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Director, Bureau of Laboratories Sandip Shah, PhD, HCLD(ABB)







Infectious Substances Transport, Advanced Training Opportunity CDC Laboratory Outreach Communication System LRN-Biological: Sentinel Rule-Out Spotlight

Bureau Vision

In this issue...

The Bureau of Laboratories is a stronger, more diverse team within an integrated public health system. We utilize advanced technology and innovative leadership to provide comprehensive public health services in our dynamic global community.

Bureau Mission

We are dedicated to continuing leadership in providing quality laboratory science for healthier people and communities through partnerships, communication, and technical innovation.

LabLink is published quarterly by the Michigan Department of Health and Human Services Bureau of Laboratories, to provide laboratory information to Michigan health professionals and the public health community.

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Editor: Teresa Miller



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Infectious Substances Transport, Advanced Training Opportunity

MDHHS Bureau of Laboratories (BOL) is pleased to announce an educational opportunity. APHL is partnering with Saf-T-Pak to provide free comprehensive training for the safe transport of infectious substances, biological specimens, dry ice, and related materials. This partnership offers exclusive training for laboratory healthcare professionals interested in development of internal subject matter expertise in infectious substances transport training and shipping regulations.

For more information about this full day virtual seminar or to obtain a flyer containing comprehensive training details please contact the BOL Bioterrorism Training Coordinator, Jason Wholehan, <u>wholehanj@michigan.gov</u>, 517-335-9653.

Training Schedule for APHL/Saf-T-Pak Training

#	Date of Training	TIME	Registration DEADLINE
12	Thursday, December 8, 2022	9:00 a.m 4:00 p.m. EST	Tuesday, November 22, 2022
14	Tuesday, January 10, 2023	9:00 a.m 4:00 p.m. EST	Thursday, December 22, 2022
15	Thursday, January 12, 2023	9:00 a.m 4:00 p.m. EST	Thursday, December 22, 2022
17	Friday, January 20, 2023	9:00 a.m 4:00 p.m. EST	Tuesday, January 3, 2023
19	Thursday, February 9, 2023	9:00 a.m 4:00 p.m. EST	Monday, January 24, 2022
20	Friday, February 17, 2023	9:00 a.m 4:00 p.m. EST	Friday, February 3, 2023
22	Thursday, March 9, 2023	9:00 a.m 4:00 p.m. EST	Thursday, February 23, 2023
23	Friday, March 17, 2023	9:00 a.m 4:00 p.m. EST	Saturday, March 4, 2023
25	Thursday, April 6, 2023	9:00 a.m 4:00 p.m. EST	Tuesday, March 21, 2023
26	Friday, April 21, 2023	9:00 a.m 4:00 p.m. EST	Friday, April 7, 2023
28	Thursday, May 4, 2023	9:00 a.m 4:00 p.m. EST	Tuesday, April 18, 2023
29	Friday, May 19, 2023	9:00 a.m 4:00 p.m. EST	Friday, May 5, 2023



CDC Laboratory Outreach Communication System Have You Subscribed to the CDC Laboratory Outreach Communication System Yet?

Centers for Disease Control and Prevention Laboratory Outreach Communication System (LOCS) provides timely information to the laboratory and testing community. Topics include emergency preparedness and response, point-of-care testing, specimen collection, antigen testing, biosafety, laboratory data reporting, and regulatory requirements, as well as training and other resources to support your work. Click here to opt in now.





LRN-Biological: Sentinel Rule-Out Spotlight

A Tricky LPX Survey: Burkholderia pseudomallei vs. Pseudomonas stutzeri.

In the recent CAP LPX 2022 Set A (College of American Pathologists, Laboratory Preparedness Exercise), Organism LPX-03 presented a tricky challenge for participating Sentinel Laboratories. The case description presented, along with the organism, lead the microbiologist to assume the organism was a potential Burkholderia pseudomallei. B. pseudomallei is often referred to as the Vietnam "time-bomb," because several decades ago many American soldiers were sent to Vietnam where the organism is endemic. This organism may have a long incubation period before symptoms appear. In one extreme case, the incubation period lasted 62 years before a *B. pseudomallei* infection occurred.

The LPX-03 case clearly fits the risk factors for a latent *B. pseudomallei* infection. The patient is described as a Vietnam veteran who developed lung cancer. However, the organism was a close mimic of *B. pseudomallei—Pseudomonas stutzeri*. The two bacteria share many characteristics: both are slow-growing, Gram-negative rods nurtured on MacConkey agar, nonhemolytic on Sheep blood agar, grow at 42°C, catalase positive, indole negative, and oxidase positive.

The main difference between *B. pseudomallei* and *P. stutzeri* is that *B. pseudomallei* is resistant to colistin and polymyxin B and is Arginine positive; while *P. stutzeri* is susceptible to colistin and polymyxin B and is Arginine negative.

Testing for colistin resistance and Arginine reaction, as described in the American Society for Microbiology (ASM) Sentinel Laboratory Guidelines, *LRN Sentinel Level <u>Clinical Laboratory Protocols (asm.org)</u>*, presents a challenge to many laboratories that may not stock these items. It is recommended to stock all biochemicals and be able to perform the full ASM testing catalog. However, if this is not possible, there are some work arounds noted in the ASM. "As an alternative to polymyxin B or colistin testing, growth on *B. cepacia* selective agars or modified Thayer Martin may substitute for the colistin or polymyxin B disk test, because these media contain polymyxin B or colistin. However, the lack of growth on these media should be

confirmed by the disk test." If possible, Arginine "biochemical reactions" can even be evaluated from the API 20E, if the laboratory has no system specific for glucose nonfermenting organisms."

Interpretation of growth from a modified Thayer Martin agar can be a challenge as seen in the *Picture 1*.



B. thailandensis: **Pinpoint growth throughout**

Picture 1: Colistin resistant *B. thailandensis* and colistin susceptible *P. stutzeri* are shown here, inoculated to modified Thayer Martin media. Notice that *B. thailandensis* shows pinpoint growth throughout the streaking pattern. *P. stutzeri* is able to breakthrough and grow only where the inoculum is heaviest and fails to grow throughout the streaking pattern.

P. stutzeri: **Growth only where** heavily inoculated

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LRN-Biological: Sentinel Rule-Out Spotlight

Interpretation of growth from a modified Thayer Martin can be a challenge as seen in *Picture 1.* Breakthrough of the susceptible organism can be misinterpreted as slow growth or pinpoint growth. Subbing a visual "control" of a colistin resistant organism may be helpful. In LPX-03, participants also reported difficulty interpreting the lactose reaction from MacConkey agar. As seen in *Picture 2*, organisms like *B. pseudomallei* and *P. stutzeri* may uptake some pigment and appear light pink. However, this reaction is not true lactose fermentation, and should be recorded as negative.



Picture 2: MacConkey Agar 24hr 35°C. A true Lactose Positive, and Lactose Negative are shown along with *P. stutzeri*. Make note of the pale pink color of the P. stutzeri. This is not caused by Lactose fermentation and should not be recorded as such; the pale pink hue comes from other pathways.

LPX-03 correct responses include:

- 1. A statement that says the isolate should be forwarded to an LRN Reference Laboratory according to the ASM rule-out guidelines because of the biochemical similarity of the organisms presented in this case.
- 2. An answer that the isolate should be forwarded to an LRN Reference Laboratory because the ASM rule-out guidelines were followed, but biochemicals were not available to complete the recommended testing.
- 3. Affirmation that the isolate should be forwarded to an LRN Reference Laboratory because Sentinel Laboratories need to limit manipulation of isolates that mimic biothreat agents.

If you would like more information about Sentinel Testing Guidelines or to schedule a lecture reviewing "Bacterial Biothreat Agents; Recognize, Rule-out, Refer", please contact the Bioterrorism Training Coordinator, Jason Wholehan wholehanj@michigan.gov.



The APHL Bench Card Booklet contains a helpful flowchart for identifying Biothreat Gram Negative Bacilli on page 13. Additionally, the booklet presents each organism with colored pictures of Sentinel tests and biochemical reactions. To request printed and laminated copies of this resource, please contact Jason Wholehan, wholehanj@michigan.gov.

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