

## ***Bordetella***

*Bordetella* species are obligately aerobic, small Gram-negative rods or coccobacilli. Most species grow on BA and MCA (not *B. pertussis*) at 37° C. Selective media are useful for isolation from contaminated sites.

Catalase positive, oxidase positive (not *B. parapertussis*).

Do not breakdown carbohydrates.

Other characteristics that are useful for the identification of *Bordetella* sp include motility and urease production.

*B. bronchiseptica* can be differentiated from other nonfermentative Gram-negative rods such as *Alcaligenes* sp using OxiFerm Tubes.

### ***Bordetella bronchiseptica***

#### **Specimen Collection**

Ideally transtracheal aspirates are collected from dogs with tracheobronchitis. Alternatively nasal or tracheal mucus can be collected using sterile, flexible stemmed, cotton-tipped swabs.

Nasal swabs are used to detect infected pigs. Swabs should be placed in a suitable transport medium for dispatch to the laboratory.

Pneumonic lung is collected at PM. Lung samples should be emulsified in tryptose phosphate broth before inoculation onto agar.

Specimens should be cultured as soon as possible after collection.

While there may be some loss of viability, short term storage at -5° C is generally satisfactory if the specimen cannot be immediately cultured

#### **Microscopic Examination of Tissues**

Smears of nasal mucosa or impression smears of pneumonic lung should be stained by Gram's method.

Little to be seen that is of diagnostic significance. Small Gram negative rods.

#### **Cultural Examination**

SBA or bovine BA and MCA plates should be inoculated and incubated in air for 18-48 hr at 37° C. MCA plates may be supplemented with 1% glucose.

*Bordetella* selective media should be used for culturing clinical specimens likely to be contaminated with other bacteria. The selective medium described by Smith and Baskerville is useful for isolating and recognising *B. bronchiseptica* and *B. avium* in contaminated specimens. The medium contains clindamycin, gentamicin, potassium tellurite, amphotericin B and bacitracin as selective agents. Some canine isolates are sensitive to gentamicin so it should be omitted when culturing specimens from canines.

Bordet-Gengou agar used for isolation of *B. pertussis* in man can also be used for primary isolation of *B. bronchiseptica*.

Grows readily on BA and MCA. Small, smooth, round, dewdrop colonies at 48 hr, which enlarge and become flatter and glistening after further incubation. May be haemolytic.

Phase modulation occurs (also in *B. avium*) and is probably due to loss of a capsule-like structure following subculture.

Phase I colonies are convex and shiny (virulent, capsulated).

Phase II colonies are larger, circular and convex with a smooth surface.

Phase III colonies are large, flat and granular with an irregular margin.

Colonies are small and pale on MCA with a pinkish hue and amber discoloration of the underlying medium.

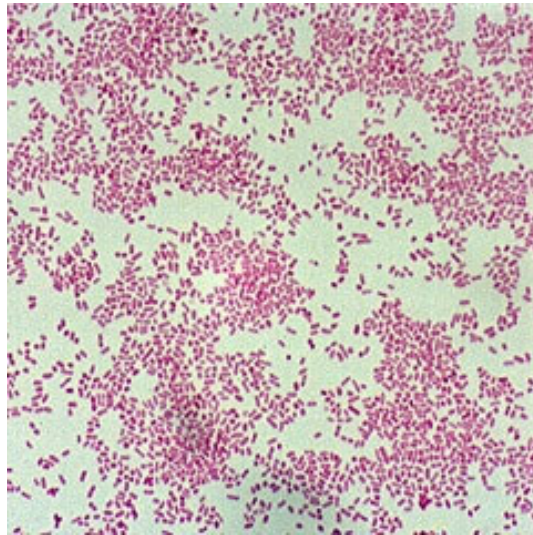
Small Gram-negative rods.

*Bordetella bronchiseptica* on SBA (48 hr culture). Oblique view x2.



Small, smooth, round, dewdrop colonies after 48 hr incubation. Colonies enlarge and become flatter and glistening after further incubation. Usually non haemolytic although some strains may produce a zone of haemolysis.

*Bordetella bronchiseptica*. Gram stained smear of culture.



*B. bronchiseptica* is a small Gram-negative rod.

## Identification

The following tests are useful —

β-haemolysis (bovine blood agar) — variable

Growth on MCA — positive (NLF)

Growth on SS agar — negative

Catalase — positive

Oxidase — positive

Motility — positive

Nitrate reduction — positive

Urease — positive

Citrate as a sole source of carbon — positive

Oxidation of xylose — negative

## Serological Tests

A serum agglutination test and ELISA can be used to measure antibody responses in pig herds.

Serum agglutinins to *B. bronchiseptica* are common in pig herds. Although serology is of some use in making a herd diagnosis, it is not routinely used because it offers no advantages over culture of nasal swabs.

## Dermonecrotic Toxin

A method for detecting the heat labile demonecrotic toxin of *B. bronchiseptica* has been described.

## Antibiotic Susceptibility

Intrinsically sensitive to a wide range of antimicrobials including benzyl penicillin, ampicillin, furazolidone, tetracyclines, , neomycin, gentamicin, , sulphonamides, trimethoprim. However, strains showing acquired (R-plasmid) resistance to one or more antimicrobials have been reported. Resistant strains are frequently encountered from pigs, so sensitivity testing should be done.