CREDIT - I

Saturday
24/8/24

Concepts of Medicle Micro-Biology

(A) Factors Determining of Microbial Pathogenicity —

1) Host Suceptibility —

- Resistance to bacterial infections is enhanced by phagocytic cells and an intact immune system.
- Initial resistance is due to nonspecific mechanisms. Specific immunity develops over time.
- Suceptibility to some infections is higher in the very young & the very old and in immunosuppressed patients.
- Suceptible hosts are also known as people who are at risk, such as patients or healthcare workers.

2) Host Resistance —

- Numerous physical & chemical attributes of the host protect against bacterial infection.
- These defence include the antibacterial factors in several secretions covering mucosal surfaces & rapid rate of replacement of skin & mucosal epithelial cells.
- once the surface of the body is penetrated, bacteria encounter an environment virtually devoid of free iron needed for growth, which requires many of them to scavenge for this essential element
- Although easily damaged, the skin represents one of the most important barries of the body to the microbial world, which

contains a divers array of bacteria in enormous membrane.

- fortunately, most bacteria in the environment are relatively begin to individuals with normal immune system.
- however, patients who are immune-suppressed such as individuals receiving cancer chemotherapy or have AIDS, opportunistic microbial pathogens can establish life threatining infections.

* Presence of bacterial virulence factors —
- Virulence factors in bacteria may be encoded on chromosomal DNA, bacteriophage DNA, plasmids or transposones in either plasmids or the bacterial chromosome.
- The virulence factors of bacteria can be divided into a no. of functional types.

* Host Susceptibility —
- Depends upon physiologic and Immunologic condition of host and virulence of bacteria

• Bacterial pathogen —
Non specific response of host resistance

① Polymorphonuclear Neutrophils

② Macrophage clearance.

- they ↑ no. of specific antibodies.

- Normal skin bacterial flora and mucosal surfaces protect the host against colonization.

- Individual suceptible to infection with the variety of bacteria if skin or mucosa is breached.
  - severe wounds
  - burns
  - surgical contamination.

- Cystic fibrosis (damages lung and digestive system).
- unable to clear mucous from respiratory tract.
- Susceptible to mucoid strains of pseudomonas aerogenosa.

- E. coli → Ascending urinary tract infection.
- transmission → rocky mountain spotted fever & Lyme disease caused by ticks.
- Bubonic plague → Fleas.

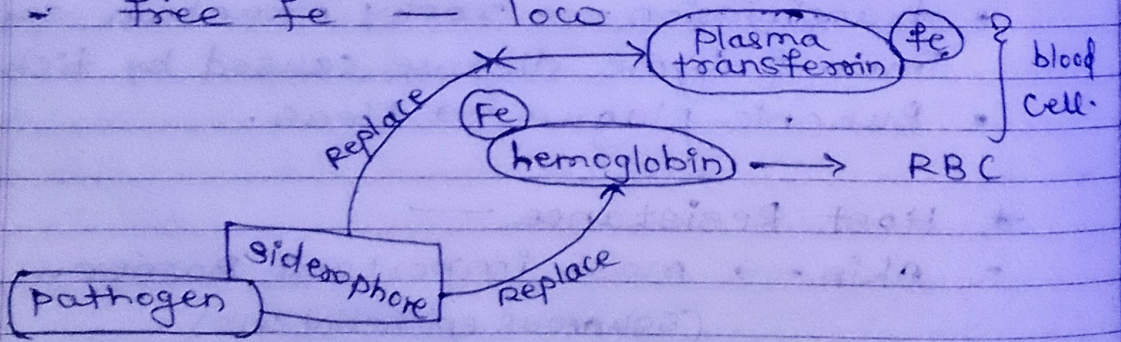
* Host Resistance —
- Skin → most important Barrier.
  (Squamous epithelial cells) outer layer
  Dead cells.
  dry, acidic environment, temp. less than 37°C.

- Immunosuppressed → weak immune system
- ~~Immunocompressed~~ →

- receives cancer chemotherapy.
- have AIDS
- oppurtunistic pathogens can establish life threatning infections.

- Mucous — respiratory tract, Gastrointestinal tract, Urogenital system through which bacteria can gain access to the body.
  - mucosal epithelial cells divides rapidly.
  - 36 – 48 hrs → complete replacement of epithelium.
  - Lysozyme.
  - Lactoferrin } To kill bacteria or resist growth.
  - Lactoperoxidase }
  - contains secretory immunoglobulins IgA.

- Competition of Iron —
  - free fe — loco



- Infection → Phagocytic cell.
  Lymphocytes.

* Bacterial Virulence Factors ——

→ Virulence factor ⟶ to cause severe infection.

- Adherence ⟶ not absorb only stick.

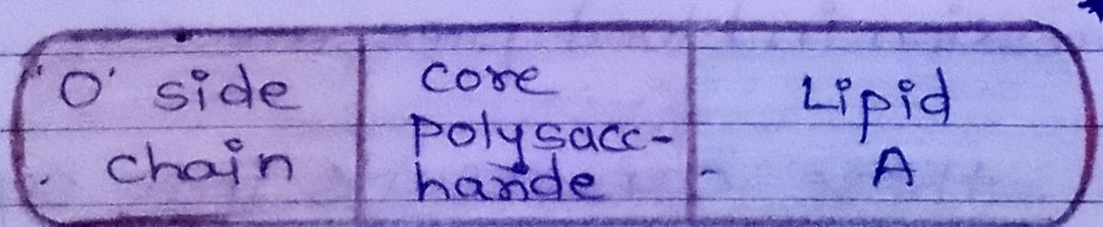
1) Adherence & Colonization factors ——

- To cause infection many bacteria first adhere to a mucosal surface.
- eg. Alimentary tract mucosa is continuously cleans by the release of mucous from goblet cells & by the peristaltic flow of gut contents over the epithelium.
- Similarly ciliated cells in the respiratory tract sweepe mucous & bacteria upward.
- The turnover of epithelial cells at this surfaces is farely rapid.
- The intestinal epithelial cells at this surfaces is farely rapid.
- The intestinal epithelial cells monolayer is continuously replenished & the cells are pushed from the crypts to villar tips in about 48 hrs.
- To establish an infection at such a site a bacterium most adhere to the epithelial & multiply before the mucous extruded epithelial cells.
- To accomplish this bacteria have evolved attachment mechanisms such as pili (fimbriae) that recognize an attached bacteria to cells.
-

* Endotoxins –

- Class of toxic substances release after lysis of bacteria.
- It is a molecular complex of lipid & polysaccharide so that, called lipopolysaccharide.
- It is a toxic lipopolysaccharide component of the outer membrane of Gram Negative Bacteria.
- It can have Harmful effects on the host.
- It can be so severe may result into death.
- Omnipresent (present everywhere at same point)

• Basic structure of Endotoxin of Gram –ve Bacteria ——



* All Endotoxin molecules are similar in chemical structure & biologic activity though some diversity has evolved.

O side chain	core Polysaccharide	Lipid A
- Oligosaccharide		
- consists variety of repeating oligosaccharide residues.	- forms back bone of macromolecules.	- composed of Glucosamine disaccharide. with attached long chain fatty acids & phosphate
- specier / serotype antigen	- Genus specific antigen.	
- eg. Family Enterobacteriaceae	- Responsible for antigenic diversity	- Toxic moiety (group)
		- Gives toxicity.
	- Eg. N. gonorrhoeg N. meningitidis B. pertusis	- Examples N. gonorrhoea N. meningitidis B. pertusis

* Biological Activity of Endotoxin —

1) Pyrogenicity → Induce Fever
2) Leukoperia → Decrease WBCs
3) Leukocytosis → Increase WBCs
4) Complement action → clear Infection.
5) Hypothermia → dropped body temp.
6) Depression in blood pressure.

- All these can results into sepsis or lethal shock. (Near death)

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* Responses Beneficial to Host —

1) Mitogenic effect on B Lymphocytes —
- Essential for growth, tissue repair.
- increase resistance to viral & bacterial infections.

2) Induction of gamma interferon production by B-Lymphocytes —
- Enhance antiviral state p cm
- promote rejection of tumor cell.
- Activate macrophages & Natural killer (NK) cells.

3) Induction of formation of IL-1 → by macrophages —
- IL-2 aftermediaters → by T lymphocyte.

4) Activation of complete cascade with the formation of —

a) C3a → fragment promote inflammation by inducing release of histamine.

b) C5a → stronger than C3a → activate
- Neutrophils & other immune cells
- vascular permeability.

5) Endotoxin —
- Largely accumulated in the liver following

injection of sublethal dose by intravenous route can be demonstrating because its ability to affect variety of cells & host proteins.

CD_{14} → Kupffer cells

Receptor on surfaces (CD_{14}) } → Granulocytes → macrophages → platelates & Lymphocytes

- CD_{14} binds endotoxin enhance by interaction with a host protein made in liver.
- Result in a complex array of host responses that can culiminate in the serious condition gram negative species.
- Leads to shock & death.

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* Detection of Endotoxin in medicle solution —

1) Pyrogenic (Induced fever) —
2) Destructive properties
3) Need Extreme care to avoid patient exposure.

- Solutions such as salines, intravenous administrations becomes contaminated because of improper handling.
- Autoclave sterilization } ⇒ does not work
- Bacterial membrane filtration

- filtration through ion exchange resins is necessary to remove endotoxin.
- If endotoxin containing solutions were used in :
  (i) renal dialysis → patient could suffer immediate fever,
  (ii) Heart by-pass machines → alteration in blood pressure.
  (iii) Blood transfusions
  (iv) Surgical lavage.

• To ensure sterility :—
1) Limulus lysate test (only for gram-ve B'A)
2) Rabbit pyrogenicity test

* Exotoxins —
- protein toxins released from viable (live) bacteria
- class of poisons - most probable / unit weight.
- present in both Gram +ve & -ve genera.

Exotoxins

① Heat stable
- Low molecular sized

② Heat Labile
- Heigher molecular sized

- Site of action → more localized
  (confirmed to particular cell types / cell receptors)
  eg. Tetanus toxin → affects only interneuncial neurons.

- Some antibodies —(reacts with)→ Important binding sites
  - enzymatic sites
  - complete inhibition → Neutralization

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Exotoxins

① Neurotoxins

- Clostridium sp.
Botulinum toxin acts on motor neuron, by preventing the release of acetylcholine at myoneural junction, prevents muscle excitation and produce flaccid paralysis.

② Cytotoxins (Kill cells)

- larger
- more heterogenous grouping with a wide array of host cells.
- toxin blocks elongation of peptide chain.
eg Diphtheria toxin Corynebacterium diptheriae

③ Enterotoxins (Intestinal)

- Highper secretion of water & electrolytes.
- Intestinal epithelium damage results into water diarrhea.

(i) Cytotoxic

- Directly kill or damage intestinal cell.
- bloody diarrhea
- eg. shiga like enterotoxin from E. coli

(ii) Cytotonic

- Alter cellular function.
- Increase ion, chloride and water.
- watery diarrhea.
eg. cholera toxins.

- Distrub normal smooth muscle function results, into abdominal decrease time for water absorbtion in intestine.

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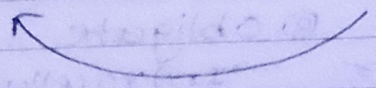
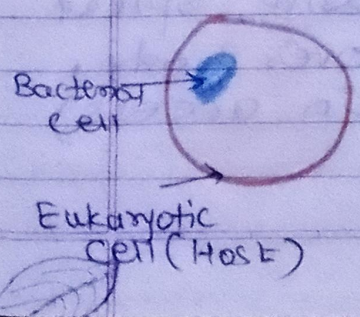
* Presence of Host mediated pathogenesis —

- Bacterial Infection ⟷ Host Immune Response
  (Pathogenesis) ↓ Tissue damage more than



- Most common gram negative bacterial sepsis (Shivering fast heart beat, low bp, sweaty skin, pain or discomfort in the body)
- Tuberculosis (Mycobacterium tuberculosis)
  Tuberculoid leprasy → skin lession
- Tissue damage → toxic substances released from Neutrophils, lymphocytes, macrophages at the site of infection.
- Host Response is so intense that host tissues are destroyed allows resistant bacteria to invade in body.
- In lepromatous leprasy there is a contrast absensce of cellular response to Mycobacterium leprae allows bacteria to multiply to such large nos. in skin, which replaces healthy tissue.

* Ability for Intracellular Growth —



- Bacterial cell invade the Host to survive.
- shielded from host immune Response of humoral antibodies from special mechanism to protect from lysozyme.

- Bacteria can be call intracellular once ingested by Neutrophils & macrophages.

- It can be eliminated by any cellular immune response (T cell) to target infected cell.

- **Pathogenic Bacteria** ——

① Facultative Intracellular
- Salmonella
- Shigella
- Mycobacterium

② Obligate Intracellular
- Rickettsia
- Coxiella
- Chlamydia

③ Extracellular
- vibrio cholerae
- B. pertusis

- Bacteria produces phospholipase (enzyme that breaks down phospholipids into fatty acids & other substances) that dissolves phagocytic veside that surrounding the bacteria who have preffered ~~for~~ intracellur environment for growth.

- In Extracellular bacteria they do not invade the cell, get enriched by body fluids.
  eg ① vibrio cholerae
  ② B. pertusis (do not penetrate body tissue, adhere epithelia surface, secret protein toxins that cause disease)

③ E. coli
④ P. aeroginosa
(Non invasive, spread rapidly over body, once gain access)

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* Molecular Basis of bacterial pathogenesis —

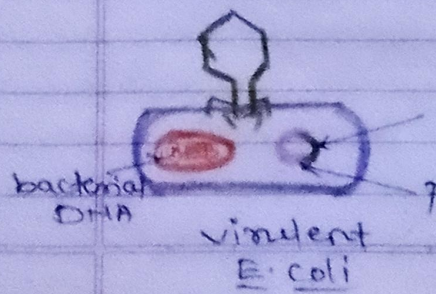
1) Virulence Genes

- Virulence factors in bacteria may be encoded on chromosomal DNA, bacteriophage DNA & plasmid
- This virulence factors transferred from one bacterium to another by conjugation, Transduction and Transformation.

1) conjugation → through conjugation tube formation.
2) Transduction → via the bacteriophage transfer of genetic materials
3) Transformation → transfer of genetic material through the plasmids.

- In this there are several steps mentions:

(i) Transfer of virulent gene to avirulent (Non-pathogenic) bacteria.
(ii) Change to virulence
(iii) Intestinal disease due to virulence gene or symptoms on the host by:
  (a) adherence
  (b) Enterotoxins
  (c) Invasiveness
  (d) Cytotoxicity.



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1 mda → 1 million dalton

1) Plasmid — carry genes for specific circumstances
- present in cytoplasm

eg. (i) - Shigella species → To invade cells, this property is encoded on 140 MDa plasmid.

(ii) - Heat labile enterotoxin (LTI) of E. coli is also encoded on plasmid

2) Bacteriophage — transfer genetic material through a process called Lysogamy
- Virulence factors encoded on bacteriophage
- Bacteriophage integrates its genome into the bacterial chromosome.
- Temperate Bacteriophage → Infect bacteria & archeae basis of toxin production in pathogenic bacteria.
- eg Corynebacterium diptheriae → (diptheria toxin)
  S. pyogenes → Erythrogenic toxic
  E. coli → shiga like toxin
  C. botulinum → Botulinum toxin

3) Chromosomes —
- present in nucleus, carries essential genetic information for life

eg. (i) Heat labile toxin (LTIT) of E. coli encoded on chromosome.
(ii) Cholera toxin
(iii) Salmonella enterotoxin.
(iv) Yersinia invasion.

* Enzymes are proteins that can speed up chemical reaction.

* ENZYMES —

* Transfer of Gene Reduces antibiotic resistance which creates medicle problem.

- This provides the opportunity for resistant bacteria proliferate and (invade in body) produce other virulence factors in patients if, they receive inappropriate antibiotics.
-
- Several bacterial proteins toxins are enzymes required for ——
  (i) Cellular signalling
  (ii) Membrane dynamics
  (iii) Cell Migration
  (iv) Cell Growth
  (v) Cell Death

eg: ① cholera toxin
② Diptheria toxin
③ pertusis toxin
} toxic effect on host is related to the pathogenesis of bacterial infection

- The function of this enzymes in normal bacterial physiology in not known.
- from the bacteria's view it requires to much energy to synthesize such as high molecular weight & complex molecules (bacterial toxins proteins), if they offered no advantage.

* Primary goal of microorganism is to acquire nutrients & multiply rather than to harm the host.

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* Siderophore —

- Substances reduce by many bacteria to capture iron from host.
- Iron require for metabolism & growth of a cell which results into conflict between pathogen & host.
- Animals evolve mechanism of with holding iron from tissue fluids to limit growth of bacteria.
- Iron is not free if any of the cell, <u>withholding mechanism.</u>

<u>RBCs</u> — Iron — hemoglobin

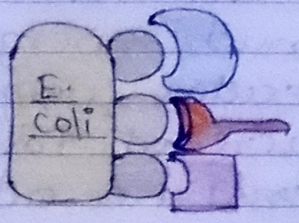
<u>plasma</u> — Iron — Transferrin

- Milk
- Tears
- Saliva
- Bile juice
- Gastro-intestinal fluid

} — Iron — Lactoferin

- Some bacteria have receptors for Eukaryotic iron binding proteins.

- Hemoglobin
- Transferin
- Lactoferin



- Iron Acquisition is facilitate for bacterial growth.
- This is important virulence mechanism.

<u>eg.</u> Enterochelin produced by <u>E. coli</u> & <u>Salmonella</u>

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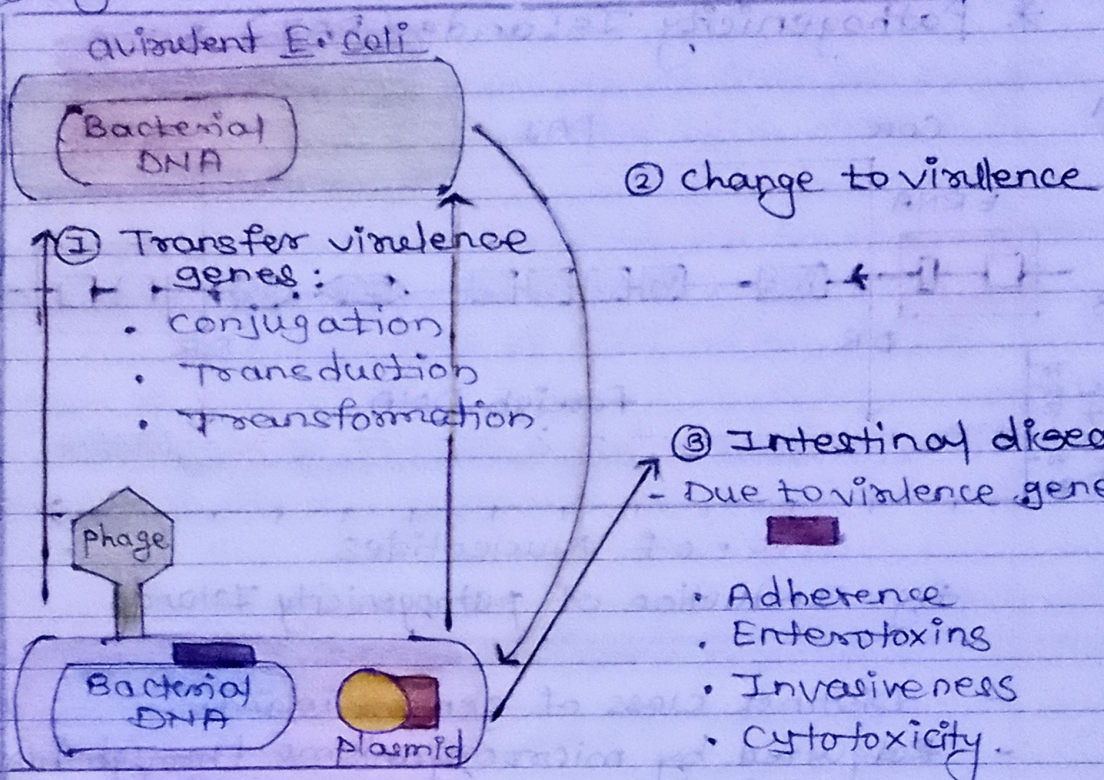

fig. Mechanism of Acquiring bacterial virulence gene

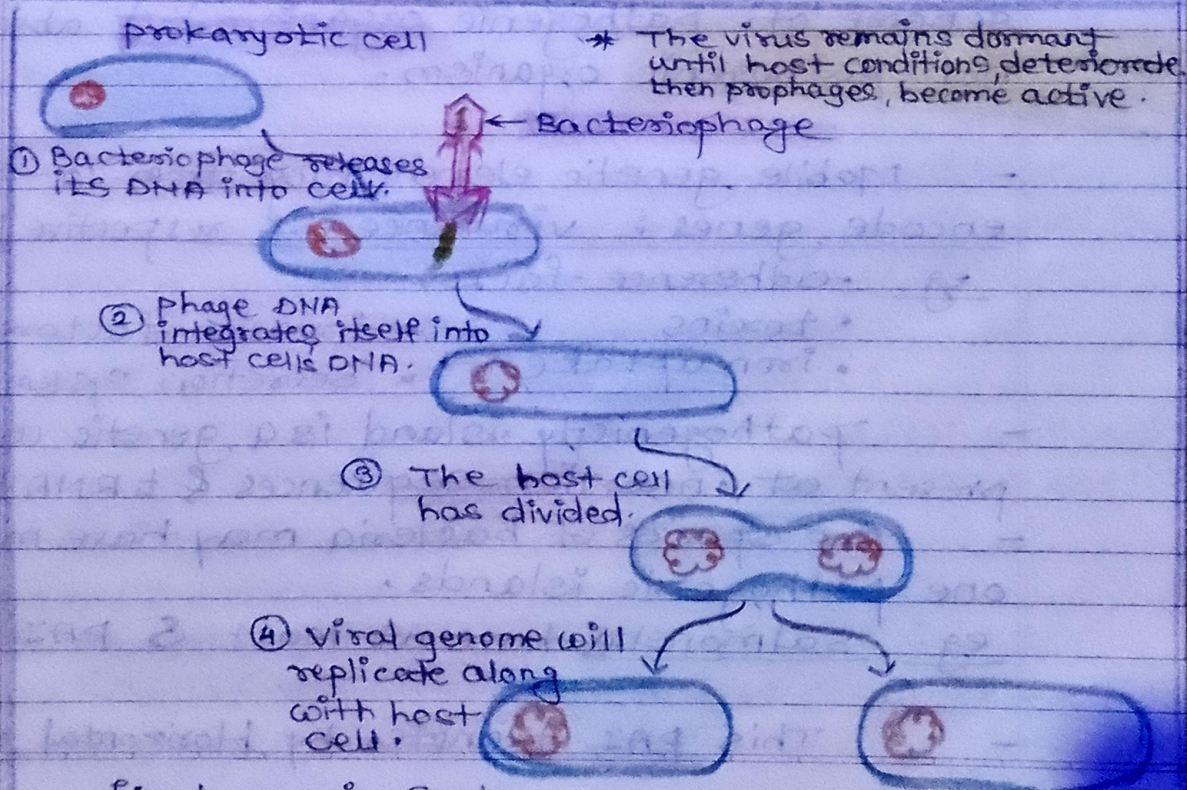

fig. Lysogenic Cycle.

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* Pathogenicity Islands (PAI)

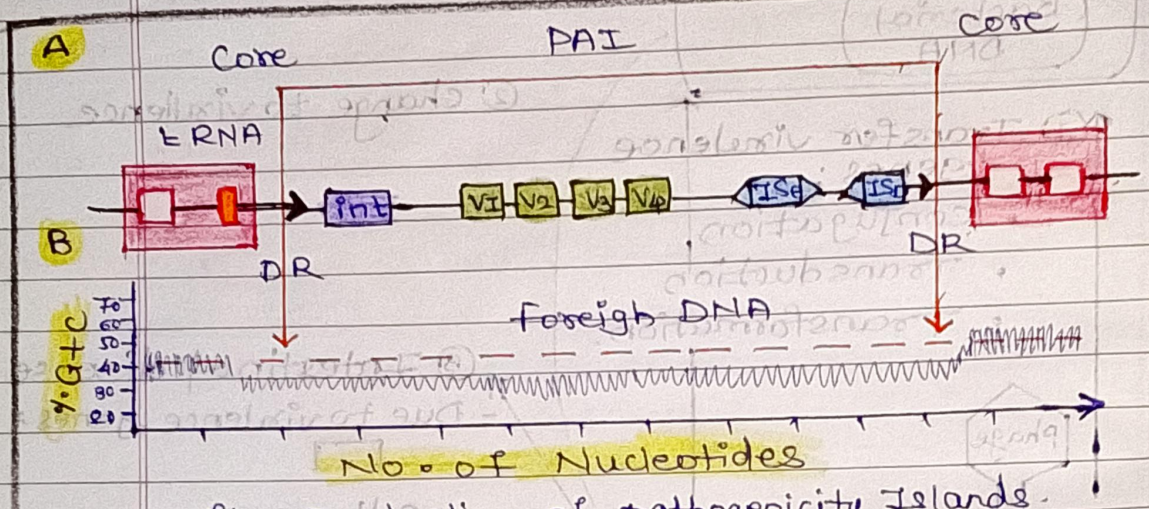

fig. Induction of pathogenicity Islands.

- Distinct class of genome island.
- Acquired by microorganisms through horizontal gene transfer : conjugation, transduction and transformation.
- Pathogenicity island incorporated in genome of pathogenic organism but absent in non-pathogenic organism.
- Mobile genetic element range from 10-200kb encode genes : virulence of respective pathogen
  eg. • adherence factor
  • toxins • invasion factor
  • iron uptake • Secretion system (SS)
- Pathogenicity island is a genetic unit present at insertion sequences & t RNA sequence
- one species of bacteria may have more than one pathogenic islands.
  eg. Salmonella have at least 5 PAI.
- This PAI transfer by Horizontal gene

transfer event, carries genes for one or more virulence factors like adhesins, toxins & invasives.
- PAI may be located on bacterial chromosome plasmid or transferons.
- G+C content of PAI often differs from rest of genome.
- DNA segments more than one link with virulence genes, were described as virulence gene blogs.
- Pathogenicity DNA island — pathogenicity island

• Structure of PAI —

(A) Typical PAI —
- Distinct region of DNA present in pathogenic organism absent in non-pathogenic
- Mostly inserted in a backbone of genome of host strength in specific site.
• t-RNA → mobile genes / Integrases (int) located begining of island close to tRNA locus.
- PAI harbours one or more virulence gene V_1, V_2, V_3 & V_4 & transfer with mobility element i.e. Insertion elements (IS), either
ISe - complete insertion element.
ISd - Defective insertion element.
- PAI boundry's determine by DR's (Direct Repeats → triangle) DR = A Direct repeat use for insertion & deletion process.

(B) A characteristic feature of PAI is :
- a G+C content different from that of core genome.

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- Common features of pathogenicity islands -
(i) presence of virulence genes
(ii) Specific presence in pathogens
(iii) Large distinct chromosomal region (10-200kb)
(iv) Characteristic based composition different from core Genome (G+C content %)
(v) Insertion of pathogenicity islands (PAI) adjacent to t-RNA genes.
(vi) Frequent Association with mobile genetic elements i.e presence of :
  - Direct Repeats (DR)
  - functional Integrates (int)
  - OR
  - Transposes
  - IS Elements (Insertion Elements)
  - Chromosomally integrated conjugative Transposons, plasmids and phages.
(vii) Genetic instability if functional mobility elements are present.

- Virulence factors Encoded by pathogenicity Islands —
- Bacterial virulence determinants are encoded by and associated with mobile genetic elements :
  * phages
  * plasmids
  * Insertion Elements.
  * Transposons (Translocation of genetic sequence)

- Most pathogenicity factors interacts with eukaryotic host cells, expose :
  - either at the surface of bacterial cell.

    OR
  - Transported out of the bacterial cell probably into eukaryotic cell.

- To Export virulence factors, five, different protein secretion system is present :

(1) Protein Secretion System Encoded by pathogenicity Islands - (PSS)

- secretion of protein is a general requirement of bacteria for cell envelope assembly.
- Metabolism, Defence against host, Interaction with host during pathogenesis.

Gram -ve Bacteria	Gram +ve Bacteria
(i) presence of outer membrane. (ii) Evolve variety of structurally & functionally different secretion system (SS)	(i) Extracellular & surface proteins are secreted by, the general secretion pathogen mechanism.

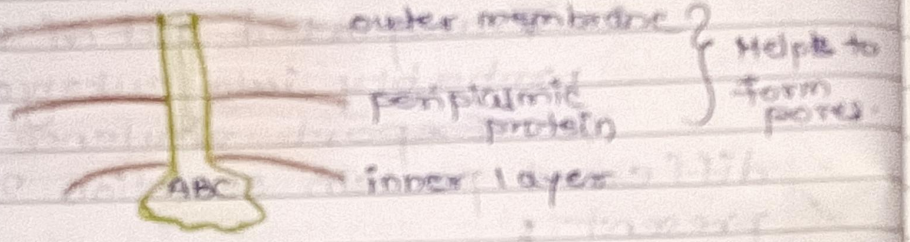
- following systems only present in Gram - ve

(a) Type I Secretion System (T1SS) :-

- Consist of simple assembly of an ATP binding cassette (ABC), Transported protein, located within inner membrane.
- A periplasmic protein & outer membrane protein forms the secretion pores.

- Substrate type 1 secretion system (T1SS) are delivered into extracellular medium.

eg. hemolysis.



(b) Type 2 secretion system (T2SS) —

- Complex assemblies.
- Main terminal branch of the general secretion pathway.
- Default machinery for protein secretion in pathogenic & non-pathogenic bacterial species.
- In Gram +ve and Gram -ve bacteria proteins transported across cytoplasmic membrane by the secretary system.

(c) Type 3 secretion system (T3SS) —

- Complex assemblies required function of 20 genes.
- virulence in flagellum assembly machinery system.

(d) Type 4 secretion system (T4SS) —

- Translocates proteins into a eukaryotic target cell.
- Complex structure
- 10 subunits.

(e) Type 5 secretion System (T5SS) —

- Autotransporters
- Secretion of proteins.
- Secretion.

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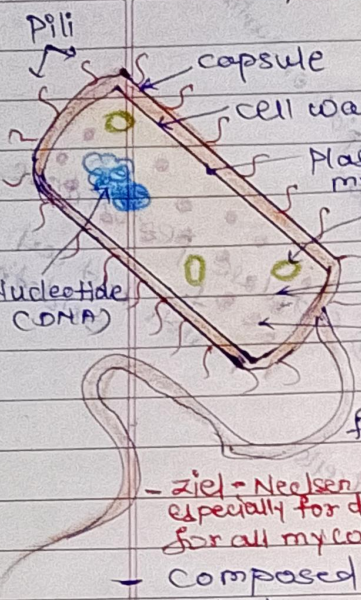
Mycobacterium tuberculosis (TB)



- Ziel-Neelsen staining especially for detection of Tb & for all mycobacterium sp.
- composed of waxy cell wall
- strictly aerobes (Needs O_2)

1) General characters —

- Gram +ve Bacilli.
- Straight / Slightly curved rod shape
- about 3 µm x 0.3 µm size
- non-motile
- Also called AFB bcoz — when stained with carbon fuschion by ziel-Neelsen method, they resist decolourization by 20% H_2SO_4.
- Acid fastness is because of mycolic acid present in semi-permeable membrane of bacc. wall of mycolic acid & also called as Mycobacteria.
- Mycolic acid contains lipids & it make lipid rich cell wall.
- Doesn't stain with Gram Stain — (because of its thick, waxey cell coating.

2) Cultural characters —

- an obligate aerobes.
- Grows slowly with generation time of 14-15 hrs.
- Colonies appear in about 2-6 weeks
- temp. range 25°C - 40°C, optimum 37°C & PH 6.4 - 7
- Growth media enriched with eggs, glycerol, potatoes, meat, bone marrow infusions.
- Media used → (a) Dorset-egg medium (b) Lowenstein-Jensen's medium (c) Lofflers serum slope.

3) Biochemical Reactions —

- for Identification, Some imp. biochemical Reab. are as follows:

(i) **Niacin Test** - It forms niacin (nicotinic acid) when grown on egg medium.

Test → 10% cyanogen bromide + 4% aniline in 96% Ethanol + culture suspension → 37°C for 24 hrs.

Yellow colour +ve test

- useful for Identifying Human Strains, as no other Mycobacterium is positive for this test.
  i.e. It is specific for only M. tuberculosis.

(ii) **Aryl Sulfatase test** - +ve only with atypical Mycobacterium

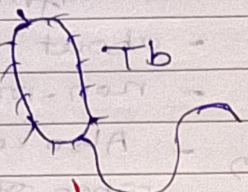
Test → bacilli grown on media containing tripotassium phenolphthalein disulfate taken.
add alkali drop by drop → pink colour +ve test
eg. $CaCO_3$, Sodium Hydroxide, potassium hydroxide, sodium carbonate & bicarbonate

(iii) **Neutral Red Test** -

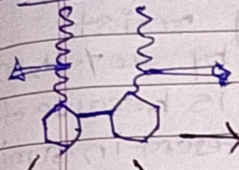
(iv) **Catalase-peroxidase test**

4) pathogenesis —

- **virulence factors** —



① Trehalose Dimycolate (cord factor)



→ cytokine release

↓ Evades immune response

↓ Granuloma for^n & trigger cytokine release

② sulfatides → These are glycolipids on surface of mycobacteria

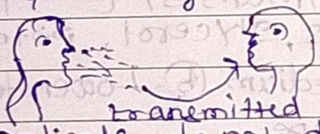
- prevent phagocyte activation
- prevents phagolysosome fusion.

↳ This allows survival of b'a inside of cells.

③ catalase peroxidase → Resists host cell oxidation.

- **Transmission**

- TB is transmitted through inhalation & that is how it gains access to your lungs.



transmitted through inhalation.

- Most patients do not develop active TB inf^n, usually it either clears up or remain latent.

- **pathogenesis** —

TB enters alveoli → where macrophage phagocyte them → usually with most pathogens macrophages packaged them in phagosome & fuse with lysosome → which breaks it but that is not possible bcoz various factors.

↳ so what happens is the TB starts to grow inside macrophage.

↳ eventually it kill macrophage & spreads into lungs.

macrophage →

Phagosome →

few weeks later → cell mediated immunity fight against it → Type-4 Hypersensitivity reac^n: TH1 Response (CD4 T-cell) ↓ Inf-γ → T-lymphocytes activated

↓ This leads to formation of granuloma. → It tries to resist spreading of TB by process called caseating Necrosis. → It forms GHON complex in lungs → Leads to Clear inf^n OR Remains Latent in lungs.

- Latent → Remains dormant
- person with weak immunity eg. HIV can active latent TB rapidly.

• Signs & symptoms —
- cough with blood - fever - weight loss - night sweats.
- chest pain - fatigue

5) Diagnosis —
- M. tb. grows slowly, becoz growth is slow cultures of clinical specimens must be held for 6-8 weeks before being recorded as -ve
- they need a special media to grow called Lowenstein Jensen media

① TST (Tuberculin Skin test) — also called Mantoux test.
- it is done through a subcutaneous inj. of purified protein derivative PPD (Purified protein derivative.
↓
-Diagnosis made on basis of diameter of induration

② QFT-G (Quantiferon TB-Gold) —
it is new test for TB & it has very high specificity & sensitivity & hence is considered the gold standard test for latent TB

If above tests are postive.
↓
-we can proceed for chest X-ray, if it has findings suggestive of TB then the confirmatory test could be sputum sample to stain for AFB.

③ BAL → Bronchoalveolar lavage : is a procedure that can be used to diagnose TB

6) Treatment —
- Active patients requires a multi drug regiment which includes
RIPE → stands for Rifampin, Isoniazid, pyrazinamide & Ethambutol.
- patients with latent TB can be given isoniazid as a monotherapy

⑦ Prevention —
- Better Housing & Nutrition can decrease your chances of getting TB.
- The infection spread can be reduced with prompt identification & adequate treatment of patients.
- Using masks & other respiratory isolation procedures.
- Vaccine for TB known as BCG vaccine → it is live attenuated vaccine contains a strain of M. bovis (Bacillus Calmette-Guerin)
↓
It provides immunity against tb.

8) Chemotherapy :—

- Chemotherapy for TB patients primarily involves a regimen of multiple antibiotics (combination of multiple antibiotics → A treatment that involves administering more than one antibiotic to patient at same time) to ensure effective treatment & reduce risk of drug resistance.

- The standard treatment for active TB typically lasts about 6 months & consists of two phases :

(a) Intensive Phase (first 2 months) —
It uses RIPE antibiotics —

① R → Rifampicin :—
Dosage → Typically 10 mg / kg daily
Mechanism → Inhibits bacterial RNA synthesis by binding to RNA polymerase.
side effects → Hepatotoxicity, orange discoloration of bodily fluids.

② I → Isoniazid :—
Dosage → 5 mg / kg daily
Mechanism → Inhibits cell coell synthesis of mycobacteria
side effects → Rashes on skin, Hepatotoxicity.

③ P → Pyrazinamide :—
Dosage → 25 mg / kg daily
Mechanism → Disrupts mycobacterial cell membrane metabolism & transport functions.
side effects → Heapatotoxicity & possible joint pain.

④ E → Ethambutol :—
Dosage → 15 mg / kg daily
Mechanism → Inhibits synthesis of mycobacterial cell wall.
side effects → optic neuritis (vision changes), allergic reactions.

(b) Continuation phase (Next 4 Months) —

Isoniazid & Rifampicin are typically continued for an additional 4 months.