Brucella
“Lessons learnt”

Introduction
Brucella is a class 3 organism and as such poses a high risk to laboratory workers; it has been implicated as a potential agent of bioterrorism. In 2007, LabPLUS isolated three Brucella species from blood cultures and tissue. This lead to a strict protocol being introduced to minimize risk of exposure to Brucella species. In May 2013, we isolated a further Brucella suis which initially did not meet the criteria for performing all manipulations in a biosafety cabinet. This lead to staff exposure procedures being put into action and an additional step being added to our current protocol.

Background
Brucellosis is a zoonosis usually transmitted to humans by contact with infected animals and consumption of contaminated animal products. The disease is one of the most common causes of laboratory transmitted-infection with 2% of all cases being laboratory acquired. The reason brucellae are easily transmissible to laboratory worker is mainly due to its low infective dose and the way it enters the body primarily through respiratory mucosa, conjunctivae, gastrointestinal tract, or abraded skin (1).

2013 Brucella case
• 54 year old Tongan Male
• August 2006 seen in Rheumatoid clinic 3-4 year history
• Left knee pain attributed to osteoarthritis Rx flucloxacillin
• Admission November 2011 pain and swelling left knee - unsuccessful aspiration
• 20th May 2013 Total knee joint replacement
• 24/5/13 Positive blood culture: Gram negative coccobacillus seen

What happened next
Day 1
• Blood culture plates examined in biohazard safety cabinet as per protocol. Isolate Oxidase negative so protocol not required and processed on open bench.
• Biomerieux VITEK MS = no valid identification
Day 3
• Repeat Oxidase test = Positive
• API 20 NE possibility of Brucella species

Identification tests for brucella species
Specimens suitable for investigation are Blood, bone marrow, pus, synovial fluid, tissue, CSF, urine and genital exudates.
• Small gram negative bacillus (coccobacilli poor staining)
• Oxidase and Catalase positive
• Urea positive in 15mins-24hours
• Can be mis-identified as Moraxella phenylpyruvica
• Detection of agglutinating antibodies using an overnight Serum Agglutination Test (SAT)

Symptoms
• Fever (undulating)
• Night sweats
• Headache
• Malaise
• Arthralgia
• Muscle aches

Incubation period
Ranges from 1-3 weeks to several months.

Laboratory exposure protocol

High risk exposures classified as:
• Individuals who have performed a specifically implicated practice (e.g. sniffing bacteriological cultures, direct skin contact, present when aerosols have been generated).
• Individuals who were near (<5 feet) if work was performed with Brucella species on an open bench.
• Individuals in the laboratory during aerosol generating event.

Low risk exposure classified as:
• Other staff in the laboratory at the time of manipulation on an open bench but who do not have high-risk exposures as defined above.

Occasional health to ensure followed up of all laboratory workers in high risk category and staff in low risk category should be noted in their occupational health records (2).

Treatment
Oral doxycycline and rifampicin for 6 weeks, or a combination of oral doxycycline for 6 weeks and gentamicin or streptomycin for the first 2 weeks. Pregnant women trimethoprim-sulfamethoxazole for 6 weeks and complicated or life threatening clinical manifestations, or endocarditis or meningitis require the use of 3 drugs (3).

Prevention
• Raise awareness
• Occupational hygiene
• Laboratory safety

Conclusion
This experience has been a valuable lesson to us and has shown that suspicious Brucella cultures may require at the very least 18-24 hours growth on plate before a valid Oxidase result should be accepted. Awareness needs to be made that Brucella suis is not actually in the Biomerieux MS database. Strict adherence and enforcement of standard precautions is critical when Brucella is suspected and will help to reduce the chance of laboratory worker exposures. It is strongly recommended to follow-up directly exposed workers and give prophylaxis where necessary.

References
2. Procedural checklist to follow after a laboratory exposure to Brucella spp. (accessed 14th June 2013)
5. CDC Brucellae: laboratory procedures. Consultation report. (accessed 10th June 2013)

Identification tests for brucella species
Specimens suitable for investigation are Blood, bone marrow, pus, synovial fluid, tissue, CSF, urine and genital exudates.
• Small gram negative bacillus (coccobacilli poor staining)
• Oxidase and Catalase positive
• Urea positive in 15mins-24hours
• Can be mis-identified as Moraxella phenylpyruvica
• Detection of agglutinating antibodies using an overnight Serum Agglutination Test (SAT)

Symptoms
• Fever (undulating)
• Night sweats
• Headache
• Malaise
• Arthralgia
• Muscle aches

Incubation period
Ranges from 1-3 weeks to several months.

Laboratory exposure protocol

High risk exposures classified as:
• Individuals who have performed a specifically implicated practice (e.g. sniffing bacteriological cultures, direct skin contact, present when aerosols have been generated).
• Individuals who were near (<5 feet) if work was performed with Brucella species on an open bench.
• Individuals in the laboratory during aerosol generating event.

Low risk exposure classified as:
• Other staff in the laboratory at the time of manipulation on an open bench but who do not have high-risk exposures as defined above.

Occasional health to ensure followed up of all laboratory workers in high risk category and staff in low risk category should be noted in their occupational health records (2).

Treatment
Oral doxycycline and rifampicin for 6 weeks, or a combination of oral doxycycline for 6 weeks and gentamicin or streptomycin for the first 2 weeks. Pregnant women trimethoprim-sulfamethoxazole for 6 weeks and complicated or life threatening clinical manifestations, or endocarditis or meningitis require the use of 3 drugs (3).

Prevention
• Raise awareness
• Occupational hygiene
• Laboratory safety

Conclusion
This experience has been a valuable lesson to us and has shown that suspicious Brucella cultures may require at the very least 18-24 hours growth on plate before a valid Oxidase result should be accepted. Awareness needs to be made that Brucella suis is not actually in the Biomerieux MS database. Strict adherence and enforcement of standard precautions is critical when Brucella is suspected and will help to reduce the chance of laboratory worker exposures. It is strongly recommended to follow-up directly exposed workers and give prophylaxis where necessary.

References
2. Procedural checklist to follow after a laboratory exposure to Brucella spp. (accessed 14th June 2013)
5. CDC Brucellae: laboratory procedures. Consultation report. (accessed 10th June 2013)

Identification tests for brucella species
Specimens suitable for investigation are Blood, bone marrow, pus, synovial fluid, tissue, CSF, urine and genital exudates.
• Small gram negative bacillus (coccobacilli poor staining)
• Oxidase and Catalase positive
• Urea positive in 15mins-24hours
• Can be mis-identified as Moraxella phenylpyruvica
• Detection of agglutinating antibodies using an overnight Serum Agglutination Test (SAT)

Symptoms
• Fever (undulating)
• Night sweats
• Headache
• Malaise
• Arthralgia
• Muscle aches

Incubation period
Ranges from 1-3 weeks to several months.

Laboratory exposure protocol

High risk exposures classified as:
• Individuals who have performed a specifically implicated practice (e.g. sniffing bacteriological cultures, direct skin contact, present when aerosols have been generated).
• Individuals who were near (<5 feet) if work was performed with Brucella species on an open bench.
• Individuals in the laboratory during aerosol generating event.

Low risk exposure classified as:
• Other staff in the laboratory at the time of manipulation on an open bench but who do not have high-risk exposures as defined above.

Occasional health to ensure followed up of all laboratory workers in high risk category and staff in low risk category should be noted in their occupational health records (2).

Treatment
Oral doxycycline and rifampicin for 6 weeks, or a combination of oral doxycycline for 6 weeks and gentamicin or streptomycin for the first 2 weeks. Pregnant women trimethoprim-sulfamethoxazole for 6 weeks and complicated or life threatening clinical manifestations, or endocarditis or meningitis require the use of 3 drugs (3).

Prevention
• Raise awareness
• Occupational hygiene
• Laboratory safety

Conclusion
This experience has been a valuable lesson to us and has shown that suspicious Brucella cultures may require at the very least 18-24 hours growth on plate before a valid Oxidase result should be accepted. Awareness needs to be made that Brucella suis is not actually in the Biomerieux MS database. Strict adherence and enforcement of standard precautions is critical when Brucella is suspected and will help to reduce the chance of laboratory worker exposures. It is strongly recommended to follow-up directly exposed workers and give prophylaxis where necessary.