

Michigan 2010 CAP LPX-A and LPX-B Survey Analysis

Introduction

One purpose of the CAP LPX survey is to provide laboratories with an educational exercise that can be used to help prepare for the detection of pathogens of epidemiologic importance, including pathogens that can be used as biothreat agents. Another purpose of the LPX is to prepare participant laboratories for effective and efficient communication of critical information related to potential BT agents. This report summarizes the results of the Michigan Laboratory Response Network (LRN) Sentinel Laboratories on the 2010 LPX-A and LPX-B survey panels in aggregate and compares Michigan lab responses to those of participating labs throughout the country.

Performance Summary

The LPX survey consists of organism identification (rule-out) plus a notification component to test communications between LRN Sentinel Laboratories and LRN Reference Labs. In these exercises, LRN Sentinel Labs are required to contact their LRN Reference Lab if, after following the established Sentinel Laboratory Guidelines on a challenge isolate, they are unable to rule out an agent of bioterrorism. Both organism rule-out and notification are summarized below.

Approximately 40% of Michigan sentinel labs participated in the 2010 LPX-A and LPX-B surveys.

The 2010 LPX-A survey contained the following samples:

LPX-01	<i>Yersinia pestis</i>
LPX-02	<i>Pseudomonas stutzeri</i>
LPX-03	<i>Bacillus anthracis</i>

The 2010 LPX-B survey contained the following samples:

LPX-04	<i>Haemophilus influenzae</i>
LPX-05	<i>Burkholderia thailandensis</i>
LPX-06	<i>Francisella tularensis</i>

Correct Result Reporting LPX-A		N = 44
Sample Number	% of MI Labs with Intended Response	
LPX-01	79.55% (35/44)	
LPX-02	76.7% (33/43) # ^	
LPX-03	97.7% (43/44)	

Correctly identified as Non-BT culture

^ One lab did not indicate they identified an agent in the specimen thus N=43

Correct Result Reporting LPX-B		N = 44
Sample Number	% of MI Labs with Intended Response	
LPX-04	95.45% (42/44) #	
LPX-05	97.7% (43/44) #	
LPX-06	97.67% (42/43) ^	

Correctly identified as Non-BT culture

^ One lab did not indicate they identified an agent in the specimen thus N=43

Notification Drill Results

Notification Drill LPX-A				N = 44
Sample Number	Notification Required	% MI Labs Indicating Would Notify the LRN Ref Lab	% MI Labs Actually Notified the LRN Reference Lab	
LPX-01	Yes	79.5% (35/44)	56.8% (25/44)	
LPX-02	No	18.2% (8/44) ∇	11.4% (5/44) ∇	
LPX-03	Yes	90.9% (40/44)	63.6% (28/44)	

Notification Drill LPX-B				N = 44
Sample Number	Notification Required	% MI Labs Indicating Would Notify the LRN Ref Lab	% MI Labs Actually Notified the LRN Reference Lab	
LPX-04	No	6.8% (3/44) ∇	6.8% (3/44) ∇	
LPX-05	Yes	79.5% (35/44)	61.4% (27/44)	
LPX-06	Yes	90.9% (40/44)	72.7% (32/44)	

∇ Although notification was not necessary in these cases, it is great that sentinel labs are willing and able to communicate with their LRN Reference Lab.

Analysis by Sample

LPX-01: <i>Yersinia pestis</i>		
Submitted Answers	Michigan Participants	All Participants
§ <i>Yersinia pestis</i> , confirmed	0/44 0%	53/1359 3.9%
§ <i>Yersinia pestis</i> , refer for confirmation	4/44 9.1%	187/1359 13.8%
§ <i>Yersinia</i> sp., refer to rule out <i>Yersinia pestis</i>	17/44 38.6%	350/1359 25.8%
§ Gram-negative bacillus, refer to rule out <i>Yersinia pestis</i>	14/44 31.8%	548/1359 12.6%
Gram-negative bacillus, refer to rule out <i>Burkholderia mallei</i>	1/44 2.3%	-
Non-BT Culture	8/44 18.2%	-

§ Acceptable response

LPX-02: <i>Pseudomonas stutzeri</i>		
Submitted Answers	Michigan Participants	All Participants
§ Non-BT Culture	33/43 76.7%	865/1353 63.9%
Gram-negative bacillus, rule out <i>Burkholderia pseudomallei</i>	7/43 16.3%	-
<i>Brucella</i> sp., refer for confirmation	1/43 2.3%	-
Gram-negative coccobacillus, refer to rule out <i>Burkholderia mallei</i>	1/43 2.3%	-
<i>Burkholderia</i> , refer to rule out <i>Burkholderia pseudomallei</i>	1/43 2.3%	-

§ Acceptable response

One lab failed to report identifying an agent in LPX-02, therefore N=43

LPX-03: <i>Bacillus anthracis</i>		
Submitted Answers	Michigan Participants	All Participants
§ <i>Bacillus anthracis</i> , confirmed	0/44 0%	39/1364 2.9%
§ <i>Bacillus anthracis</i> , refer for confirmation	0/44 0%	96/1364 7.0%
§ <i>Bacillus</i> sp., refer to rule out <i>Bacillus anthracis</i>	38/44 86.4%	913/1364 66.9%
§ Gram-positive bacillus, refer to rule out <i>Bacillus anthracis</i>	5/44 11.4%	74/1364 5.4%
Non-BT Culture	1/44 2.3%	-

§ Acceptable response

LPX-04: <i>Haemophilus influenzae</i>		
Submitted Answers	Michigan Participants	All Participants
§ Non-BT Culture	42/44 95.45%	1092/1183 92.3%
Gram-negative coccobacillus, refer to rule out <i>Brucella</i> sp.	2/44 4.5%	-

§ Acceptable response

LPX-05 <i>Burkholderia thailandensis</i>		
Submitted Answers	Michigan Participants	All Participants
§ Non-BT Culture	6/44 13.6%	189/1189 15.9%
§ <i>Burkholderia</i> sp., refer to rule out <i>Burkholderia pseudomallei</i>	7/44 15.9%	190/1186 16.0%
§ Gram-negative bacillus, refer to rule out <i>Burkholderia pseudomallei</i>	19/44 43.2%	475/1189 40%
<i>Burkholderia pseudomallei</i> , refer for confirmation	10/44 22.7%	-
Gram-negative bacillus, refer to rule out <i>Yersinia pestis</i>	2/44 4.5%	-

§ Acceptable response

LPX-06: <i>Francisella tularensis</i>		
Submitted Answers	Michigan Participants	All Participants
§ <i>Francisella tularensis</i> , confirmed	0/43 0%	55/1191 4.6%
§ <i>Francisella tularensis</i> , refer for confirmation	5/43 11.6%	166/1191 13.9%
§ <i>Francisella s</i> sp., refer to rule out <i>Francisella tularensis</i>	6/43 13.9%	135/1191 11.3%
§ Gram-negative bacillus/coccobacillus, refer to rule out <i>Francisella tularensis</i>	31/43 72.1%	772/1191 64.8%
Non-BT culture	1/43 2.3%	42/1191 3.5%

§ Acceptable response

One lab failed to report identifying an agent in LPX-06, therefore N=43

Discussion

LPX-01

This challenge contained *Yersinia pestis* strain devoid of the 75 kb low calcium response (LCR) virulence plasmid. Nationally, the majority of participants appropriately managed this sample. 83.8% of all laboratories replied with an intended response. However, 12.6% indicated that *Y. pestis* was a non-bioterrorism culture, a concern for the LRN sentinel network and those laboratories in particular.

Seventy-nine percent of the 44 Michigan laboratories who participated in this survey responded with an acceptable response identifying this as either *Yersinia pestis* or recognizing the possibility of *Y. pestis* being present in the sample. This compares to 83.7% of all participating laboratories. One laboratory indicated the presence of *Burkholderia mallei* and eight indicated this was a non-BT culture.

The isolate from this challenge should have triggered a communication with the LRN Reference Laboratory. One Michigan laboratory that identified the possibility of *Y. pestis* being present in this sample did not indicate they would contact their LRN Reference Lab but stated they would refer the sample to their commercial reference lab. Participants in LPX exercises are required to contact their LRN Reference Laboratory if, after performing the established Sentinel Laboratory Guidelines on a challenge isolate, they are unable to rule out an agent of bioterrorism.

LPX-02

This challenge contained *Pseudomonas stutzeri* in pure culture. The appropriate response was non-BT culture. Only sixty-four percent of all participating labs indicated that this culture was a non-BT isolate. Thirty-five percent of participating labs indicated they would contact the appropriate LRN reference lab, which for this isolate was an unnecessary step. *Burkholderia* sp., which was among the responses of labs not reporting non-BT agent, can be ruled out based on the Sentinel Level Clinical Microbiology Laboratory Guidelines.

33/43 (76.7%) of Michigan labs participating in this survey responded with an acceptable response indicating this was not a potential biothreat agent. This compares to 63.9% of all participating labs. Nine Michigan labs indicated the possibility that this sample contained *Burkholderia mallei* or *pseudomallei* and another indicated the possibility of *Brucella* being present. Two of the Michigan labs that suspected *Burkholderia pseudomallei* indicated they would refer the specimens to their commercial laboratory instead of their LRN Reference Lab. Participants in LPX exercises are required to contact their LRN Reference Laboratory if, after performing the established Sentinel Laboratory Guidelines on a challenge isolate, they are unable to rule out an agent of bioterrorism. This challenge would not require notification of the LRN Reference Laboratory unless the Sentinel lab could not rule out the presence of a biothreat agent.

LPX-03

This challenge contained the Sterne strain of *Bacillus anthracis* devoid of the plasmid pX02. Nationally, the majority of participants appropriately managed this sample. Ninety-five percent of all participating labs provided an intended response for this sample.

43/44 (98%) of participating Michigan labs correctly identified the possibility of *Bacillus anthracis* being present in this sample. One Michigan lab indicated this was a non-BT agent. Three participating Michigan labs identified the possibility of *Bacillus anthracis* in this challenge yet indicated they would refer it to their commercial reference laboratory rather than to the LRN Reference Lab. LRN Sentinel Labs are required to notify their LRN reference laboratory when they are unable to rule out an agent of bioterrorism. The isolate from this challenge should have triggered a communication with the LRN Reference Laboratory.

LPX-04

This challenge contained *Haemophilus influenzae* in pure culture. The appropriate response was non-BT culture. Ninety-two percent of all participating labs indicated that this culture was a non-BT isolate.

42/44 (95%) of participating Michigan labs correctly reported this as a non-BT culture. However, two Michigan laboratories suspected this challenge contained a *Brucella* sp. When bioterrorism agents have been ruled out, LRN Reference Lab notification is not required. The two Michigan labs suspecting a biothreat agent, both indicated they would notify their LRN Reference Laboratory. This challenge should not have triggered a communication with the LRN Reference Laboratory.

LPX-05

This challenge contained *Burkholderia thailandensis* in pure culture. *B. thailandensis* is a mimic for *B. pseudomallei* as differentiation of the two species requires further testing by an LRN reference Laboratory. The intended response for Sentinel Laboratories using the Sentinel Level Clinical Microbiology Laboratory Guideline was either “*Burkholderia* sp., refer to rule out *Burkholderia pseudomallei*” or “Gram-negative bacillus, refer to rule out *Burkholderia pseudomallei*.”

Participant laboratories showed variable performance for several key biochemical reactions on this challenge: oxidase, catalase and motility. Laboratories that reported oxidase negative, catalase negative or motility negative are encouraged to review the Sentinel Level Clinical Microbiology Laboratory Guidelines to ensure their protocols are compliant. (<http://www.asm.org/images/pdf/BpseudomalleiBmalleiRevision2008.pdf>)

Surprisingly, a significant number of labs did not perform key tests from the Sentinel Level Clinical Microbiology Laboratory Guidelines on this challenge sample. Catalase was not performed by 21% of participants; Indole was not performed by 21%; Motility was not performed by 32%; Colistin or Polymyxin B resistance was not tested by 47% and 48% respectively. **These laboratory practices do not comply with the Sentinel Level Clinical Microbiology Laboratory Guidelines, and it is unclear how these labs were able to rule-in or rule-out a BT agent in this challenge specimen.** Not following the guidelines is likely to increase the risk of laboratory acquired infection for laboratory staff and may delay appropriate communication with LRN Reference Laboratories and other relevant Public Health Authorities. Laboratories are encouraged to review the Sentinel Level Clinical Microbiology Laboratory Guidelines and integrate these into their routine laboratory protocols.

32/44 (73%) of participating Michigan labs correctly identified this challenge.

The isolate from this challenge should have triggered a communication with the LRN Reference Laboratory if a biothreat agent could not be ruled out. Three participating Michigan labs whose testing indicated a potential biothreat agent (two *B. pseudomallei* and one *Y. pestis*) did not indicate the need to notify the LRN Reference Laboratory but, instead, would refer the sample to their commercial reference lab. **LRN Sentinel Labs are required to notify their LRN reference laboratory when they are unable to rule out an agent of bioterrorism.**

LPX-06

This challenge contained *Francisella tularensis*, the live vaccine strain (LVS). Overall, participants performed well in this challenge with >90% of labs providing one of the expected results. As discussed in the LPX-05 discussion, a surprising number of participants did not perform key tests: oxidase was not performed by 3% of participants; catalase was not performed by 11%; beta-lactamase was not performed by 32%; satellite/XV was not performed by 71%; urease was not performed by 20%. **These laboratory practices do not comply with the Sentinel Level Clinical Microbiology Laboratory Guidelines, and it is unclear how these laboratories were able to rule-in or rule-out a BT agent in the challenge specimen.** Not following the guidelines is likely to increase the risk of laboratory acquired infection for laboratory staff and may delay appropriate communication with LRN Reference Laboratories and other relevant Public Health Authorities. Laboratories are encouraged to review the Sentinel Level Clinical Microbiology Laboratory Guidelines and integrate these into their routine laboratory protocols.

42/43 (98%) of participating Michigan labs correctly identified this challenge as *Francisella tularensis*, *Francisella* species or a gram-negative bacillus/coccobacillus. This compares to 95% of all participating labs. One (2.3%) Michigan lab incorrectly identified this specimen as a non-BT culture.

The isolate from this challenge should have triggered a communication with the LRN Reference Laboratory. Two (4.6%) Michigan participating labs that detected this pathogen that could potentially be used as a biothreat agent did not indicate the need to notify the LRN Regional Laboratory but instead would have referred it to a commercial reference laboratory. **Notification of the LRN Reference Laboratory is required if unable to rule out an agent of bioterrorism.**

Comparison between Michigan 2009 and 2010 LPX Survey Results

Michigan Result Reporting		
Organism Present	% of MI Labs with Intended Response 2009 (LPX A LPX B)	% of MI Labs with Intended Response 2010 (LPX A LPX B)
<i>Y. pestis</i>	74.4% 78.6%	79.55% NI*
<i>F. tularensis</i>	97.6% 93%	NI* 97.7%
<i>B. anthracis</i>	NI* 88.4%	97.7% NI*
Non-BT culture	93% 97.7%	76.7% 95.5%

* NI = Not included in challenge set.

Comparison between Michigan 2009 and 2010 Notification Drill Results

Michigan Notification Results		
Year	Lowest % of MI Labs that Actually Notified the LRN Reference Lab When Notification Required	Highest % of MI Labs that Actually Notified the LRN Reference Lab When Notification Required
2009	41.9%	53.5%
2010	56.8%	72.7%

Overall Performance

Nationally, LRN sentinel laboratories are more appropriately limiting the identification of bioterrorism agents and referring the isolates to LRN Reference Labs for their confirmatory identification. It is of some concern that LRN sentinel laboratories may not have all the bioterrorism screening tests available. An example is urease testing, an important test for the identification of *Brucella* sp. This year, between 7-9% of labs reported that they do not have urease testing available. As seen in LPX-05, a significant number of labs did not perform key tests from the Sentinel Level Clinical Microbiology Laboratory Guidelines. Not following the guidelines is likely to increase the risk of laboratory acquired infection for laboratory staff and may delay appropriate communication with LRN Reference Laboratories and other relevant Public Health Authorities. Laboratories are encouraged to review the Sentinel Level Clinical Microbiology Laboratory Guidelines and integrate these into their routine laboratory protocols.

In Michigan, the data from 2009 to 2010 shows that Sentinel Laboratories are improving their detection techniques for biothreat agents as the percentage of labs with the intended response was stable for *Y. pestis* and *F. tularensis* and improved for *B. anthracis*. The Notification Drill shows improvement as well. However, more improvement is needed in this area as only 53-73% of Sentinel Labs actually notified their LRN Regional Laboratory when they could not rule out an agent of bioterrorism during the 2010 LPX challenge. Remember, Sentinel Labs MUST actually contact their LRN Regional Laboratory when a biothreat agent can not be ruled out. It is not sufficient to simply state you would make that contact.

Thank you for participating in the CAP LPX Challenges. If your laboratory desires refresher training on any of the LRN Rule-Out Procedures, please contact the Michigan Department of Community Health Bureau of Laboratories Bioterrorism Coordinator, Valerie Reed, via e-mail at ReedV@michigan.gov.