PASTEURELLA

Learning objectives

To know in detail about,

- Fowl cholera, Haemorrhagic septicaemia and shipping fever
- Morphology, cultural and biochemical characters of Pasteurella sp
- Difference between *P.multocida* and *P.haemolytica* (Mannheimia haemolytica)
- Antigns, toxins and pathogenesis of Pasteurella sp
- General approaches used to isolate and identify Pasterurella sp
- Leishman staining technique

SYSTEMATICS

<u>Order</u>	Pasteurellales
Family	Pasteurellaceae
Genus	Pasteurella
Genus	Mannheimia
Genus	Haemophilus
Genus	Actinobacillus
Genus	Lonepinella

HISTORY AND HABITAT

History

- The first significant report of the *P.multocida* was made by Bollinger (1878) and Perroncito (1878) and it was extensively investigated in 1880 by Pasteur as the cause of Fowl cholera.
- The *P.haemolytica* was isolated from disease of calves and sheep by Jones (1921).
- The name Pasteurellacea was proposed by Mannheim (1981).

Habitat

- *Pasteurella* organisms are world wide in distribution with a wide spectrum of hosts.
- They infect practically all domesticated and many wild animals and birds.
- They occur as commensals in the mucous membrane of the Upper respiratory and intestinal tract of animals.

MORPHOLOGY

• They are of small Gram –ve rods or coccobacilli that show*bipolar staining* particularly when fresh isolates stained with Leishman's or Wrights or Giemsa. They are non motile and non spore forming. ,

Cultural characteristics

• Growth takes place at an optimum temperature of 37°C. Growth is<u>enhanced</u> by the addition of blood or serum. On serum agar *Pasteurella* exhibit 3 different colonial forms

- Smooth form: Virulent for rabbits, growing diffusely in broth. Forming 0 smooth form. Virtuent for rabbits, growing underly in broth. Forming smooth, moderately opaque iridescent colonies on serum agar. It contains a type specific polysaccharide capsular antigen. Rough form: Completely avirulent for rabbits, giving a granular deposit in broth and forming translucent bluish colonies. It has neither a capsular nor a
- 0 mucoid antigen.
- Mucoid form: Intermediate virulence. It is rich in hyaluronic acid. 0

- The routine medium for isolation of *Pasteurella* species is Brain heart Infusion agar and Ox or Sheep blood agar.
- *P. multocida* has a characteristic sweetish odour, is non-haemolytic, and does not grow on MacConkey agar
- *Mannheimia haemolytica* and *P. trehalosi* are beta haemolytic. On sheep blood agar, colonies are surrounded by a single narrow zone of hamolysis, but in lamb blood agar a double zone haemolysis may be seen. *Mannheimia haemolytica* and *P. trehalosi* usually tolerate the bile salts in MacConkey agar to grow as pin point pink red colonies. On TSI produce Y/Y/H₂S^{-ve}.
- *P. anatipestifer* produces dewdrop colonies within 48 hrs.

BIOCHEMICAL PROPERTIES, ANTIGENS, TOXINS ANDRESISTANCE

Biocehmical properties

- They are oxidase +ve, catalase +ve and fermentative (except *P*. *trehalosi* and *R.anatipestifer*). *P.multocida* is indole positive.
- *Mannheimia haemolytica* is indole negative. They ferment several sugars (glucose, sucrose, galactose and sorbitol) with acid and no gas.
- *P.anatipestifer* is non fementative, non haemolytic, does not grow on MacConkey agar, indole –ve, urease –ve.

Antigens and toxins

- Based on capsular polysaccharide antigens, using PHA, Carter classified *P.multocida* into 5 serotypes (A-F). A type C is not a valid capsular serotype.
- The capsular types may be subdivided further into 16 somatic types based on heat stable somatic antigen, using AGPT (i.e. serologic differences in LPS). The capsular type followed by the number representing the somatic type designates a serotype
- For e.g. Type A:5, A:8, A:9 produce fowl cholera
- Type B:6 cause haemorrhagic septicaemia in Asia
- Type E:6 cause haemorrhagic septicaemia in Africa
- The toxin produced by *P.multocida* (mostly type A and D strain) is mainly cell associated, polypeptide, heat stable, dermonecrotic, lethal and immunogenic.
- In case of *Mannheimia haemolytica*/*P. trehalosi*, on the basis of soluble surface antigens so far 17 serotypes have been recognized.
- There are two distinct biotypes, A and T, based on fermentation of arabinose and trehalose respectively.
- Serotypes 3,4,10 and 15 are *P. haemolytica* biotype T *P. trehalosi*, all others are A *Mannheimia haemolytica*.
- Biotype A strains are associated with ruminant pneumonias, septicaemia in young lambs and ovine mastitis, while biotype T strains are usually associated with septicaemic infection in older lambs and sheep.
- All strains of *Mannheimia haemolytica* elaborate a soluble cytotoxin (Leukotoxin).
- It is a protein of relatively large mol.wt, thermolabile, resembles the alpha haemolysin in *E.coli* and is immunogenic.

Resistance

- They are readily killed at 55°C for 15mts or if exposed to sunlight for 3-4 hrs.
- They are also killed by 0.5% phenol in 15 mts.
- They are susceptible to penicillin but resistant to sulphonamides.

PATHOGENESIS

- The mechanism of disease production is not fully understood.
- *Pasteurella* infections result from the invasion of commensal organism during period of stress such as overcrowding, chilling, transportation or intercurrent infection. But exogenous transmission may occur by aerosal or contact exposure.
- In birds, the organisms are transmitted from nasal, oral or conjuctival secretions of infected birds, often-convalescent carriers, and by aerosal or contamination of drinking water and feed. Clinical disease is precipitated by stress such as overcrowding, laying, moulting and severe climatic change.
- Passage of infecting agent from animal to animal results in<u>enhancement</u>♂ of virulence.
- *P.multocida* is a primary cause of fowl cholera and Haemorrhagic septicaemia. However, *P.multocida* is a frequent secondary invader in pneumonic disease. When it is primary, septicaemia frequently occurs.
- Septicaemia results from penetration of the pharyngeal mucous membrane by highly pathogenic strains. They resist phagocyotsis and elaborate toxins.
- Pneumonias of cattle and sheep are the most important *Mannheimia haemolytica* infection.
- Disease usually follows within 1-2 weeks of a stressing experience such as transportation, hence the name shipping fever in cattle.
- Serotype 1 predominates in bovine pneumonia, serotype 2 in ovine disease.
- Pathogenesis is related to the adherence and colonization of the lower respiratory epithelium.
- All serotypes produce soluble leucotoxin. The toxin impairs phagocytosis and causes of lysis of cells, which allows the movement of K, Na and calcium through trans membrane gradients. This leads to lung damage.
- Lysis of platelets results in pulmonary vascular thrombosis and fibrin exudation typically associated with shipping fever pneumonia.



Diseases caused by Pasteurella

Speices	Disease
<i>P.multocida</i> (Based on Capsular polysaccharide typing)	 Fowl cholera Haemorrhagic septicaemia / Barbone (Asiaand Australia)

 Type A Type B Type D Type E Type F 	 Atrophic rhinits of pigs Haemorrhagic septicaemia (Africa) Disease in turkey
 Mannheimia haemolytica (P.haemolytica BiotypeA) Pasteurella trehalosi (P.haemolytica Biotype T) 	 Pneumonia (Shipping fever) in cattle and sheep Septicaemia in lambs
• P.anatipestifer (Rimerella anatipestifer)	• New duck disease (1-8 wk old ducklings)

PATHOGENECITY

Symptoms

Cattle

- In acute cases of haemorrhagic septicemia the symptoms include a rise in temperature, a sudden drop in milk yield, signs of abdominal pain, severe diarrhoea and dysentery.
- Respiration becomes rapid and shortly before death the mucous membranes appear cyanotic.
- In less acute cases there will be odema development in the region of the head, neck and brisket.
- The nasal discharge may be blood stained or purulent. Death occurs within 2-4 days.
- *P.haemolytica* give rise to symptoms of bronchopneumonia with pleuritis. This condition is referred as transit or shipping fever.

Sheep

• *Pasteurella* infection results in pneumonia and mastitis.

Pigs

• *Pasteurella* type D infection causes atrophic rhinitis in pigs and is a secondary invader in respiratory infections.

Poultry

- The acute form of fowl cholera can affect many species of wild and domesticated birds including chicken, turkey, ducks and geese causing high mortality.
- Death occurs suddenly without any predominant symptoms.
- In ailing birds, breathing is rapid through the open beak, the feathers are ruffled and the comb and wattles become cyanotic.
- In chronic cases swollen wattles and comb, hot and painful joints are characteristic.

Lesions

Cattle

- In acute form, the lesions include multiple haemorrhages on the serous membranesand organs together with blood stained exudates in the thorax and abdomen.
- There will be severe gastroenteritis with enlargement of mesenteric lymphnodes.
- In edematous form the striking lesion is odema occurring in the subcutaneous tissues in the region of throat, and brisket region.
- In pectoral form, the lesions are confined to the lungs and pleural cavity with enlargement of bronchial and mediastinal lymph glands.

Sheep and goat

• In acute cases there may be exudates in the pleural cavity and pericardial sac.

Poultry

- The liver and spleen are enlarged, hyperaemic with necrotic foci.
- There will be peritonitis, fluid in the pericardial sac and petechial haemorrhages on the serous surfaces.
- In chronic infections there will be swelling of the comb and wattles and accumulation f serofibrinous fluid in the joints.

DIAGNOSIS, CONTROL AND PREVENTION

Diagnosis

- By symptoms and lesions
- Leishman's stain from smears of heart blood, liver, spleen, lungs or exudates reveal bipolar stained organisms.
- Isolation and Identification of the organisms from heart blood and fluid exudates in blood agar and brain heart infusion agar will show dewdrop like colonies after 18hrsincubation.
- In putrified material bone marrow of long bones are useful to get pure cultures.
- Animal inoculation is done by inoculating the suspected material or bacterial culture in mice and rabbits by scarification or subcutaneous route.
- The animal dies in 24-72 hrs with haemorrhagic tracheitis.

Control and prevention

- Immunity is predominantly humoral. *Pasteurella multocida* P52 strain have proved useful. It gives protection for 6 months to 1 year.
- Vaccines for fowl cholera are not successful. So vaccines should be prepared fromtypes prevalent in that area for protection.
- *Pasteurella* is amenable to Penicillin-G, streptomycin, chloramphenicol, chlortetracycline, sulpha and tripmethoprim, enrofloxacin and oxytetracycline.
- It is one of the few Gram-negative bacteria sensitive to penicillin.