College Of Veterinary Medicine .

3rd class First Semester

MICROBIOLOGY- BACTERIOLOGY *The Eighth – Lecture*

VIRULENCE AND GENETICS BACTERIA :-

Introduction:-

Virulence is the ability of a microorganism to produce disease. Virulence depends on the number of infecting bacteria, their route of entry into the body, the response of the host immune system and any characteristics specific to that bacteria – its virulence factors. Bacterial virulence factors are typically proteins or molecules synthesized by protein enzymes.

Mechanisms of Typical Virulence Factors :

The general categories for which virulence factors can be broken down into are as follows:

- **Adhesion** - Virulence factors which facilitate adhesion are known as adhesins, they are what allow bacteria to bind to host cells. For example *Bordetella bronchiseptica* adhesins include; fimbriae, filamentous haemagglutinin adhesin (FHA) and pertactin. These adhesins bind to specific epithelium receptors and epitopes, which is what derives tissue specificity. Some bacterial cell walls and capsules are also adhesins, these are able to maintain even closer contact.

- **Colonization –** Some virulence factors ease colonisation of certain areas of the body, for example *Helicobacter* species counter the low pH of the stomach by producing urease. This helps to neutralise the stomach acids making the stomach more colonisable for the *Helicobacter*.
- **Invasion –** Virulence factors which facilitate bacterial invasion of a host. This is done by disrupting host cell membranes, the result is the facilitation of transport across epithelial layers of tissue and skin. For example, the internalin surface proteins found on *Listeria monocytogenes* allow them to invade mammalian cells via transmembrane proteins. Thus these virulence factors are facilitating invasion into human epithelial cells.
- **Immune Response Inhibitors** - These are virulence factors which target the immune system and of the host and inhibit immune responses initiated against the bacteria. Even a low dose of bacterial exotoxin can alter the immune response. For example *Escherichia coli* heat liable toxin reduces the expression

of inter leukin 12 (IL-12) a cytokine which is indicative of the immune response.

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- ## Virulence Factors :

1- Enzymes .

Numerous enzymes have been implicated in microbial virulence. Although the number of enzymes in this category is vast. Enzymes that are considered virulence factors are generally active against host components and contribute to virulence by damaging host tissue damage makes the host permissive for microbial infection. Enzyme virulence factors that damage tissue include proteases, neurominidases and phospholipases.

2- Modulins .

A large group of microbial compounds can damage a host by eliciting inflammatory responses. These compounds often do not meet the classical criteria for virulence factor definition because they are necessary components of bacterial cells. Bacterial lipopolysaccharide is a wellknown example of a microbial compound that can cause

massive host damage by interacting with Toll-like receptors and triggering an inflammatory cascade. Microbial products that elicit detrimental cytokine responses, such as lipopolysaccharide,

3- Motility .

Motility is a complex trait that has been associated with virulence in both bacteria and parasites. Motility is manifested by approximately 80% of known bacterial species and is critical for the adaptation of mobile microbes to new environments .

Bacterial cells can move by the action of specialized organelles called flagella. For movement in intracellular spaces, many microbes exploit host actin to propel themselves forward . Actin-based motility is used by several intracellular pathogens including Shigella spp., Listeria monocytogenes and Rickettsiae for cell-to-cell spread Like bacteria,

4- Capsules .

Many pathogenic bacteria possess polysaccharide capsules, which are required for virulence in mammalian hosts. Encapsulated bacteria with polysaccharide capsules include Streptococcus pneumoniae, Neisseria meningiitidis and Haemophilus influenzae. Among the eukaryotes, only C. neoformans has a polysaccharide capsule. Most capsules function in microbial pathogenesis by protecting the microbe against host immune mechanisms, although for some the capsular structures can serve as adhesins. For example, polysaccharide capsules are usually poorly immunogenic.

5-Pigments .

Pigment production, and specifically melanin-like pigments, have been associated with virulence in several microbes .

Melanin in melanotic organisms can protect against a variety of host defense mechanisms that include free radical fluxes.

6-Toxins – A large proportion of virulence factors are proteins produced by bacteria which are toxic to the host, these toxins cause damage to hosts cells and tissues.

GENETICS:-

Genetics is the study of genes including the structure of genetic materials, what information is stored in the genes, how the genes are expressed and how the genetic information is transferred. Genetics is also the study of heredity and variation.

The arrangement of genes within organisms is its genotype and the physical characteristics an organism based on its genotype and the interaction with its environment, make up its phenotype. The order of DNA bases constitutes the bacterium's genotype. A particular organism may possess alternate forms of some genes.

Such alternate forms of genes are referred to as alleles. The cell's genome is stored in chromosomes, which are chains of double stranded DNA. Genes are sequences of nucleotides within DNA that code for functional proteins.

The genetic material of bacteria and plasmids is DNA. The two essential functions of genetic material are replication and expression.

Some fundamentals of bacterial genetics :

◙Bacterial DNA (like any DNA) can be altered by mutations .

◙Mutations can result in changes in proteins diversity acquisition of resistance new traits can be transmitted to other microbes .

◙What happens when DNA is mutated?

◙Bases (nucleotides) are changed (base substitution), Bases are inserted or deleted Sequences of DNA move around the genome (transposons).

Mutations :

The term "mutation" was coined by Hugo de Vries, which is derived from Latin word meaning "to change". Mutations are heritable changes in genotype that can occur spontaneously or be induced by chemical or physical treatments. (Organisms selected as reference strains are called wild type, and their progeny with mutations are called mutants.) The process of mutation is called mutagenesis and the agent inducing mutations is called mutagen.
Changes in the sequence of template DNA (mutations) can drastically affect the type of protein end product produced.

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For a particular bacterial strain under defined growth conditions, the mutation rate for any specific gene is constant and is expressed as the probability of mutation per cell division. Spontaneous mutation occurs naturally about one in every million to one in every billion divisions. Most spontaneous mutations occur during DNA replication .

Mechanisms of mutation :

(A) . Substitution of a nucleotide:

(B). Deletion or addition of a nucleotide:

Results of mutation :

(a) Missense mutation

(b) Nonsense mutation:

(c)-- Silent mutation:

(d)-- Frameshift mutation:

(e)-- Lethal mutation:

(f)-- Suppressor mutation

(g)-- Conditional lethal mutation:

(h)-- Inversion mutation:

Significance of mutation:

1• Discovery of a mutation in a gene can help in identifying the function of that gene.

2• Mutations can be induced at a desired region to create a suitable mutant, especially to produce vaccines.

3• Spontaneous mutations can result in emergence of antibiotic resistance in bacteria.

4• Mutations can result in change in phenotype such as appearance of novel surface antigen, alternation in physiological properties, change in colony morphology, nutritional requirements, biochemical reactions, growth characteristics, virulence and host range.

GENE TRANSFER:

1- Vertical transmission.

Genes transmitted stably from one generation to another .

An *E.coli lacz* will divide into two *E. coli lacz* mutants .

2- Horizontal transmission.

An *E.coli lacz* will can be converted into lac + by receiving a gene from a wild type *E. coli* .

Occurs by three mechanisms :

1- TRANSFORMATION:

Transformation is gene transfer resulting from the uptake by a recipient cell of naked DNA from a donor cell. Certain bacteria (e.g. Bacillus, Haemophilus, Neisseria, Pneumococcus) can take up DNA from the environment and the DNA that is taken up can be incorporated into the recipient's chromosome

2- CONJUGATION:

In 1946 discovered that some bacteria can transfer genetic information to other bacteria through a process known as conjugation. Bacterial conjugation is the transfer of DNA from a living donor bacterium to a recipient bacterium. Plasmids are small autonomously replicating circular pieces of double-stranded circular DNA. Conjugation involves the transfer of plasmids from donor bacterium to recipient bacterium. Plasmid transfer in Gram-negative bacteria occurs only between strains of the same species or closely related species. Some plasmids are designated as F factor (F plasmid, fertility factor or sex factor) because they carry genes that mediate their own transfer.

3- TRANSDUCTION:

Bacteriophage are viruses that parasitize bacteria and use their machinery for their own replication. During the process of replication inside the host bacteria the bacterial chromosome or plasmid is erroneously packaged into the bacteriophage capsid. Thus newer progeny of phages may contain fragments of host chromosome along with their own DNA or entirely host chromosome.

PLASMIDS:

Plasmids are extrachromosomal elements found inside a bacterium. These are not essential for the survival of the bacterium but they confer certain extra advantages to the cell.

Number and size: A bacterium can have no plasmids at all or have many plasmids (20-30) or multiple copies of a plasmid. Usually they are closed circular molecules; however they occur as linear molecule in *Borrelia burgdorferi*. Their size can vary from 1 Kb to 400 Kb.

Multiplication: Plasmids multiply independently of the chromosome and are inherited regularly by the daughter cells.

Significance of plasmids:

1. Codes for resistance to several antibiotics. Gram-negative bacteria carry plasmids that give resistance to antibiotics such as neomycin, kanamycin, streptomycin, chloramphenicol, tetracycline, penicillins and sulfonamides.

2. Codes for the production of bacteriocines.

3. Codes for the production of toxins (such as Enterotoxins by *Escherichia coli*, *Vibrio cholerae*, exfoliative toxin by *Staphylococcus aureus* and neurotoxin of *Clostridium tetani*).

4. Codes for resistance to heavy metals (such as Hg, Ag, Cd, Pb etc.).

5. Plasmids carry virulence determinant genes. Eg, the plasmid Col V of *Escherichia coli* contains genes for iron sequestering compounds.

6. Codes resistance to uv light (DNA repair enzymes are coded in the plasmid).

7. Codes for colonization factors that is necessary for their attachment. Eg, as produced by the plasmids of *Yersinia enterocolitica*, *Shigella flexneri*, Enteroinvasive Escherichia *coli*.

8. Contains genes coding for enzymes that allow bacteria unique or unusual materials for carbon or energy sources.

Some strains are used for clearing

Recombination :

*Is the integration of the DNA that transferred from the donor cell to the recipient cell in into the chromosome ofhost cell (recipient cell).

*** There are two types of recombinations:**

1- Homologous recombinations:

is a type of genetic recombination in which nucleotide sequences are exchanged between two similar or identical molecules of DNA. It is most widely used by cells to accurately repair harmful breaks that occur on both strands of DNA, known as double-strand breaks. . Homologous recombination also produces new combinations of DNA sequences during meiosis.

2- Non homologous recombinations:

Little, if any, homology is necessary .