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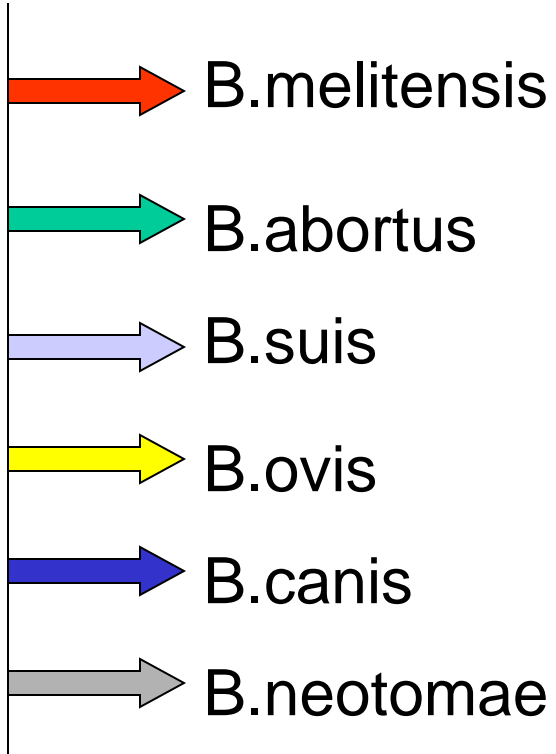
F. Brucellaceae

General characters

- ✚ G-ve rods, small in size (0.5-0.7 x 0.6-1.5 μm), that often appear cocco bacilli.
- ✚ Non motile, non sporulated and non capsulated.
- ✚ Partially acid fast as they are not decolourized by 0.5% acetic acid in modified Ziehl-Neelsen (MZN) stain.
- ✚ Catalase +ve and oxidase +ve (except *B. ovis* and *B. neotomae*)
- ✚ The growth is enhanced on enriched media

A light purple starburst shape with a black outline, containing the word "Species" in a bold, black, sans-serif font.

Species



| Species | Criteria |
|--------------|--|
| B.melitensis | <p>3 biotypes, it cause brucellosis in sheep and goat (mainly)</p> <p>Infect cattle, buffaloes, camels, swine</p> <p>Zoonotic</p> |
| B.abortus | <p>9 biotypes (differ in their biochemical and serological reactions)</p> <p>Cattle: Bang's disease or contagious abortion</p> <p>Man: Mediterranean fever, undulent fever, malta fever (Zoonotic)</p> |
| B.suis | <p>4 biotypes (pathogenic for pig)</p> <p>Highly pathogenic for man</p> |
| B.ovis | <p>Ram: ram epidmyitis</p> |
| B.canis | <p>Brucellosis in dogs (zoonotic)</p> |
| B.neotomae | <p>Isolated from wood rat</p> |

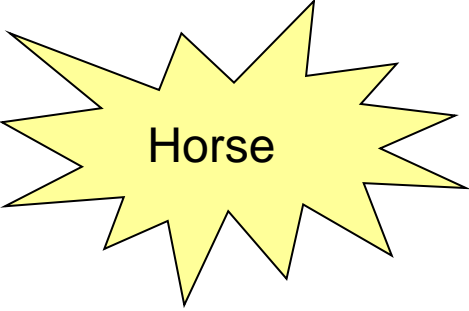
Brucellosis in Animals

1. Abortion (after 5th month) or weak neonates
2. decreased weaning weight
4. extended birthing interval
5. lower fertility
6. Decreased milk



Bull


Orchitis, epididymitis, necrosis of testicles



Horse

Fistulus withers

B.suis: abortion in mare



Sheep
goat

Abortion, retained placenta, mastitis,
lameness due to arthritis

Mode of transmission of Brucellosis in man and animals.

- ❑ Infection in animals occurs venereally (coitus or artificial insemination).

Inhalation of aerosolized Brucella .

Ingestion of contaminated food

- ❑ In man mostly occur through GIT by infected food or water (milk and meat)

or through

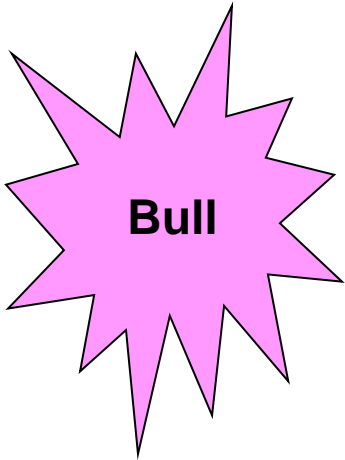
skin and mm (Abrasion or even intact)

Laboratory diagnosis of brucellosis

Samples



Aborted foeti (stomach content, fetal fluids)
Mother (vaginal mucous, milk, blood)



Testicular abscess
semen
preputal wash

Diagnosis may be:

Direct

Indirect

Direct diagnosis

- 1- Direct microscopic examination
- 2- Cultivation on specific media
- 3- Biochemical tests
- 4- agglutination with mono-specific antiserum
- 5- Pathogenicity test { Male: Strauss`s reaction
- 6- Allergic test (brucellin test)

Morphology

Gram`s stain

Gram-ve coccobacilli, arranged singly, pairs or short chain

Modified Ziehl Neelsen

As Ziehl Neelsen but 0.5% acetic acid is used as decolourizing agent

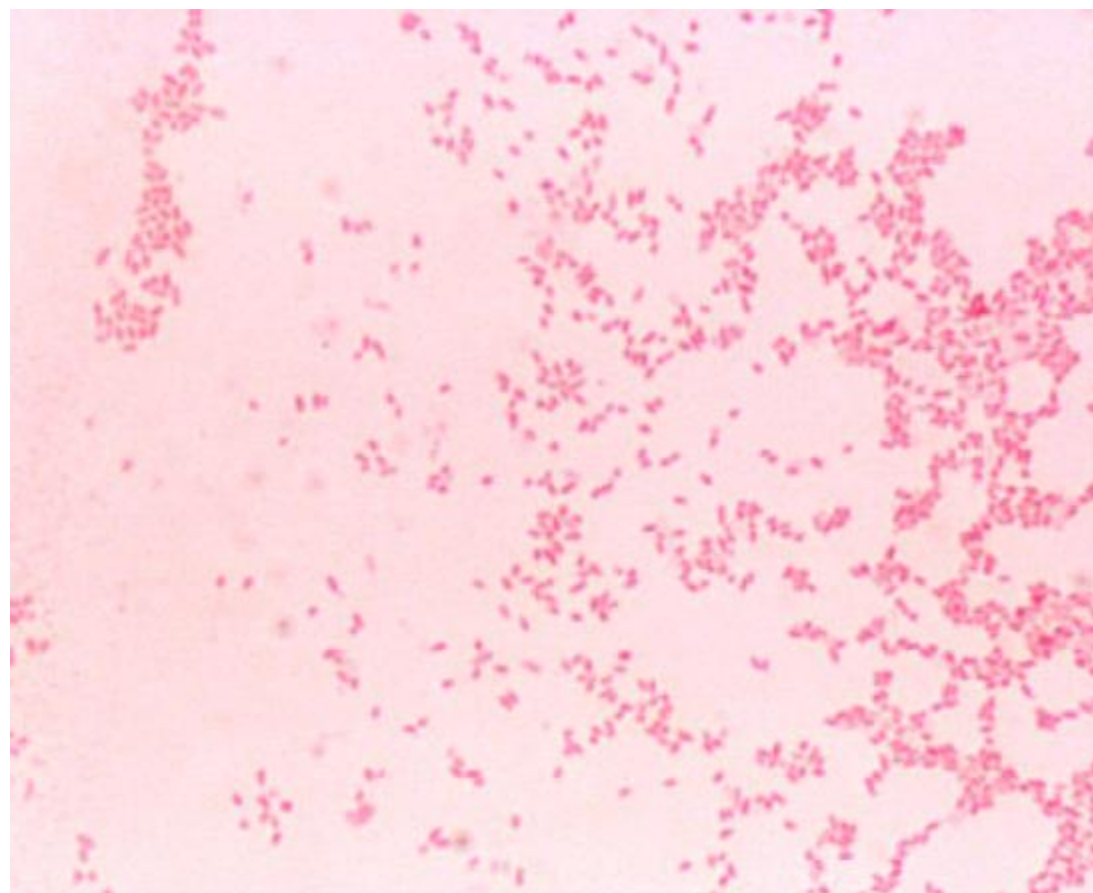
M.O. is stained red against blue back ground

Modified Koster`s

M.O. stained red against blue back ground

Han`s stain

M.O. bright blue against violet background



Culture characters

The media that used for brucella must be:

- Slightly acidic media (6.6-6.8)
- Contains enriched substance (serum, blood)
- Contains antibiotics as Polymyxin, Bacitracin, Actidione and Nystatin
- Brucella ovis and some biotypes of B. abortus media enriched with blood or serum with and incubation under 5-10% CO₂ tension for primary isolation

Media

Albimi brucella agar

Trypticase soya agar

Serum dextrose agar

BHI agar

Potato glycerol agar

❑ colonies appear small slightly convex translucent reach 1-2mm after 1-2 days.

❑ Smooth rounded delicate with entire edge and bluish tinge (sky color) by transmitted light.

changed to deep brown on aging

❑ easily transferred from smooth to rough colonies and become large, irregular,

❑ Pigmentation is better demonstrated on potato glyceol agar media.

❑ It should be incubated for at least 4 weeks before being discarded



Colonies are usually small, rounded with entire edge, translucent

Isolation of brucellae

- Isolation is the most definitive diagnosis when it is positive.
 - Failure to isolate the organism does not mean negative result.
-

Isolation failure may be due to:

- the viability and numbers of organisms in the sample
 - the nature of the sample, which is commonly contaminated.
-

Biochemical reactions

Catalase, oxidase, urease and nitrate reduction tests are positive : +ve (except *B.ovis* and *B.neotomae*)

Indole, gelatin liquefaction, citrate utilization, MR &VP tests are negative

H₂S production:

B.suis: +++ within 4 days

B.abortus, *B.melitensis*: + within 2 days

Urease activity:

B.suis: +++ (0.5-1 hour)

B.abortus, *B.melitensis*: + (1-8 hours)

| | <i>B. abortus</i> | <i>B. melitensis</i> | <i>B. suis</i> |
|--|-------------------|----------------------|----------------|
| CO2 Requiremen | + | - | - |
| H2S Production | ++ | ± | ++++ |
| Urease activity | Within 8 hs | Within 8 hs | ½-1 h |
| Inhibition/Sensitivity to dyes: | | | |
| - Thionin (20 µg/ml) | - | + (inhibit growth) | + |
| - Basic fuchsin (20 µg/ml) | + | + | - |
| - Methyl Violet (20 µg/ml) | + | + | - |
| - Pyronine (10 µg/ml) | + | + | - |
| Agglutination by monospecific antiserum | | | |
| <i>B. Abortus</i> | + | - | + |
| <i>B. melitensis</i> | - | + | - |
| <i>B. suis</i> | + | - | + |

❖ **Pathogenicity test**

Strauss reaction

❖ **Brucellin test**

Indirect diagnosis (Serological diagnosis)

1- tube agglutination test (Tube test):

Constant amount of Brucella Ag is added to a serially diluted serum samples then titre is calculated (1/40 in cattle and 1/20 in sheep).

2- Complement fixation test (Widely accepted confirmatory test).

3- Rose Bengal test (screening test)

4- Rivanol test ((screening test)

5- ELISA especially competitive ELISA because depends on mono clonal antibodies (Reliable screening and confirmatory test).

Abortus Bang ring test (ABRT) Or Milk Ring Test (MRT)

Samples: milk

1 ml milk

**+
One drop of
whole cell
antigen stained
with
tetrazolium or
haematoxyline**



one hour / 37°C



Serological Diagnosis of brucellosis

- Although the serological diagnosis is not 100% reliable when positive
- It is the main tool for the rapid recognition of infected herd and individual animals

A positive serology means:

- field strain infection
 - vaccination infection
 - residual vaccination titre
 - cross-reactivity with other organisms, like Yersinia, Salmonella, Pasteurella etc
 - human errors.
-

**CONTROL OF
BRUCELLOSIS
IN ANIMALS**

Treatment is not a practical solution.

Test and Slaughter policy.

Vaccination of calves and adults.

Good management.

VACCINATION

Killed vaccine
(Bacterin)

Living vaccine

Living virulent
(Attenuated)

Living avirulent

45/20

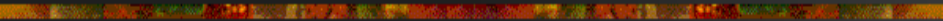
H38

S19

RB 51

B. melitensis

Rev. 1 (Rev1)

- 
- Natural infection gives life-long immunity
 - This means the best immunity is achieved by using live vaccines
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