

**2nd Professional Year, Veterinary Microbiology
(Unit-I)**

Pasteurella and Mannheimia

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Characteristics

- Family *Pasteurellaceae*
- small (0.3–1.0 μm in width and 1.0–2.0 μm in length)
- facultatively anaerobic, Gram-negative coccobacilli or rods
- non-motile, non-spore-forming, fermentative
- generally oxidase-positive (except some *P. dagmatis*)
- catalase-positive (except for *P. caballi*)
- *non-haemolytic* (except for *M. haemolytica*)
- Isolates of *P. haemolytica* previously known as biotype A-*Mannheimia haemolytica*

Characteristics

- unenriched media support growth
- nutritionally fastidious
- grow best on media supplemented with serum or blood
- usually remain viable for only a few days on culture plates
- *M. haemolytica*- small, pinpoint colonies on MacConkey agar
- commensals on mucosae of upper respiratory tract of animals
- survival in environment relatively short

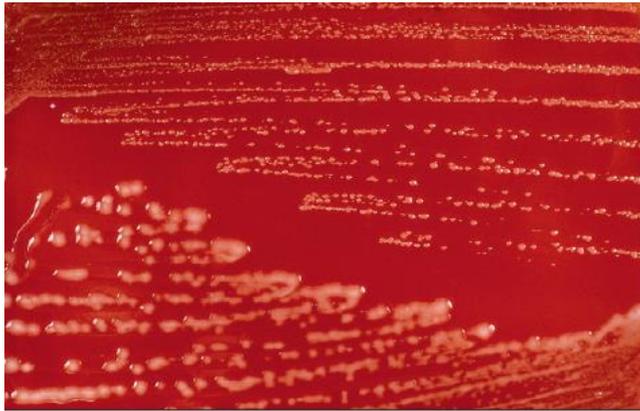


Figure 21.2 Comparison of the colonial types of *P. multocida*. The non-mucoid strain (top) of low virulence was isolated from a dog, while the mucoid colonies (bottom) are those of a virulent type A strain from a pig.

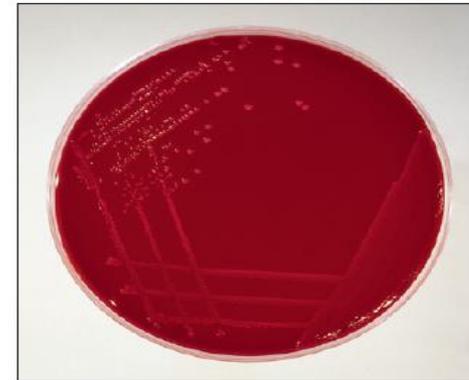


Figure 21.4 *Pasteurella multocida* on sheep blood agar. The colonies are non-haemolytic and have a characteristic sweetish odour.

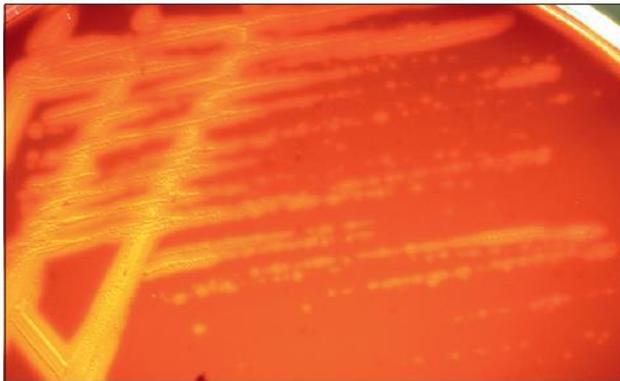


Figure 21.6 *Mannheimia haemolytica* on sheep blood agar, isolated from the pneumonic lung of a lamb, showing small colonies surrounded by a narrow zone of beta-haemolysis.



Figure 21.7 *Mannheimia haemolytica* on MacConkey agar: the small, red, pinpoint colonies indicate a tolerance of the bile salts in the medium.

(Markey *et al.*, 2013)

Bipolar staining of *Pasteurella* species

Bacteria in Giemsa-stained

blood smears from lesions have this characteristic staining pattern

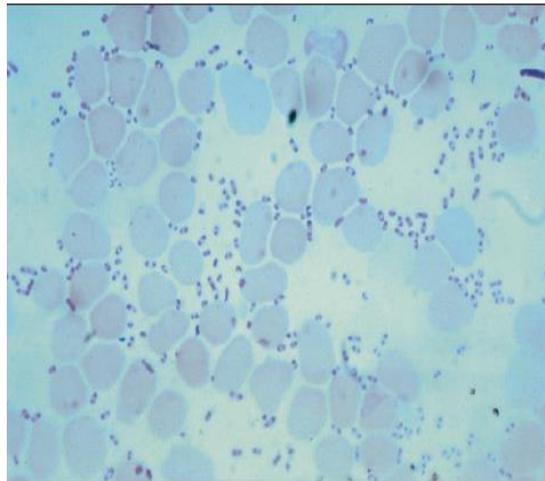


Figure 21.1 *Pasteurella multocida* in a bovine blood smear from a case of haemorrhagic septicaemia showing the characteristic bipolar staining. (Leishman stain, $\times 1000$)

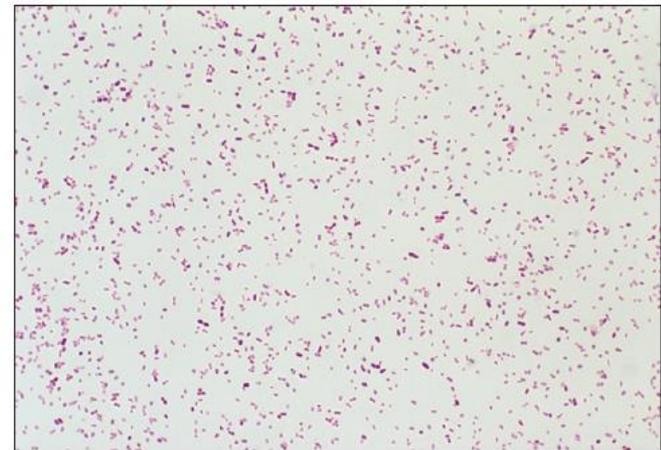


Figure 21.8 Gram-stained smear from a culture of *P. multocida*. Small Gram-negative rods with a tendency towards coccobacillary forms. ($\times 1000$)

(Markey *et al.*, 2013)

Differentiation

- Pasteurellae and *Mannheimia* species- colonial and growth characteristics, by biochemical reactions
- Strains of *P. multocida* - serotyping and biotyping

Capsule

- Types A and B - composed of hyaluronic acid
- Type D contains heparin
- Type F contains chondroitin

Colonial characteristics:

- *P. multocida*- round, greyish, shiny and non-haemolytic
- Colonies of some pathogenic strains mucoid due to the production of thick hyaluronic acid capsules
- The colonies have a subtle but characteristic sweetish odour
- *M. haemolytica*, *M. granulomatis* – haemolytic and odourless
- other *Pasteurella species* - round, greyish and mostly nonhaemolytic
- Most pathogenic *Pasteurella species* do not grow on MacConkey agar

Serotyping of *Pasteurella*

- On the basis of differences in capsular polysaccharides (Carter, 1955)
- Five types or serogroups of *P. multocida*- A, B, D, E and F, Using a passive hemagglutination test
- Lipopolysaccharide (LPS)- two main somatic typing systems reported
- The Namioka system - based on a tube agglutination test and able to recognize 11 serotypes (Namioka and Murata, 1961)
- Heddleston system- uses a gel diffusion precipitation test and can recognize 16 serotypes; currently the preferred method
- Current classification of *P. multocida* strains combines capsular typing with Heddleston somatic typing
- Strains- first letter indicates the capsular group and the number designates the Heddleston LPS serotype

- *M. haemolytica* – serotypes A1, A2, A5, A6, A7, A8, A9, A12, A13, A14, A16, and A17
- serotype A11 -renamed *M. glucosida*
- *P. multocida* has three subspecies - differentiated by minor differences in their fermentation of carbohydrates
- *Pasteurella multocida* subsp. *Multocida*- cause significant disease in domestic animals
- *Pasteurella multocida* subsp. *Septica*- from various sources including dogs, cats, birds and man
- *Pasteurella multocida* subsp. *gallicida* strains are recovered from birds and may occasionally cause fowl cholera

Diseases:

Bacterial species	Hosts	Disease conditions
<i>P. multocida</i>		
type A	Cattle	Associated with bovine pneumonic pasteurellosis (shipping fever); associated with enzootic pneumonia complex of calves; mastitis (rare)
	Sheep	Pneumonia, mastitis
	Pigs	Pneumonia, atrophic rhinitis
	Poultry	Fowl cholera
	Rabbits	Snuffles
	Other animal species	Pneumonia following stress
type B	Cattle, buffaloes	Haemorrhagic septicaemia (Asia)
type D	Pigs	Atrophic rhinitis, pneumonia
type E	Cattle, buffaloes	Haemorrhagic septicaemia (Africa)
type F	Poultry, especially turkeys	Fowl cholera
	Calves	Rare cases of peritonitis
<i>M. haemolytica</i>	Cattle	Bovine pneumonic pasteurellosis (shipping fever)

(Quinn et al.,2011)

Differentiation of the main pathogenic *Pasteurella* and *Mannheimia* species

Feature	<i>M. haemolytica</i>	
	<i>P. multocida</i>	
Haemolysis on sheep blood agar	+	-
Growth on MacConkey agar	+	-
Distinctive odour from colonies	-	+
Indole production	-	+
Catalase activity	+	+
Urease activity	-	-
Ornithine decarboxylase activity	-	+
Acid from:		
lactose	+	-
sucrose	+	+

(Markey *et al.*, 2013)

Pathogenesis and pathogenicity

- Many *P. multocida* infections- endogenous
- commensals of upper respiratory tract, may invade the tissues of immunosuppressed animals
- Exogenous transmission- by direct contact or through aerosols
- Adhesion to mucosa and avoidance of phagocytosis
- Capsule- antiphagocytic, may also function in adhesion in some strains
- Virulence factor of *P. multocida* strains of type A, D- PMT toxin, a cytotoxic protein

Haemorrhagic septicaemia

- Haemorrhagic septicaemia or barbone- acute, potentially fatal septicaemia
- mainly affecting buffaloes and cattle
- Predisposing factors such as overwork, poor body condition and monsoon rains
- *Pasteurella multocida* serotype B:2 causes the disease in Asia, the Middle East and some southern European countries
- Serotype E:2 - in Africa
- The Namioka-Carter and the Carter-Heddleston systems- 6:B or B:2 (Asian serotype) respectively
6:E or E:2 (African serotype) respectively
- Serotypes B and E - only *P. multocida* serotypes with hyaluronidase activity

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- Buffaloes tend to be more susceptible to the disease than cattle
 - All ages can be affected but in endemic areas
 - the disease most common in animals between 6 and 24 months of age
 - Many older animals -latent carriers, with pasteurellae located in the tonsillar crypts
 - Periodically, these animals shed *P. multocida* in nasal secretions and in aerosols
 - Explosive outbreaks- active carrier introduced into a stressed, susceptible population

Clinical signs

- The incubation period - 2 to 4 days
- course ranges from 2 to 5 days
- Death, without prior signs of illness, may occur within 24 hours of infection
- Sudden onset of high fever, respiratory distress and a characteristic oedema of the laryngeal region are features of the disease
- The oedema may extend to the throat and parotid regions and to the brisket
- Recumbency followed by death from endotoxaemia
- Mortality rates are usually over 50% and can approach 100%

Diagnosis

- History - acute disease with high mortality in endemic areas
- Lesions-
 - widespread petechial haemorrhages
 - enlarged haemorrhagic lymph nodes
 - blood-tinged fluid in the pleural cavity and pericardial sac
- Giemsa-stained blood smears from a recently dead animal often reveal large numbers of bipolar-staining organisms
- Isolation, identification and serotyping the *P. multocida* isolate
- PCR

Treatment and control

- Antibiotic therapy in the early febrile stage- usually effective
- organism susceptible to penicillin, tetracyclines more often used
- A slaughter policy for affected and in-contact animals in countries where the disease is exotic
- Vaccines - bacterins and a live vaccine, Modified live deletion mutant vaccines

Bovine respiratory disease

- Both shipping fever and enzootic pneumonia of calves
- infection by *M. haemolytica* and *P. multocida*. either separately or jointly
- Shipping fever, characterized by severe bronchopneumonia and pleurisy
- most commonly in young cattle within weeks of being subjected to severe stress, such as transportation, assembly in feedlots and close confinement
- condition is commonly associated with *M. haemolytica*, although *P. multocida* also isolated from lungs of affected cattle

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- The principal serotype of *M. haemolytica* associated with disease is A1
 - The principal serotype of *P. multocida* isolated from cases of respiratory disease in cattle is A3
 - Several respiratory viruses including parainfluenzavirus 3, bovine herpesvirus 1 and bovine respiratory syncytial virus may predispose to the bacterial invasion

Clinical signs

- Clinical features of shipping fever include sudden onset of fever, depression, anorexia, tachypnoea and serous nasal discharge
- In mixed infections, usually a marked cough and ocular discharge
- Morbidity rates can reach 50% and mortality rates range from 1 to 10%
- Enzootic calf pneumonia- occurring in housed calves from 2 to 6 months of age with morbidity rates of up to 30% and mortality rates of between 5 and 10%

Diagnosis

- There may be a history of exposure to stress factors and of sudden onset of respiratory disease
- Gross pathological findings - cranial lobes of lungs are red, swollen and consolidated
- Cytospin preparations from bronchoalveolar lavage usually reveal large numbers of neutrophils
- Isolation of *M. haemolytica*, often in association with other pathogens, from transtracheal wash samples or affected lung tissue- confirmatory

Treatment and control

- Affected animals must be isolated and treated early in the course of disease
- Treatment with oxytetracycline, potentiated sulphonamides and ampicillin usually effective
- Stress factors must be kept to a minimum
- Vaccination

Pasteurellosis in sheep

- Outbreaks of ovine pneumonic pasteurellosis - usually caused by *M. haemolytica*
- *P. multocida* -sporadic cases
- *Mannheimia haemolytica* - commensal of the upper respiratory tract
- Predisposing Factors- adverse climatic conditions or concurrent infections with respiratory viruses such as parainfluenzavirus 3
- Flock outbreaks usually start with sudden deaths of some sheep and acute respiratory distress in others

- Post-mortem findings - ventral consolidation in the cranial lobes of the lungs and fibrinous pleural and pericardial effusions
- Laboratory confirmation - isolation of a heavy growth of *M. haemolytica* from lung lesions
- Long-acting oxytetracycline- usually effective for treatment
- Multivalent bacterins are available and may be of value in control programmes
- Septicaemic pasteurellosis in lambs less than 3 months of age- *M. haemolytica*
- *Mannheimia haemolytica*- can cause a severe necrotizing mastitis in ewes
- Both *P. multocida* and *M. haemolytica* have been isolated occasionally from cases of bovine mastitis

Atrophic rhinitis of pigs

- Toxigenic strains of *P. multocida* type D or A severe, progressive form of atrophic rhinitis
- These toxigenic *P. multocida* isolates - AR+ (atrophic rhinitis-positive) strains
- Infection with *Bordetella bronchiseptica* - mild, nonprogressive turbinate atrophy without significant distortion of the snout
- Presence of this organism predisposes to infection with *P. multocida* AR+
- Other factors - overstocking and poor ventilation
- *Bordetella bronchiseptica* and non-toxigenic strains of *P. multocida* widely distributed in pig herds
- The introduction of a *P. multocida* AR+ carrier may initiate an outbreak of progressive atrophic rhinitis in a susceptible herd
- young pigs - particularly vulnerable to infection, non-immune pigs of any age can be infected by these toxigenic strains

Clinical signs

- Early signs, usually encountered in pigs between 3 and 8 weeks of age
- include excessive lacrimation, sneezing and, occasionally, epistaxis
- snout gradually becomes shortened and wrinkled
- As the disease progresses, a distinct lateral deviation of the snout may develop
- Atrophic rhinitis rarely fatal
- Affected pigs are usually underweight
- damage to the turbinate bones may predispose to secondary bacterial infections of the lower respiratory tract

Diagnosis

- In severely affected pigs, characteristic facial deformities diagnostic
- Visual assessment of the extent of turbinate atrophy can be made following
- slaughter by transverse section of snouts between the first and second premolar teeth
- Isolation and identification of *P. multocida* , followed by tests to confirm that the isolate is a toxigenic strain
- PCR

Control

- Chemoprophylaxis with sulphonamides, trimethoprim, tylosin or tetracyclines in weaner, grower and sow rations could be considered
- Improvement in husbandry and management
- Vaccination with a combined *B. bronchiseptica* bacterin and *P. multocida* toxoid
 - may reduce the severity of the disease and improve growth rates

Fowl cholera

- Fowl cholera is a primary avian pasteurellosis caused by *P. multocida capsular* types A and F
- highly contagious and affects both domestic and wild birds
- Usually as an acute septicaemia, often fatal
- Turkeys tend to be more susceptible than chickens
- Post-mortem lesions include haemorrhages on serous surfaces and accumulation of fluid in body cavities
- In sporadic chronic cases- signs and lesions often related to localized infections
- The wattles, sternal bursae and joints - usually swollen due to the accumulation of fibri-nopurulent exudates

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- In the acute septicaemic form of the disease, numerous characteristic bipolar staining
 - organisms can be detected in blood smears
 - *P. multocida* can be isolated from blood, bone marrow, liver or spleen
 - The bacterium may be difficult to isolate from chronic lesions
 - Medication of the feed or water early in an outbreak of acute disease may decrease the mortality rate
 - Polyvalent adjuvant bacterins are widely used and usually contain the most commonly isolated serotypes, 1, 3 and 4
 - Modified live vaccines

Snuffles in rabbits

- Common, recurring, purulent rhinitis in rabbits - type A strains of *P. multocida*
- Stress factors such as overcrowding, chilling, transportation, concurrent infections and poor ventilation, resulting in high levels of atmospheric ammonia
- Purulent nasal discharge - cakes on the fore legs because affected rabbits paw their noses
- Sneezing and coughing may be observed
- Sequelae - conjunctivitis, otitis media, subcutaneous abscessation
- Bronchopneumonia may develop in young rabbits
- Treatment or prophylactic therapy with antibiotics