



PASTEURELLA

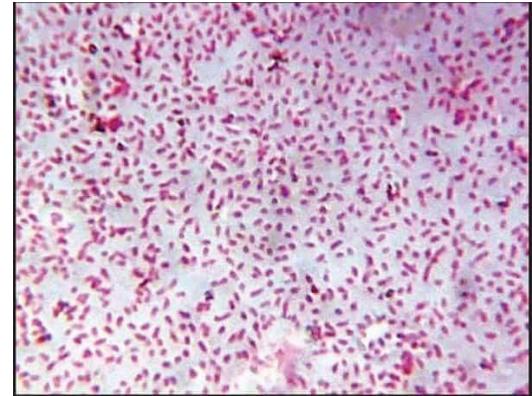
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PASTEURELLA

- *Pasteurella multocida* subsp. *multocida*
- The species *Pasteurella multocida* is Gram- negative bacteria that are inhabitants of the upper respiratory tract of many vertebrate hosts including birds, cattle, swine, cats, dogs and rodents.
- Members of this species are responsible for a number of infections that normally are secondary to colonization of the upper respiratory tract
- **Avian cholera/ Fowl cholera** (in waterfowl, chickens and turkeys),
- Respiratory disease and **hemorrhagic septicemia** in ruminants (cattle, sheep, goats and buffalo)
- **Atrophic rhinitis** in pigs
- **Snuffles/septicemia** in rodents (mice & rabbits).
- *P. multocida* is also a rare cause of infection in humans that is normally associated with dog or cat bites or scratches.

MORPHOLOGY

- Members of the Genus *Pasteurella* are Gram-negative cocco-bacilli.
- They are non-motile and non-sporulating organisms.
- They possess a capsule that is composed of **polysaccharide and hyaluronic acid**.
- They are facultative anaerobic organisms, fermentative and **oxidase positive**.
- These organisms are known for **their bi-polar staining property** in tissue smears stained by Leishman's stain or methylene blue



CLASSIFICATION

- The latest classification of *Pasteurella* is based on DNA analysis. Based on this method, the important species of veterinary importance and the diseases produced by them are listed as follows;
 - ***P.multocida* subsp *multocida*** – HS in cattle (type B2 and E2)
 - ***P.multocida* subsp *septica*** - Wound contamination
 - ***P.multocida* subsp *gallicida*** - fowl cholera (type A)
 - ***P.gallinarum*** - respiratory tract infection
 - ***P.canis*** - dog bit wound infections
 - ***P.avium* - *Haemophilus avium***

RESISTANCE

- They are not very resistant organisms
- They are killed by chemical and agents like 0.5% phenol when exposed for 15 minutes
- They are also killed on exposure to sunlight for 3-4 hours and heating at 55°C for 15 minutes.



ANTIGENS AND TOXINS

- Based on differences in capsular substances, *P. multocida* classified into A, B, D, E and F
(carter system of classification)
- These types are further subdivided into 16 subtypes based on differences in somatic antigens (Robertson system of classification)
- These subtypes are assigned numerals
- Thus a serotype is designated by an alphabet followed by a number eg. **Serotype B2 causes HS.**
- A toxin in addition to endotoxin (LPS) is produced by B serotype



CULTURAL CHARACTERISTICS

- They are aerobic or facultative anaerobic organisms and grow at a temperature of 37°C.
- In ordinary media (nutrient agar), three types of colonies – mucoid, smooth (iridescent) and non-iridescent colonies are produced after overnight incubation
- The mucoid colonies are round, flat, mucoid and sticky of 2-3 mm in diameter. Organisms with abundant capsular material produce these types of colonies.
- Organisms with less capsular material produce the iridescent colonies.
- Whereas rough colonies are produced by organisms that have no capsular material
- **Blood agar** is preferred medium for growth
- There is darkening of the medium with no haemolysis and a specific odour (**tender coconut smell**).
- In broth cultures they produce uniform turbidity.



BIOCHEMICAL CHARACTERS

- **Biochemical characters:** HS organisms produce oxidase, catalase and indole, and will reduce nitrates.



HAEMORRHAGIC SEPTICAEMIA

- Haemorrhagic septicemia (HS) is an acute pasteurellosis, caused by particular serotypes of *Pasteurella multocida* and manifested by an acute and **highly fatal septicemia** principally in cattle and water buffaloes the latter are thought to be more susceptible than cattle
- HS occurs infrequently in swine and even less commonly in sheep and goats.
- It has been reported in bison, camels, elephants, horses, and donkeys, and there is evidence of its occurrence in yak.
- Presently, two serotypes are recognized – the Asian serotype and the African serotype
- The Asian strains belong to capsular type B only while the African strains are types B and E.
- Epidemic HS is caused by one of two serotypes of *P multocida*, designated B:2 in Asia and E:2 in Africa



PATHOGENESIS

- Infection is exogenous and animals acquire the infection by contact, inhalation or ingestion
- It is hypothesized that animals become susceptible as a result of various stresses, eg. the inanition seen in cattle and water buffalo at the beginning of the rainy season.
- The heaviest losses occur during the monsoon rains in south east Asia, and it is thought that the organisms, which can survive for hours and probably days in the moist soil and water, are transmitted widely at this time.
- Biting arthropods rarely transmits infection.
- *Pasteurella* organisms are found in the upper respiratory and digestive tract of animals and birds as commensals.



PATHOGENESIS CONTINUES....

- Most of the animals carry these organisms with them without exhibiting any symptoms. The organisms produce infection when they are exposed to stress conditions like extreme weather conditions, transport, immunosuppression etc.
- During these conditions the organisms rapidly multiply and excreted via droplets and through digestive tract. The droplets with organisms are the main source of infection for other animals.
- In *Pasteurella* infection, the morbidity is always higher. *P. multocida* is also a secondary invader in respiratory tract infection. The important virulence factor is the endotoxin (LPS). Besides this a thermostable, cell associated toxin that is released by dying cells also plays a role in pathogenesis.

SYMPTOMS

- The clinical syndrome of HS consists of an initial phase of temperature elevation (often unnoticed),
- A phase of respiratory involvement, and a terminal phase of septicaemia and recumbency leading to death
- The incubation period is usually 1–3 days, and the course of the disease may range from sudden death, with no observable clinical signs, to a protracted course extending up to 5 days
- Buffaloes are generally believed to be more susceptible to HS than cattle, and in this species, the disease course is shorter.
- In endemic areas, most deaths are confined to older calves and young adults.
- In nonendemic areas, massive epizootics may occur.
- Case fatality approaches 100% if treatment is not carried out sufficiently early (in the pyrexic stage).



- Three forms of HS are noticed among animals – acute, sub-acute (oedematous form) and sub acute (pectoral form).
- The important symptoms in acute cases are rise in body temperature, drop in milk yield, abdominal pain, severe diarrhoea and dysentery.
- The respiration becomes rapid before death and the mucous membrane becomes cyanotic.
- Less acute cases are characterised by rise in body temperature and oedema in the head, neck and brisket region.
- A purulent, blood stained nasal discharge is also noticed.
- Occasional cases linger for several days. Recovery is rare
- There appears to be no chronic form.

LESIONS

- The most obvious changes in affected animals are the edema, widely distributed hemorrhages, and general hyperemia.
- In most cases, there is an edematous swelling of the head, neck, and brisket region.
- Incision of the swellings reveals a clear or straw-colored serous fluid.
- The edema is also found in the musculature, and the subserous petechial hemorrhages, which are found throughout the animal, are particularly characteristic.
- Blood-tinged fluid is often found in the pericardial sac and in the thoracic and abdominal cavities.
- The lesions are also coupled with gastroenteritis, marbled lungs, blood stained stools and enlarged and haemorrhagic mesenteric lymph nodes.
- Petechial hemorrhages are seen in the pharyngeal and cervical lymph nodes.
- Gastroenteritis is seen only occasionally and, unlike pneumonic pasteurellosis, pneumonia usually is not extensive.



DIAGNOSIS

- **Based on symptoms and lesions.**
 - **Microscopical examination:**
 - The septicaemia in HS occurs at the terminal stage of the disease.
 - Therefore, blood samples taken from sick animals before death may not always contain *P. multocida* organisms.
 - Also, they are not consistently present in the nasal secretions of sick animals.
 - A blood sample or swab collected from the heart within few hours after death is most ideal.
 - Blood smears from affected animals are stained with Gram, Leishman's or methylene blue stains.
 - The organisms appear as Gram-negative, bipolar-staining short bacilli.
 - No conclusive diagnosis can be made on the basis of direct microscopic examinations alone.



- **Isolation and identification:** Blood in transport medium is the most ideal material for isolation. If the animal has been dead for a long time, a long bone, free of tissue, can also be taken.
- The most ideal medium for isolation of Pasteurella is casein/sucrose/yeast (CSY) agar containing 5% blood (calf blood).
- Freshly isolated *P. multocida* forms smooth, greyish glistening translucent colonies, approximately 1 mm in diameter, on blood agar after 24 hours' incubation at 37°C. Colonies grown on CSY agar are larger.
- Old cultures, particularly those grown on media devoid of blood, may produce smaller colonies.

- **Test to confirm the production of hyaluronidase:**
- HS-causing strains of *P. multocida* has the ability to produce the enzyme hyaluronidase.
- A hyaluronic-acid-producing culture is streaked across the centre of a dextrose starch agar plate.
- The pasteurella culture to be tested for hyaluronidase production is streaked at right angles.
- The plates are incubated at 37°C for 18 hours.
- At the point of intersection, the mucoid growth of the hyaluronic acid producer will diminish into a thin line of growth, indicating the production of hyaluronidase by the test culture.

- **Immunological methods:** Several immunological tests are used for the identification of the HS-causing serotypes of *P. multocida*.
- These consist of a rapid slide agglutination test, an IHA test for capsular typing, the AGID test, and the counter immunoelectrophoresis test (CIEP).



- **Nucleic acid recognition methods:** PCR amplification of specific DNA sequences allows rapid detection and presumptive identification of organisms directly from either clinical specimens or from small amounts of mixed or pure bacterial cultures. Some of the common methods performed to identify *Pasteurella* are as below
 - *Pasteurella-multocida*-specific PCR assay
 - *Pasteurella multocida* multiplex capsular PCR typing system
 - HS-causing type-B-specific PCR assay
 - Genotypic differentiation of isolates
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- **Serological methods:** Serological tests for detecting antibodies are not normally used for diagnosis.
- **Animal Inoculation:** The mouse usually serves as a biological ‘screen’ for extraneous organisms.
- A small volume (0.2 ml) of eluted blood swabs or a portion of bone marrow in saline is inoculated subcutaneously or intramuscularly into mice.
- If viable *P. multocida* is present, the mice die 24–36 hours following inoculation, and a pure growth of *P. multocida* can be seen in blood smears

- **Treatment:** Various sulfonamides, tetracyclines, penicillin, and chloramphenicol (are effective if administered early).
- Because of the rapid course of the disease and the frequent difficulty of access to animals, antimicrobial therapy often is not practicable.
- **Control:** The principal means of prevention is by vaccination.
- Three kinds of vaccine are widely used: plain bacterin, alum-type precipitated bacterin, and oil-adjuvant bacterin. The most effective bacterin is the oil-adjuvant— one dose provides protection for 9-12 months; it should be administered annually.
- The alum-precipitated-type bacterin is given at 6-months intervals.
- A live vaccine prepared from a B:3,4 serotype of deer origin is being used with reported success in southeast Asia.

FOWL CHOLERA (AVIAN CHOLERA)

- Fowl cholera (avian pasteurellosis) is a commonly occurring avian disease that can affect all types of birds and is often fatal.
- It is contagious and usually occurs as a septicemia of sudden onset with high morbidity and mortality, but chronic and asymptomatic infections also occur.



- Turkeys are more susceptible than chickens, older chickens are more susceptible than young ones, and some breeds of chickens are more susceptible than others.
- Chronically infected birds are considered to be a major source of infection.
- Dissemination of *P multocida* within a flock is primarily by excretions from mouth, nose, and conjunctiva of diseased birds that contaminate their environment.



SYMPTOMS

- These vary greatly depending on the course of disease.
- In acute fowl cholera, dead birds are usually the first indication of disease.
- Fever, depression, anorexia, mucoid discharge from the mouth, ruffled feathers, diarrhea, and increased respiratory rate are usually seen.



LESIONS

- Many of the lesions are related to vascular disturbances
- Hyperemia is especially evident in the vessels of the abdominal viscera.
- Petechial and ecchymotic hemorrhages are common, particularly in subepicardial and subserosal locations.
- Increased amounts of peritoneal and pericardial fluids are frequently seen.
- Livers may be swollen and often develop multiple, small, necrotic foci.
- Pneumonia is particularly common in turkeys.
- In **chronic fowl cholera**, signs and lesions are generally related to localized infections. Sternal bursae, wattles, joints, tendon sheaths, and footpads are often swollen because of accumulated fibrinosuppurative exudate.
- There may be exudative conjunctivitis and pharyngitis.
- Torticollis may result when the meninges, middle ear, or cranial bones are infected.

DIAGNOSIS

Based on the symptoms and lesions

- **Microscopical examination:** . The cells are coccobacillary or short rod-shaped, usually 0.2–0.4 by 0.6–2.5 µm in size, stain Gram negative, and generally occur singly or in pairs. Recently isolated organisms or those found in tissue smears show bipolar staining with Wright or Giemsa stains or methylene blue, and are usually encapsulated.
- **Isolation and identification:** Isolation of the organism from visceral organs, such as liver, bone marrow, spleen, or heart blood of birds that succumb to the acute form of the disease, and from exudative lesions of birds with the chronic form of the disease, is generally easily accomplished.
- *Pasteurella multocida* is a facultative anaerobic bacterium that grows best at 35–37°C.
- Primary isolation is usually accomplished using media such as blood agar, trypticase–soy agar or dextrose starch agar, and isolation may be improved by supplementing these media with 5% heat-inactivated serum.

- **Nucleic acid identification methods:** The most ideal method is the DNA fingerprinting of *P. multocida* by restriction endonuclease analysis (REA).
- **Serological tests:** Serological tests for the presence of specific antibodies are not used for diagnosis of fowl cholera.

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- **Treatment:** Sulfonamides and antibiotics are commonly used; early treatment and adequate dosages are important.
- High levels of tetracycline antibiotics in the feed (0.04%) or administered parenterally may be useful.
- **Control:** Good management practices are essential to prevention.
- Adjuvant bacterins are widely used and generally effective
- Autogenous bacterins are recommended when polyvalent bacterins are found to be ineffective
- Attenuated vaccines are available for administration in drinking water to turkeys and by wing-web inoculation to chickens.
- These live vaccines can effectively induce immunity against different serotypes of *P multocida*.
- They are recommended for use in healthy flocks only.

OTHER IMORTANT PASTEURELLA INFECTIONS

- - **Rabbits:** Peracute infection is common in rabbits. In chronic infection coryza like respiratory symptoms are common.
 - **Sheep:** Mastitis (blue bag) and pneumonia
 - **Dogs and cats:** Septic contamination of wounds
 - **Pigs:** Respiratory infection and rhinitis.

ATROPHIC RHINITIS

- An infectious disease of swine characterised by purulent nasal discharge, shortening or twisting of the snout, atrophy of the turbinate (conchal) bones and reduced productivity.
- It may occur enzootically or more sporadically, depending on a variety of factors including herd immunity.
- The most severe progressive form is caused by infection with toxigenic strains of *Pasteurella multocida* type D alone or in combination with *Bordetella bronchiseptica*.



- ***Mannheimia haemolytica* (*P. heamolytica*):**
- Unlike *P. multocida*, it is beta haemolytic.
- Causes transport or **shipping fever** in cattle.
- It is a pneumonic condition with rise in temperature, rapid respiration followed by death with 12-48 hours.
- In less acute cases coughing, debility and death are noticed.
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RIEMERALLA ANATIPESTIFER **(*P.ANATIPESTIFER*):**

- It is a non-fermenting bacterium.
- It causes infectious serositis (New duck disease) in ducklings.
- Anatipestifer infection causes high mortality, weight loss and condemnation.
- In the acute form, listlessness, eye discharge and diarrhea are commonly seen.
- Ducks show incoordination, shaking of the head and twisted neck. Birds are commonly found on their backs, paddling their legs.
- Typical lesions found in dead birds are infected air sacs, membranes covering the heart and liver, and meningitis.
- Preventive management and vaccination are effective means of control.
- Penicillin, enrofloxacin and sulfadimethoxine-ormetoprim (0.04-0.08% in feed) are effective in reducing mortality.