

The Microbiology Lab:

Bartonella henselae

PATH 417A

Case 1

Week 3

What are potential causes of fever and swollen lymph nodes? Provide examples of both bacterial and non-bacterial pathogens, as well as non-infectious causes

Fever

- Is a result of cytokine release from stimulation of macrophages & T-lymphocytes¹

Lymphadenopathy

- Swollen lymph nodes are a result of immune cells' migrating to a localized area¹
- There are 2 categories of lymphadenopathy presentation³
 - 1. Localized: local cause originating from the lymph drainage area³
 - 2. Generalized: root systemic cause³

Non-Infectious Causes of Fever & Lymphadenopathy

- Autoimmune disorders such as sarcoidosis, systemic lupus erythematosus & rheumatoid arthritis²
- Malignant cancer neoplasms such as lymphoma & leukemia²
 - Hodgkin & non-Hodgkin lymphomas also cause weight loss, fever & drenching night sweats⁴
- Lymphoproliferative disorders such as hemophagocytic lymphohistiocytosis²
- Differential diagnoses of a Bartonella henselae infection may include: atypical mycobacterial diseases, valley fever, Lyme disease, lymphogranuloma venereum, nocardiosis, sarcoidosis, sporotrichosis, syphilis, & toxoplasmosis⁶

Infectious Causes

Bacterial Causes

- Bartonella henselae²
- Brucellosis²
- Bacterial pharyngitis²
- Syphilis²
- Tuberculosis²
- Typhoid fever²
- Francisella tularensis (Tularemia)⁵
 - May result from cat bites⁵

Non-Bacterial Causes

- Fungal & Protozoan⁵
 - Blastomycosis⁵
 - Toxoplasmosis⁵
- Viral
 - Hepatitis, cytomegalovirus, herpes simplex, Epstein-Barr virus, rubella, & viral pharyngitis²

What samples should be taken for diagnostic testing?

- Bartonella henselae infections are routinely detected & confirmed by blood samples or lymph node samples⁷
 - Primary lesion on skin is sampled for PCR testing¹²

Blood Samples

- Utilized for serological testing & bacterial culturing in systemic infections⁷
- Identify bacterium's present in blood^{8,10}
- Prevention of cross-contamination with sterile techniques is crucial⁹
- Blood cultures are preferred in bacteremia⁸
- Blood cultures allows susceptibility testing¹¹

Lymph Node Samples

- Aspirates are utilized for PCR diagnostics⁶
- Not routinely recommended⁷
- May be utilized for uncertain diagnosis⁷

Explain the tests that can be performed on samples to detect any of the potential bacterial pathogens associated with this disease. Discuss any specialized needs for this pathogen and limitations/advantages of mentioned tests

Direct detection methods identify bacterial components:⁶

- Microscopy⁶
- bacterial culture⁶
- gram staining⁶
- detection of pathogen antigens⁶
- histological spectrum⁶
- polymerase reaction chain testing⁶

- Indirect detection methods use serology to identify antibodies⁶
 - Current vs prior infection⁶

Microscopy

- Identifies bacteria¹¹
- Identified bacteria strain must be further test to determine the strain¹¹

Bacterial Culturing

- Identifies bacterial strain⁸
- Utilize selective or non-selective media¹¹
- Bartonella alpha proteobacteria growth medium (BAPGM) with added growth supplements is used for Bartonella henselae cultures^{13,9}
- Visual identification
 - turbidity, discrete colonies^{8,11}
 - Color change, fluorescent signals^{8,11}
- Gram stains
 - Pink gram-^{8,15}
 - Purple gram+^{8,15}
- Time Limitations due to incubation time^{6,8}

Enzyme-linked immunosorbent assays (ELISA)

- Indirect or direct identification¹¹
- Direct method uses bacterial antigens¹¹
- Coloured product for + results¹¹

Serology

- Preferred initial diagnostic for *Bartonella henselae*⁶
 - Does not differentiate between *Bartonella* spp as a result of cross-reactivity of antibodies
- Indirect fluorescent assays or ELISA⁶
- Antibodies interpreted as current or previous infection^{7,10}
- May test positive for *Bartonella henselae* after clearance of infection⁷
- May test negative for *Bartonella henselae* during early infection due to antibody level threshold^{6,7,8}

Histological Spectrum

- Samples collected by skin test, ESSA (clinical cases) or lymph biopsy^{17,18}
- Samples are stained & identified by granulomatous lesions & surrounding parenchymal changes^{17,18}
- High potential for differential diagnosis^{17,18}
 - Early infection exam results may yield misleading results^{17,18}

Polymerase Chain Reaction

- May be used to identify *Bartonella henselae*¹⁹
 - Gel electrophoresis¹⁹
- Blood or lymph tissue may be used & inoculated into cultures^{19,20}

What are the expected results from these tests allowing for the identification of the bacteria named in this case? Discuss what would be interpreted as a positive result for each type of test; for culture discuss appearance on stains and growth characteristics

Blood Culture

- Positive gram stain for *Bartonella henselae* is pink, pleomorphic & bacillary shaped⁶
- Can not differentiate between *Bartonella* spp as there are no unique characteristics to distinguish between them^{6,11}
- Not a preferred method for *Bartonella henselae* diagnosis
 - Slow growing & incubation up to 21 days
- Bacterial DNA analysis is recommended to identify *Bartonella henselae*^{13,8}

Enzyme-linked immunosorbent assays (ELISA)

- Wells containing *Bartonella henselae* are coloured¹¹

Serology

- Anti-Bartonella henselae antibodies interpreted as either active or prior infection¹⁰
- Anti-Bartonella henselae IgM titres 1:16 or higher: acute infection⁶
- Anti-Bartonella henselae IgG titres of 1:256 or higher: current or prior infection⁶
 - Current: marked increase in IgG levels between consecutive samples 7-10 days apart¹⁶
 - Prior: IgG levels do not change significantly during the 7-10 days¹⁶

Histological Spectrum

- Positive Bartonella henselae: granuloma with stellate central microabscess surrounded by histiocytes, lymphocytes, and dense fibrosis¹⁷
 - Prone to high amount of inconsistencies¹⁷

Polymerase Chain Reaction

- Gel electrophoresis PCR^{23,24}
 - Negative control: no DNA bands^{23,24,25}
 - Postive Control & Sample Lanes: bands occurring at same molecular weight marker^{23,24,25}
 - Postive Bartonella henselae: sample & postive control bands occur at the same markers^{23,24,25}
- Positive PCR-RFLP for Bartonella henselae: bands mirror positive gel electrophoresis sample^{23,24,25}
- Samples from PCR-RFLP & confirmed give a postive for Bartonella hensela²⁵

References

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