BRUCELLA

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General characteristics

- **Brucella** species are small, nonmotile, Gram-negative coccobacilli.
- They are MZN-positive.
- In MZN-stained smears they characteristically appear as clusters of red coccobacilli.
- They are intracellular parasites.
- Grow poorly on ordinary media.
- They are non-hemolytic on blood agar.
Important species

- *Brucella abortus*- affects cattle
- *Brucella melitensis*- affects sheep and goats
- *Brucella suis*- affects pigs
- *Brucella ovis*- affects sheep
- *Brucella canis*- affect dogs
- *Brucella neotomae*- affect desert wood rat

- *B. abortus* is most prevalent *Brucella* spp
- *B. melitensis* is most virulent *Brucella* spp.
General characteristics

- Aerobic, capnophilic and catalase-positive.
- Except *B. ovis* and *B. neotomae*, others are oxidase-positive.
- All *Brucella* species are urease-positive except *B. ovis*.
- Growth of other *Brucella* species is enhanced in an atmosphere of CO₂.
- *B. abortus*, *B. melitensis* and *B. suis* occur in smooth forms
- *B. ovis* and *B. canis* always occur in rough forms.
Differentiation of *Brucella* species

- Two important surface antigens are *abortus* antigen A and *melitensis* antigen M.

- The proportion of A : M antigen
  
  - Brucella abortus - 20:1
  - Brucella melitensis: 1:20
  - Brucella suis - 11:9

- *B. abortus* are lysed by a specific bacteriophage (*Tbilisi phage*) at routine test dilution.

- *B. abortus* grows in presence of basic fuchsin but not in presence of thionin.

- *B. suis* grows in presence of thionin but not in presence of basic fuchsin.
Cultural requirement

- Do not grow on ordinary media.
- Addition of blood or serum supports growth.
- Trypstose agar or Liver infusion agar gives satisfactory growth.
- *B. abortus* and *B. ovis* requires 10% CO$_2$. 
Usual habitat

- Brucellae have a predilection for both female and male reproductive organs in sexually mature animals.

- Each *Brucella* species tends to infect a particular animal species.

- Infected animals serve as reservoirs of infection which often persists indefinitely.

- Organisms, shed by infected animals, can remain viable in a moist environment for many months.
Pathogenesis

- Rough colonies and are less virulent than smooth colonies.

- Organism can gain entry in the host even through the mucous membrane, abraded skin and conjunctiva.

- Inhibition of phagosome-lysosome function intracellular survival

- Superoxide dismutase and Catalase - resistance to oxidative killing

- Brucellae persist within macrophages but not within neutrophils
Pathogenesis

- Bacteraemia - spread and localization in the reproductive organs
- Associated glands in sexually mature animals
- *Erythritol* is 4 carbon sugar alcohol that stimulates growth of Brucella organism
- High concentrations - placentae of cattle, sheep, goats and pigs
- Also found in other organs - mammary gland and epididymis
- In chronic brucellosis, organisms may localize in joints or inter-vertebral discs.
Brucellosis in cattle

- Brucellosis in cattle due to *B. abortus* is called **Bangs disease**.

- Brucella organism are obligate parasite and have predilection for genital tract and joints.

- Cows may be infected by:
  - Ingestion
  - Inhalation
  - AI or natural service

- Infected cows secrete brucella organism in:
  - vaginal discharge
  - placenta
  - aborted foetus
  - milk
Brucellosis in cattle

- Usually abortion is seen in third trimester.
- “Abortion storm” is observed in herd with large number of susceptible pregnant animals.
- Cows abort only once.

Symptoms in cattle are:
- Abortion;
- Stillborn.
- Weak calf born.
- Retention of fetal membranes;
- Signs of infection in the membranes;
- In bull- *Epididymitis* is observed
Diagnostic procedures

- Specimens should be processed in a biohazard cabinet.

- The diagnosis of brucellosis depends on serological testing.
  - MRT
  - RBPT
  - STAT

- Isolation and identification of the Brucella species.
Serological test

- **Milk ring test (MRT) or Abortus Bang’s ring (ABR) test:**
  - This test is performed to identify brucella in herds.
  - It is based on detection of antibodies in milk against stained antigen

- **Rose Bengal Plate test (RBPT):** Qualitative test for screening.

- **Standard Tube Agglutination Test (STAT):** Quantitative test- Confirmatory
<table>
<thead>
<tr>
<th>Sl No</th>
<th>Name of the test</th>
<th>Antigen</th>
<th>Sample to be tested</th>
<th>Nature</th>
<th>Purpose</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>Milk ring test or Abortus bang ring test.</td>
<td>Killed brucella organism stained with _______</td>
<td>Milk</td>
<td>Qualitative</td>
<td>This test is carried out to detect brucellosis in herd.</td>
</tr>
<tr>
<td>2.</td>
<td>Rose Bengal Plate test (RBPT)</td>
<td>Killed brucella organism stained with “Rose Bengal” dye</td>
<td>Serum</td>
<td>Qualitative</td>
<td>This test is carried out in field condition to detect affected animals from MRT positive herd.</td>
</tr>
<tr>
<td>3.</td>
<td>Standard Tube agglutination test (STAT / SAT)</td>
<td>Killed Brucella organism unstained</td>
<td>Serum</td>
<td>Quantitative</td>
<td>A titre of 1:40 is considered positive whereas 1:20 is doubtful.</td>
</tr>
</tbody>
</table>
Diagnostic procedures

- **Specimen:** cotyledons, foetal abomasal contents and uterine discharges
- **MZN-stained smears** often reveal MZN-positive coccobacilli.
- In specimens containing cells, the organisms appear in clusters.
- The polymerase chain reaction can be used to detect brucellae in tissues.
- **Strauss reaction:** Development of orchitis in G. pig after intraperitoneal injection of infective material.
Treatment and control

• Treatment of cattle with brucellosis is not practical.

• Immunity in brucellosis is predominantly cell mediated.

• Three vaccines are in use
  - strain 19 (S19) (*Cotton strain 19*)
  - adjuvanted 45/20 vaccine ,and
  - RB51 vaccine
Brucella - Vaccine

• **S19 vaccine** is administered to female calves up to 5 months of age (Calf hood vaccination).

  (Vaccination of *mature animals leads to persistent antibody titres*)

• **45/20 bacterin** - Even when administered to adult animals, this vaccine does not induce persistent antibody titres.

• **RB51 strain** is a *stable, rough mutant* which induces good protection against abortion and does not result in serological responses detectable in tests.
Brucellosis in humans

- Humans are susceptible to *B. abortus*, *B. suis*, *B. melitensis* and, rarely, with *B. canis*.

- Severe infections occur with *B. melitensis* (Malta fever) and *B. suis* biotypes 1 and 2. Human infections due to *B. abortus* are moderately severe whereas those caused by *B. canis* are usually mild.

- Transmission to humans occurs through contact with secretions or excretions of infected animals.

- Routes of entry include *skin abrasions, inhalation* and *ingestion*.

- *Raw milk and dairy produce* made with unpasteurized milk are important sources of infection.
Brucellosis in humans

• Brucellosis in humans, known as undulant fever, presents as:
  - Chill
  - fluctuating pyrexia,
  - perspiration
  - malaise,
  - fatigue and
  - muscle and joint pains

• Abortion is not a feature of human infection.

• *Osteomyelitis* is the most common complication.

• Antimicrobial therapy should be administered early in an infection.
THANKS