















General characters of *Pasteurella* species and *Mannheimia haemolytica*

- It is considered as normal commensal in upper respiratory tract of man, animals and birds. Under stress condition, it converted into pathogenic causing septicemia.
- ✓ They are small, Gram negative rods, characterized by <u>bipolarity</u> due to accumulation of metachromatic or volutin granules at the two poles of bacterial cell (appear in smears from infected tissues stained by Leishman's or Giemsa stains).
- Non motile, non sporulated, capsulated (<u>hyaluronic acid</u>).
 Capsule appears only in freshly prepared samples????
- ✓ Aerobic and facultative anaerobic.
- ✓ Can grow on ordinary media but they grow best on media supplemented with blood or serum.
- ✓ Some species, such as *M. haemolytica*, *P. trehalosi* and *P. aerogenes* can tolerate the bile salts in MacConkey agar.

General characters of *Pasteurella* species and *Mannheimia haemolytica*

- ✓ Ferment glucose without gas production.
- <u>Oxidase</u> and catalase test +ve and <u>nitrate</u> is usually reduced to nitrite.
- Pasteurella aerogenes, P. trehalosi, and P. pneumotropica are more closely related to members of G. Actinobacillus than they are other Pasteurella species.
 P. trehalosi is now used to denote P. haemolytica biotype
- *P. trehalosi* is now used to denote *P. haemolytica* biotype
 T isolates while *P. haemolytica* biotype A have been allocated to a new genus and renamed *Mannheimia haemolytica*.



- Gram –ve coccobacilli
- Arranged singly or in pairs rarely in chains
- Non motile
- Non sporulated
- Capsulated by hyaluronic acid capsule which rapidly disappear <u>due to hyaluronidase enzyme</u>
- It could be stained by Giemsa, Leishman's stain in blood smear or Loeffler's M.B. from animal tissue showing bipolarity

Culture characters

- Aerobic and facultative anaerobic
- Optimum temperature 37C (can grow at 37-44C)
- ✤ pH is slightly alkaline (7.2-7.4)
- On nutrient agar media: colonies are fine translucent (pin point) with semen or semineferous odour
- Colonies present in 3 forms:
- 1- Mucoid (due to capsule) or intermediate colonies
- 2- <u>Smooth</u> fluorescent colonies
- 3- Rough or blue colonies

- On broth: uniform turbidity + <u>semen odour</u>
- Blood agar: no haemolysis
- MacConkey agar: no growth (bile salt inhibitors)
- Selective media for Pasteurella as:
- Trypticase soya agar (TSA)
- DAS media

- Dextrose starch agar (DSA)
 - <u>N.B.</u> Colonies of *M. haemolytica* and *P. trehalosi* are β -<u>haemolytic</u> and <u>odourless</u>. Moreover, they <u>grow on</u> <u>MacConkey's</u> agar as pin-point, red colonies.

Biochemical reactions

- Sugar fermentation: *P. multocida* ferment most sugars except lactose and maltose are –ve
- Oxidase, catalase, Indole, Ornithine decarboxylase and Nitrate reduction tests: +ve.
- Urease: -ve.
- G. Mannheimia includes <u>trehalose-negative</u> members of the *P. multocida* complex. All strains in the genus ferment mannitol, while not ferment D-mannose is a key by which *Mannheimia* species are differentiated from members of the G. Pasteurella.

Feature	M. haemolytica	Pasteurella species	
		P. multocida	P. trehalose
Haemolysis (sheep blood	+	-	+
agar)			
Growth on MacConkey agar	+	-	+
Indole production	-	+	-
Catalase activity	+	+	-
Ornithine decarboxylase test	-	+	-
Acid production from:			
- Lactose	+		-
- D-trehalose		V	+
- Maltose	+		+

Classification

P. multocida classification



Pasturella M.O. is named by Pasteur who described it in cases of:

- Fowl cholera Avian
- Snuffles Rabbit
- Septicemia Swine, wild animals
- Pneumonia Animals

He refer them as **Bacterium bipolar multocidum** or (*P. septica*)

- Bergey's manual classification
- P. multocida P. haemolytica P. pneumotropica
- P. ureae P. gallinarum P. aerogenes
- *P. anatipestifer* (infectious serositis of duckling)

Ligniere (1900)

Classify Pasteurella according to host:

- Aviseptica ----- Fowl cholera
- Boviseptica ----> Pneumonia in cattle
- Suiseptica ---- Swine
- Equiseptica ----> Equine

Classification on the basis of <u>capsular</u> and <u>surface</u> <u>polysaccharide (somatic)</u> Ag into 5 groups (A, B, D, E, F)



Antigenic structure of Pasteurella

It is detected by both capsular and somatic polysaccharide antigens which are used to designate a specific serotype.

Robert	Classify Pasteurella according to the Serum protection test in mice. Designated by Latin numbers (I, II, III, IV).
Carter	Serogrouping on the basis of differences in capsular polysaccharides using Indirect haemagglutination test (A, B, D, E and F).
Namioka and Murata	subdivided <i>P. multocida</i> into 16 somatic types on the basis of serological differences in the cell wall LPS using HA absorption test. Designated by numbers (1, 2, 3, 4,)

Examples for antigenic formula:

- IIA5,8,9 (Fowl cholera).
- A11 (Snuffle in rabbit).

- IB6& IE6 (HS in bovine).

Biotyping of *P. multocida*

- Biotyping of *P. multocida* revealed 3 biotypes or subspecies namely:
- ✓ *P. multocida* subspecies Multocida.
- ✓ *P. multocida* subspecies Septica.
- ✓ P. multocida subspecies Gallicida.



Diseases caused by *Pasteurella* and *Mannheimia* spp.

Species	Hosts	Diseases	
P. multocida:	Cattle	-Shipping fever, pneumonia in calves and	
-Type A		mastitis.	
	Sheep	- Pneumonia, encephalitis, septicemia and	
		mastitis.	
	Horse	- Pleuropneumonia	
	Poultry	- Fowl cholera.	
	Rabbits	- Snuffles (purulent rhinitis)	
- Type B	Cattle, buffaloes	- Haemorrhagic septicaemia; barbone (Asia)	
- Type E	Cattle, buffaloes	- Haemorrhagic septicaemia; barbone (Africa)	
- Type D	Swine	Pneumonia and a tropic rhinitis	
- Type F	Turkey	May cause septicaemia	
M. haemolytica	Cattle	- Bovine pneumonic pasteurellosis	
(M. haemolytica		(shipping fever).	
biotype A)			
	Sheep	-Septicaemia (3 months of age),	
		pneumonia, gangrenous mastitis.	
<u>P. trehalosi</u>			
(M. haemolytica	Sheep	Septicaemia in lambs (5 to 12 months of	
biotype T)		age)	
P. anatipestifer	Duckilng	Infectious serositis	
ur, anmed Hussein			





Fowl cholera



Lab. Diagnosis of Pasteurella and Mannheimia Diseases

- <u>Specimen</u>: According to the <u>disease</u> and its <u>stage</u>. Samples are sent to the lab within 1-3hrs after collection..
- Blood sample from suspected animals or birds during fever or acute septicaemia.
- Living animals: Nasopharyngeal swabs or larynopharyngeal swabs or mastitic milk.
- Dead animals or birds: parts of lung showing pneumonia, trachea, heart blood, liver, spleen or bone marrow.
- Specimens should be cultured on blood and MacConkey agars. Blood agar supplemented with neomycin, bacitracin and actidione can be used for the isolation of *P. multocida* from heavily contaminated specimens.

Identification of the isolates

Morphology.

- Gram stained smear.
- characteristic bipolar organisms can be detected in blood or tissue (bone marrow, liver or spleen) smears stained with Leishman's or Giemsa stains and *P. multocida* can be isolated.
- Culture characters.
- Biochemical reactions.
- Serogrouping and serotyping.
- Experimental infection (Lab animal pathogenicity test):
- Mice, rabbit and pigeon are more susceptible while G. Pigs are resistant.
- \checkmark Chicken are susceptible to avian strains.
- Sample is injected S/C or I/P in the lab animal. Death overnight due to acute septicemia.
- ✓ N.B. Laboratory animals are resistant to experimental infection with *Mannheimia* species.
 Dr. Ahmed Hussein

