

Challenge PC243

November 2024

HISTORY

This paper challenge was sent to category A and C1 laboratories. The following scenario was presented to participants:

Your laboratory received a blood culture sample from a traveler returning from the Middle East. The blood culture became positive after 4 days of incubation. The Gram stain indicated is a tiny gram negative coccobacillus.

Please indicate the following actions you would take immediately? Select all that apply.

- A. Discard the blood culture sample and subculture plates without proceeding further
- B. Proceed with your laboratory's normal protocol
- C. Tape the subculture plates closed, ensure staff and supervisory staff are informed
- D. When the organism has grown, perform oxidase, catalase and urease in a BSC from the subculture
- E. Package the blood culture sample as RG3 and ship immediately to level 3 laboratory
- F. Package the subculture plates when growth is achieved and ship immediately, as a RG3 and ship to level 3 laboratory
- G. Our lab does not process blood cultures

CMPT QA/QC

The Committee considered the combined answers "C, D, and E" as the best answer.

SURVEY RESULTS

Reference labs: see Table 1. 3/13 (23%) reported C only, 3/13 (23%) reported C/D, 2/13 (15%) reported C/D/F, 2/13 (15%) reported C/E/F, 1/13 (8%) chose C/E, 1/13 (8%) chose E, and 1/13 (8%) reference laboratory indicated no blood cultures processed. No consensus was reached amongst reference laboratories; therefore, the challenge is not suitable for grading.

Participants: see Table 2. The most frequently reported result among participant labs was C/D 15/55 (27%). and 6/55 (11%) B only, 5/55

MAIN EDUCATIONAL POINTS from PC243

1. This scenario highlights the importance of recognizing, processing, and managing a Risk Group 3 (RG3) pathogen.
2. Immediate actions need to be taken when RG3 organism is suspected (i.e. tape subculture plates), and technologists should notify supervisory staff before proceeding.
3. Handling of clinical materials and basic biochemical tests can be performed in a biosafety cabinet II to reduce risk associated with aerosolization. However, all further manipulation of cultures should be performed at a CL3 laboratory.

(9%) C only, 5/55 (9%) C/D/F; there was heterogeneity across labs for various alternative answer combinations.

Grading

This challenge was ungraded

Table 1. Reported results - Reference labs

Reported	Total
C	3
C, D	3
C, D, F	2
C, E	1
C, E, F	2
E	1
G	1
Total	13

Table 2. Reported results - Participants

Reported	cat A	cat C1	Total	Grade
B	5	1	6	ungraded
B, C, D	1		1	ungraded
B, C, D, E	1		1	ungraded
B, C, E	1	1	2	ungraded
C	5		5	ungraded
C, D	15		15	ungraded
C, D, F	5		5	ungraded
C, E	3		3	ungraded
C, E, F	3		3	ungraded
C, F	1		1	ungraded
D	1		1	ungraded
E	4		4	ungraded
no report	2	1	3	ungraded
G	4	1	5	ungraded
Total	51	4	55	

COMMENTS ON RESULTS

The results were ungraded due to a lower-than-acceptable level of agreement among reference laboratories (<80%).

The committee agreed that the most correct answer would include options C, D, and E in combination. Choice C indicates the immediate actions required to ensure safety of lab personnel once a RG3 pathogen is suspected. Choice D indicates additional steps acceptable in a CL2 lab to support identification. Choice E facilitates referral for final identification and is the most expeditious means of transporting the specimen to a CL3 lab thereby minimizing delays in patient care. Choice F was considered acceptable; however, waiting for the subculture to grow before referring out may cause additional delays in patient care.

The committee notes that including choice D may have led some participants to select F instead of E by implying that subcultures were already growing.

Choice A and B were both considered unacceptable: the former would compromise clinical care while the latter could put laboratory personnel at risk. At least one participant laboratory indicated that the normal protocol contains instructions for handling RG3 organisms, which may have led to the selection of B.

CLINICAL SIGNIFICANCE

In this scenario, recovery of gram-negative coccobacilli with slow growth from primary blood culture at 4 days and a clinical history of travel to the Middle East should raise suspicion for *Brucella* spp., the causative agent of Brucellosis.

Transmission can occur via multiple routes (ingestion, direct contact with nonintact skin/mucous membranes, inhalation of aerosols), and *Brucella* spp. are one of the most commonly reported bacterial laboratory-acquired infections (LAI).^{1,2}

A recent systematic review and meta-analysis of 164 LAIs in clinical labs from 1990-2023 implicated *Brucella* as the leading pathogen (55.5%).³ As *Brucella* spp. are considered RG3 pathogens, risk groups are used to stratify organisms by risk to the individual and community.

RG3 pathogens pose a high risk to an individual's health (likely to cause severe disease) but low public health/community risk, and for which effective treatments usually exist.⁴ With a few exceptions, most RG3 organisms require a containment level 3 (CL3) lab for manipulation of cultures, which are equipped with additional engineering controls and specialized biosafety equipment for personnel safety and to mitigate risk related to pathogen release into the environment.⁴

MALDI-TOF mass spectrometry (MS) has been a fast and reliable method for bacterial identification based on protein profile characteristics of each microorganism. Databases have been developed that include the main pathogenic microorganisms, thus allowing the use of this method in routine bacterial identification. Nevertheless, *Brucella* has not been still incorporated into

some of the main databases available because of problems derived from their potential bioterrorist use. This is an important problem for the routine use of MALDI-TOF MS for the direct diagnosis of blood cultures in countries where brucellosis is still frequent.⁵

REFERENCES

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