Pathogens of short term catheterization + PMP
    - → **P** - Proteus
    - → **M** - Morganella
    - → **P** - Providencia

How to remember

- PMP

Source of infection

Extra luminal spread	Intra luminal spread
• Mostly endogenous flora or Soiled hands of Healthcare workers	• Open drainage bag (allows organisms to enter & cause infection)

Lab diagnosis

Catheter criteria	Catheterized or history of recent Catheterization within 48hrs
Clinical criteria	Presence of at least one sign or symptom of UTI • Fever • Suprapubic tenderness • Costovertebral angle pain • Urinary urgency, frequency or dysuria
Urine culture criteria	Presence of significant bacteriuria; defined as Colony count exceeding • $\geq 10^3$CFU/mL: in symptomatic patients • $\geq 10^5$CFU/mL: in asymptomatic patients

CRBSI 00:07:54

- A/w Central venous catheter or Central line
  - Temporary or short term central line (<72 hrs)
  - Permanent or longterm central line (>72 hrs)
- Site of insertion of Central line
  - Subclavian vein
  - Femoral vein
  - Internal jugular vein
  - Peripheral veins
- Organisms gain entry
  - D/t colonization of organisms on Central line
  - Eg. CONS (Biofilm), S. aureus
- Clinical criteria for CRBSI
  - Presence of fever, chills, rigor, hypotension after central line is used
  - Signs of catheter site infection present
    - → Erythema, Tenderness, Swelling at the catheter exit site
- Microbiological criteria for CRBSI
  - Take blood culture from both central line as well as peripheral line
  - If central line blood culture bottle flags ≥2 hrs prior to peripheral line blood culture bottle: Indicative of CRBSI

VAP

00:14:25

- 2nd MC Nosocomial infection after CAUTI
- Accounts for 15-20% of total HAI's
- MCC of death among HAI (Mortality rate →40%)

Early onset VAP

- Occurs in first 4 days of Mechanical ventilation
- Caused by
  - Pneumococcus
  - H. Influenzae
  - MSSA (Methicillin Sensitive Staph aureus)

Late onset VAP

- Develops ≥5 days after Mechanical ventilation starts
- Caused by
  - P. aeruginosa
  - Acinetobacter baumanii
  - E. coli
  - Klebsiella
  - MRSA (Methicillin Resistant Staph aureus)

Source of infection

- Endogenous flora
- Exogenous infection: from Heathcare workers, Hospital environment

Clinical criteria for VAP

- CPIS score >6: Diagnostic of VAP
- CPIS: Clinical Pulmonary Infection Score

Microbiological criteria for VAP

- Specimens taken
  - Endotracheal aspirate (ETA): MC
  - BAL (Broncho Alveolar Lavage) fluid
  - Lung biopsy (by Bronchoscopy)
- Gram staining
- Culture
  - Quantitative assessment on Culture
    - → ≥10^5CFU/ml for ETA
    - → ≥10^4 for BAL
  - Semi-quantitative
    - → Moderate to heavy growth

Radiological investigations

- CT Scan
- CXR

SSI

00:21:30

- Infections which develop within 30 days of surgery at surgical site
- Exception: ≥90 days of Surgery
  - Breast Sx
  - Cardiac Sx
  - Joint Sx

Source of infection

- Endogenous flora
- Exogenous infection: from Heathcare workers, Hospital environment

Wound class types

- Class I wounds: Clean wounds
- Class II wounds: Clean contaminated wounds
- Class III wounds
  - Contaminated wounds: Mostly a/w accidents
  - Dirty & infected wounds: Surgical procedure done overactive infection

Other classification of SSI

Superficial SSI	Deep SSI

• Occurs at level of superficial incisional site
• Skin & Subcutaneous tissue involved
• Happens within 30 days of surgery
	• Occurs at level of deep incisional site (Muscle & Fascia)
	• Happens within 30 days of surgery
	• Exception: ≥90 days of Surgery
	  ○ Breast Sx
	  ○ Cardiac Sx
	  ○ Joint implant Sx

Prevention of SSI

- Pre-operative measures
  - Pre operative bathing
  - Treat MRSA carriers with Mupirocin ointment
  - Hair removal should not be done. Hair clipping is done now a days
- Intra operative measures
  - SAP (Systemic Antibiotic Prophylaxis)
  - Surgical hand disinfection by best possible ways
  - Proper surgical site preparation
  - Proper Oxygenation (Target FiO_2: 80%)
  - Maintain normal blood glucose levels during Sx(Target <200mg/dL)
- Post-operative measures
  - Proper wound dressing: Done daily
  - OT disinfection
  - Periodic monitoring of air quality in OT

36 HAND HYGIENE

- Best method to prevent Nosocomial infections

Hand Rub 00:00:41

- Alcohol based Rubs (70-80% Ethyl alcohol)
- Chlorhexidine
- Duration:20-30 seconds

Hand Wash 00:01:30

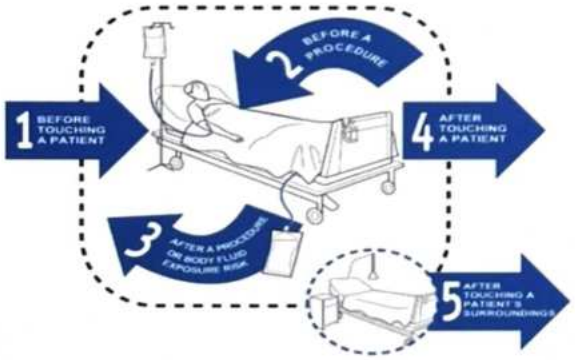
- Done with Antimicrobial soaps
- Duration: 40-60 seconds
- When to do hand wash
  - When hands are visibly soiled with blood, pus or excreta
  - Before & after eating
  - After going to toilet
  - Before and after duty (in case of Healthcare personnel)

Surgical hands scrub 00:03:37

- Done with 4% Chlorhexidine
- Duration: 3-5mins
- Done prior to any surgical procedure

Five moments for Hand Hygiene (as per WHO) 00:04:25

- Before touching a patient
- Before a procedure
- After a procedure or body fluid exposure risk
- After touching a patient
- After touching a patients surroundings



Steps of Hand Rubbing & Hand Washing 00:05:37

- Step 1 - Apply Hand Rub / Hand Wash
- Step 2 - Palm to Palm
- Step 3 - Back of the palm
- Step 4 - Interlacing of fingers
- Step 5 - Back of the fingers
- Step 6 - Rotational rubbing of thumb
- Step 7 - Scrub nails on palm
- Step 8 - Rinse the hands with water
- Step 9 - Dry hands with a single use paper towel
- Step 10- Close the tap with (Elbow >same single use paper towel)

Personal Protective Equipments (PPE) 00:09:04

1. Gloves
2. Heavy duty gloves
3. Surgical mask
4. N_{95} respirator
5. Plastic apron
6. Linen gown
7. Disposable gown
8. Coverall
9. Goggles
10. Face shield
11. Cap
12. Shoes
13. Gum boot
14. Shoe cover

Donning (Wearing PPE) 00:11:50

- Steps of Donning
  1. Gown
  2. Mask / N_{95} respirator
  3. Goggles / face shield
  4. Gloves

Doffing (Removing PPE)

- Steps of Doffing
  1. Gloves
  2. Goggles / Face shield
  3. Gown
  4. Mask / N_{95} respirator

Blood Spill Management 00:16:08

- Blood spill management is needed to remove the risk of transmission of blood borne viruses
- Steps to follow if a blood spill happens
  - Contain the spill and put a Signage at the place where spill happened
  - Call Infection Control Team
  - Sodium hypochlorite used in management of blood spill should be freshly prepared
- As per CDC guidelines
  - For large spills: ≥10cm size
    → Use Sodium hypochlorite: 0.5% (1%)
  - For small spills: <10cm size
    → Use Sodium hypochlorite: 0.05%
  - Contact time: 10 min
  - Rinse the area with clean water to remove the disinfectant residues

37 ORGANISMS TRANSMITTED BY DROPLETS AND AEROSOL

Droplet precautions 00:00:13

- Respiratory droplet are
  - Large particles >5μ in size
  - Generated by a person while coughing, sneezing or talking
  - Transmission doesn't happen beyond 3 feet

- Infections spread by droplets 00:01:32
  - **MDH** - **M**eningococci, **D**iptheria, **H**ib
  - **in**
  - **PPM** - **P**ertussis, **P**neumonic Plague, **M**ycoplasma pneumonia
  - **IS** - **I**nfluenza virus, **S**ARS COV2 (COVID 19)
  - **Very** - **V**iral Hemorrhagic fever (Ebola, Marburg, Lassa virus)
  - **Owesome** - **O**ther viruses
    - → **A** - **A**deno virus
    - → **R** - **R**hino virus, **R**ubella
    - → **M** - **M**umps
    - → **P** - **P**arvo virus B_{19}

How to remember

- MDH in PPM IS Very Owesome ARMP

- Aerosols are Particles <5μ in size
- Infections spread by Aerosols
  - **A** - **A**spergillus
  - **B** - **B**acillus anthracis
  - **C** - **C**hicken pox
  - **M** - **M**TB
  - **M** - **M**easles
  - **S** - **S**mall pox

How to remember

- A B C M M S

38 BIOSAFETY LEVELS

- Level of biocontainment precautions required to isolate dangerous biological agents in enclosed facility

BSL – 1 00:01:26

- Biological agents pose low risk to personnel & environment
  - **Big** - **B**acillus thuringiensis
  - **P** - **P**seudomonas fluorescens
  - **E** - **E**.coli K_{12}
  - **A** - **A**spergillus niger
  - **S** - **S**erratia marcescens

How to remember

- BIG PEAS

- BSL-1 is found in
  - Water testing labs
  - High Schools & College teaching introductory Microbiology classes
- Work is done on an open bench
- Lab coats & gloves are recommended

BSL-2 00:04:20

- Biological agents pose moderate risk to personnel & environment
- Risk of spread is limited & it rarely would cause infection that would lead to serious disease
- Eg. For BSL-2
  - Bacteria: Pathogenic Staph, Streptococci, Pneumococci, Clostridium species, Bacillus (except Anthrax), Enterobacteriaceae, Pathogenic E.coli
  - Viruses: Herpes, Calci, Influenza, Adeno (HCC Is Aggressive)
  - Fungi: Aspergillus, Dermatophytes, Candida, Cryptococcus (ADCC)
- Diagnostic work of COVID 19
  - PCR / Tru NAT: Done in BSL-2

BSL-3 00:07:32

- Biological agents usually produce serious disease
- Lab must be in a separate building or isolated zone with double door entry, directional inward flow.
- Culture of SARS COV 2 (COVID 19) (for vaccine production) is done in BSL-3
- Eg. of BSL -3
  - Bacteria
    - → **A** - **A**nthrax
    - → **B** - **B**rucella
    - → **C** - **C**oxiella burnetti
    - → **T** - **T**ularemia
    - → **Y** - **Y**ersinia pestis
    - → **M** - MTB

How to remember

- A B C TYM

  - Viruses (Arboviruses)
    - → Hantan virus
    - → Lymphocytic choriomeningitis virus
    - → St. Louis encephalitis virus
    - → Japanese encephalitis virus
    - → West nile encephalitis virus
    - → Chikungunya
    - → Yellow fever virus

  - Fungi (Dimorphic fungi)
    - → Coccidiodes immitis
    - → Blastomyces dermatidis
    - → Paracoccidiodes brasilensis
    - → Histoplasma capsulatum

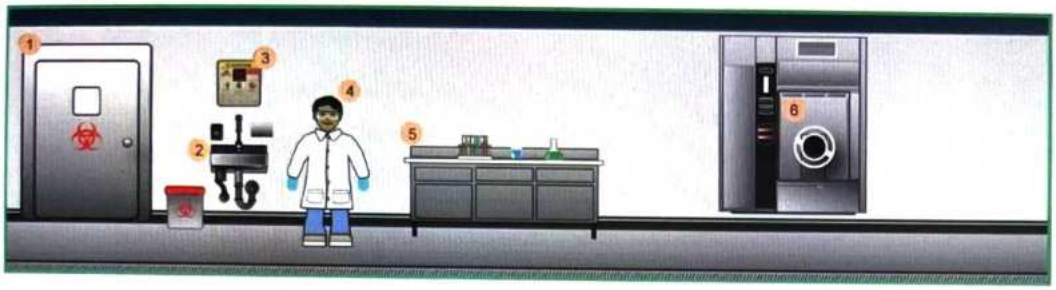
BSL-4 00:12:51

- Biological agents pose very serious infection that is often untreatable
- Easily transmitted from one to another individual, from animal to human and vice versa either by direct, indirect or by casual contact

- Eg.
  - Lassa fever virus
  - Marburg virus
  - Ebola virus
  - CCHF (Crimean Congo Hemorrhagic Fever) virus
  - Hendra virus
  - Nipah virus

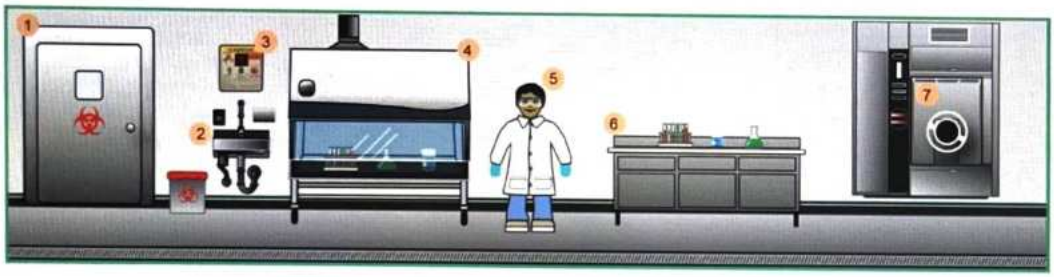
4 BIOSAFETY LAB LEVELS

BSL 1



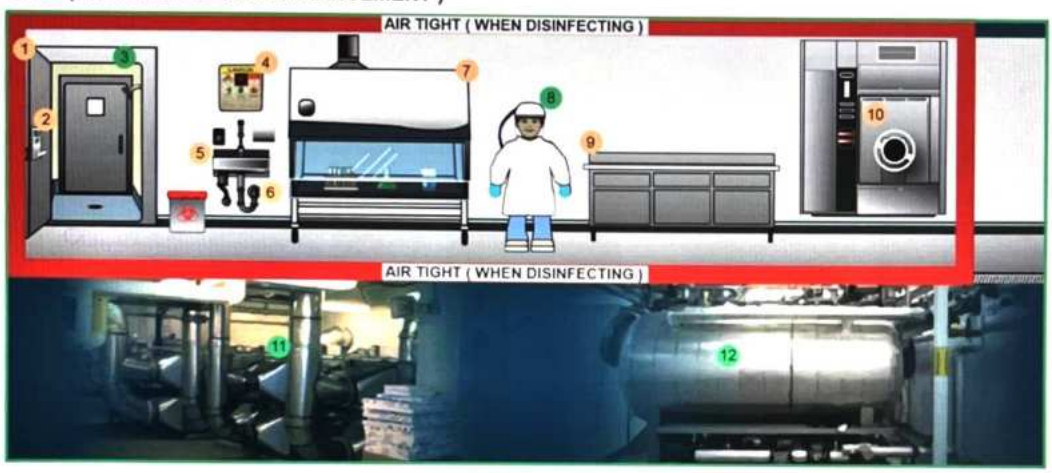
BSL 1
1 Controlled access
2 Hand washing sink
3 Sharp hazards warning policy
4 Personal protective equipment
5 Laboratory bench
6 Autoclave

BSL 2



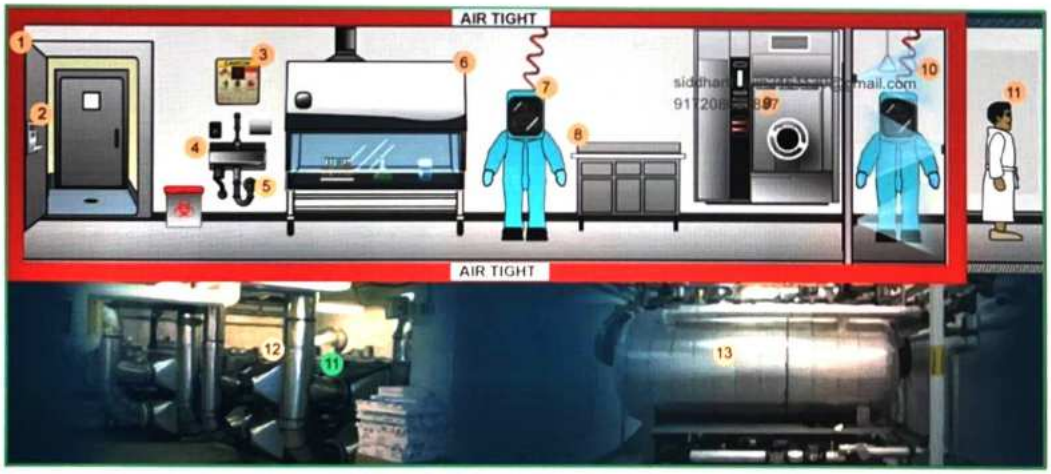
BSL 2
1 Controlled access
2 Hand washing sink
3 Sharp hazards warning policy
4 Physical protective equipment
5 Personal protective equipment
6 Laboratory bench
7 Autoclave

BSL 3 (WITH RISK-BASED ENHANCEMENT)



BSL 3
1 Self closing, double-door access
2 Controlled acsess
3 Personal shower out
4 Sharp hazards warning policy
5 Hand washing sink
6 Sealed penetrations
7 Physical containment device
8 Powered air purifying respirator
9 Laboratory bench
10 Autoclave
11 Exhaust HEPA filter
12 Effluent decontamination system

BSL 4



Required safety equipment Risk-based enhancements

BSL 4
1 Self closing, double-door access
2 Controlled acsess
3 Sharp hazards warning policy
4 Hand washing sink
5 Sealed penetrations
6 Physical containment device
7 Positive pressure protective suit
8 Laboratory bench
9 Autoclave
10 Chemical shower out
11 Personal shower out
12 Supply and exhaust HEPA filters
13 Effluent decontamination system

- Level of biocontainment precautions required to isolate dangerous biological agents in enclosed facility

39 OCCUPATIONAL EXPOSURES (HIV & HBV)

1. Percutaneous injuries 00:00:20
- Needle stick injury / Sharp injury

2. Splash injury
- Contact with mucus membrane
- Contact with non intact skin
- Contact with intact skin

Agents involved in Needle Stick Injury 00:02:05

Agents	Risk of transmission
HBV	30%
HCV	3%
HIV	0.3%

Potential infectious body fluids 00:03:15
- Blood
- Genital secretions
- All body fluids: CSF, Synovial fluid, Pleural & Peritoneal fluid

Not considered potentially infectious
- Fecal matter
- Nasal secretions
- Saliva
- Sputum
- Sweat
- Tears
- Urine
- Vomitus

Precautions during needle handling 00:06:41
1. Work surface: Disinfect with 0.5% Na hypochlorite
2. Healthcare workers: Immunized against HBV
3. If spillage of blood / body fluids happen then Management with 1% Na hypochlorite
4. Disposable needle should be used (No Recapping)

Post exposure management 00:09:50
1. First aid
2. Report to Nodal center
3. First dose of Post Exposure Prophylaxis (PEP) within 2hrs (HIV) & at the most before 72hrs
4. Test for other blood borne viruses
5. Record the incident
6. Follow up testing of Healthcare worker
7. All precautions should be taken during follow up

First Aid 00:13:18
- For Splash injury
  - Irrigate thoroughly the site vigorously for atleast 5 mins
  - In oral contact: spit fluid immediately & rinse the mouth several times
  - If wearing contact lens
    - → Leave them in place
    - → Eye is cleaned first
    - → Remove contact lens in normal fashion

Don'ts 00:16:08
1. Don't panic
2. Never put pricked finger in mouth
3. Don't squeeze finger

PEP for HIV as per NACO guidelines 00:17:25

Exposure code
- EC-1
  - Mild Exposure
  - Mucous membrane contact / non intact skin contact with small volume of blood / fluid or less duration
- EC-2 (Moderate Exposure)
  - Mucous membrane contact / non intact skin contact with large volume of blood / fluid or more duration
  - Percutaneous superficial exposure with solid needle or just a superficial scratch
- EC-3 (Severe Exposure)
  - [illegible] exposure with
    1. Large volume transfer
    2. Deep puncture of needle
    3. Visible blood on device
    4. Needles used in blood withdrawing from arteries & veins

Source Codes (SC)
- SC-1
  - HIV positive but asymptomatic
  - Viral load <400 copies/ml
- SC-2
  - HIV positive, symptomatic
  - High viral load

- SC unknown
  - Status of patient is unknown
- HIV negative
  - Tested negative according to NACO strategies

Exposure Code	Source Code	PEP
1,2 or 3	Negative	Not needed
1	1	Not needed
1	2	Needed
2	1	Needed
2	2	Needed
3	1 or 2	Needed
2/3	Unknown	Needed

PEP
- Started within 2hrs
- It is of no use if started after 72hrs
- Duration: 28 days

PEP Regimen
- Tenofovir
- Lamivudine } 1 tablet daily x 28 days
- Lopinavir
- Ritonavir } 2 tablets twice daily x 28 days

Alternative Regimen: TLE
- Tenofovir: 300mg
- Lamivudine: 300mg } 1 tablet daily x 28 days
- Efavirenz: 600mg

PEP for Hep B

00:30:55

Healthcare worker status	Source is positive (HBs Ag +ve)	Source is negative (HBs Ag -ve)
• If exposed person is completely vaccinated &Ab titer >10 mIU / ml	• No further treatment required	
• If Ab titer of exposed person <10 mIU / ml • (vaccinated person)	• Give HBIg dose immediately (max. upto 7 days) • Start 2nd series of vaccine	• Start 2nd series of vaccine
• If exposed person is partially vaccinated or not vaccinated	• Give HBIg dose • Complete remaining vaccination	• Complete remaining vaccination
• Vaccinated for 2 series still Ab titers <10 mIU/ml (Non responders)	• Give HBIg 2 doses 1 month apart (0.06ml /kg) or (10-12 IU/kg)	• Nothing required

40 BIOMEDICAL WASTE MANAGEMENT

- Any waste generated during lab diagnosis, treatment or immunization, research activities

Waste generated in hospitals 00:01:04

- General waste = 80%
- BMW = 20%
  - 10% Infectious waste
  - 5% Chemical / Radioactive waste

Aim for waste management 00:02:24

- Reduce
- Recycle
- Recover

BMW guidelines 1998

- New guidelines given by Ministry of Environment & Forests
- Given in 2016 & amended in 2018-19

Steps of BMW management 00:04:17

1. Waste segregation: At point of generation
2. Pretreat: Lab liquid waste
3. Transportation of waste from generation site: Central storage area (CSA)
4. From CSA: Common Biomedical Waste Treatment Facility (CBMWTF)
   - Waste should reach from generation site to Common Biomedical Waste Treatment Facility within 48 hrs
5. Treatment and disposal of waste in 48 hrs

BMW guidelines 00:07:27

Refer Table 40.1

Logos used for segregation of biomedical waste

BMW Treatment Modalities 00:21:02

1. Incineration
   - Dry heat sterilization method
   - High temperature: 800 - 1200° C
   - Organic waste is converted into non organic waste
   - Waste volume is decreased

2. Autoclave
   - Moist heat sterilization method
   - Used for Infectious plastic waste & sharp waste

3. Chemical disinfection
   - 1-2% Sodium hypochlorite

4. Microwaving
   - Radio frequency waves of 2450 MHz are used

   ↓

   Produce friction of water molecules

   ↓

   Generation of heat

5. Hydroclaving
   - Low temperature steam method
   - is broken into small pieces
   - Done to avoid Shredding

6. Shredding
   - Waste is cut into small pieces
   - Helps in decreasing waste volume

7. Deep Burial
   - Pit is dug at 2 meters depth
   - Pit is half filled with waste & then covered with lime within 50cms of surface before filling rest volume with soil

8. Sharp Pits
   - Constructed within Hospital

9. Encapsulation
   - Immobilizing the material & sealing up the containers

10. Inertization
    - Mixing waste with cement before disposal

11. Plasma Pyrolysis
- Ionized gas is used in Plasma state
- Costly method

12. Expired Cytotoxic waste
- Resent back to manufacturer

Table 40.1

Category	Type of waste	Type of Bag / Container	Treatment / Disposal options
Yellow (Infectious non-plastic waste) 	Human anatomical waste	Yellow coloured non chlorinated plastic bags	Incineration / Plasma pyrolysis / Deep burial
	Animal anatomical waste		
	Soiled waste		Incineration / Plasma pyrolysis / Deep burial / Autoclaving or Hydroclaving + Shredding / Mutilation
	Expired / discardedmedicines Pharmaceutical waste, Cytotoxic drugs	Yellow colored containers / non chlorinated plastic bags with Cytotoxic label	Sent back to manufacturer/ CBMWTF for Incineration (Cytotoxic drugs at temperature >1200°c)
	Chemical solid waste	Yellow colored containers / non chlorinated plastic bags	Incineration or Plasma pyrolysis or Encapsulation
	Chemical liquid waste such as discarded disinfectants, infected body fluids and secretions, liquid from house – keeping related activities	To be discharged into separate collection system, which leads to effluent treatment system Not to be discarded into yellow bag	Pre-treated before mixing with other waste water
	Discarded linen waste contaminated with blood / body fluids, mask, cap, gown and shoe cover	Non–chlorinated yellow plastic bags / suitable packing material	Non–chlorinated chemical Disinfection followed by Incineration / Plasma pyrolysis
	Microbiology, other clinical laboratory waste, blood bags, live / attenuated vaccines	Autoclave safe plastic bag / container	Pre-treat to sterilize with non –chlorinated chemicals / on-site as per NACO / WHO guidelines + Incineration

Red (Infectious plastic waste)  	Disposable items such as • Tubing • Bottles • Intravenous tubes and sets • Catheters • Urine bags • Syringes (without needles and fixed needle syringes) • Vacutainer with their needles cut • Gloves • Plastic apron	Red colored non-chlorinated plastic bags or containers	1. Autoclaving / Microwaving / Hydrovlacing+ Shredding 2. Mutilation / Sterilization + Shredding 3. Treated waste sent to authorized recyclers or for energy recovery
White (Translucent) (Waste sharps including metal sharps) 	• Needles • Syringes with fixed needles • Needles from needle tip • Cutter or burner • Scalpels • Blades • Any other contaminated sharp (used or discarded)	Puncture-proof, leak-proof, tamper-proof containers	Autoclaving / dry heat sterilization followed by 1. Shredding or Mutilation or Encapsulation in metal container or cement concrete or 2. Sanitary landfill or 3. Designated concrete waste sharp pit
Blue (Glasswares & metallic body implant) 	• Broken • Discarded & contaminated glass including medicine vials & ampoules except those contaminated with cytotoxic waste • Metallic body implants	Blue colored containers	

41 BIOTERRORIST AGENTS

Categories

- Category A: Highest risk
- Category B: Moderate risk
- Category C: Emerging threats

Category A 00:01:19

- **Play** - **P**lague
- **Vith** – **V**iral hemorrhagic fever
  - Marburg
  - Ebola
  - KFD
  - YF
- **Small** – **S**mall pox
- **B** - **B**otulism
- **A** - **A**nthrax
- **T** – **T**ularemia

How to remember

- Play Vith Small BAT

Category B 00:03:43

- **C** – **C**lostridium perfringens
- **B** – **B**rucella
- **S**^{3} – **S**almonella, Shigella, Staph aureus enterotoxin B
- **E** – **E**. coli o157:H7
- **Vill** – **V**. cholerae
- **Take** – **T**yphus fever
- **Question for** – **Q** fever
- **P** – **P**sittacosis (LCL)
- **G** – **G**landers disease
- **Medical** – **M**elioidosis (caused by B. pseudomallei)

How to remember

- CBS^{3}E Vill Take Question for PG Medical

Category C 00:10:09

- **N** - **N**ipah virus
- **H** - **H**anta virus
- **S** - **S**ARS corona virus, SARS cov-2

PREP NUGGETS

Prep Nuggets

Motility	Organisms
Tumbling	.....
.....	Vibrio
.....	Proters
Falling leaf	.....

Prep Nuggets

	Transport media
UR Media	.....
Carry Blair	.....
.....	Streptococcus pyogenes
.....	Shigella

Prep Nuggets

Meningococci	Gonococci
.....	Non capsulated
Lens shaped	.....
.....	Ferounts glucose only
.....	

Prep Nuggets

Oligate aerobic	Obligate anaerobic
.....	.....

Prep Nuggets

Condition	Safe in pregnancy	Avoid in pregnancy
Hypertension	.....	.....
Hyperthyroidism	.....	.....
Anti-epileptic	.....	.....
Bipolar	.....	.....

Prep Nuggets

Receptors	Dose of depamina
D_1	.....
β_1	.....
α_1	.....