A Practical Guidance Document for the Laboratory Detection of Toxigenic *Clostridium difficile* September 21, 2010*

HIGHLIGHTS:

1. Testing for toxigenic *C. difficile* should be limited to patients with ≥ 3 non-formed stool specimens per 24 hr period, unless ileus (obstruction) is suspected.
2. Utilizing toxin A/B EIA for *C.difficile* diagnosis is insensitive and no longer recommended as a stand alone test.
3. Glutamate dehydrogenase (GDH) antigen assays have been found to be good screening tests for *C.difficile* infection (CDI) in many studies with high sensitivity and negative predictive values.
4. Positive GDH assay results must be confirmed. A GDH positive result along with a positive toxin A/B EIA, a positive cytotoxin neutralization, or a positive nucleic acid amplification test (NAAT) result may be reported as positive for toxigenic C. difficile. If the A/B EIA or cytotoxin neutralization assay is used and is negative, specimens should be further tested by either NAAT or toxigenic culture.
5. Laboratories can also use a NAAT to detect *C.difficile* toxin genes as a stand alone diagnostic test.
6. Repeat testing following a positive test (test of cure) is not recommended since patients may carry toxigenic *C. difficile* for months after clinical cure. Repeat testing following a positive test is appropriate if the patient improves with therapy and relapses after the completion of a treatment regimen (clinical relapse).
7. Repeat testing following a negative test is not recommended if one of the suggested algorithms (see below) is used because nearly all positive patients will be detected (high sensitivity). Testing a second specimen from a negative patient is more likely to be a false-positive.
8. Up to 50% of neonates may be colonized with toxigenic *C.difficile*. Testing for *C.difficile* infection (CDI) in this population should proceed only after consultation with the clinician.

COMMENTARY:

The laboratory diagnosis of *C.difficile* remains a challenge to microbiology laboratories. A significant number of clinical laboratories are currently utilizing EIA for toxin A/B testing due to its ease of use. However, there is a wealth of information now available indicating that utilizing this test alone is not appropriate for toxigenic *C.difficile* detection.

Many laboratories have adopted NAAT testing alone or a 2- or 3-step algorithm for the most sensitive diagnosis of CDI (see below for sample algorithms). The 2- or 3-step algorithm approach utilizes GDH antigen (which is a common antigen found in both toxigenic and non-

toxigenic isolates of *C.difficile*) followed by cytotoxin neutralization or toxin A/B EIA, and NAAT or toxigenic culture when GDH-positive specimens are cytotoxin neutralization or EIA negative.

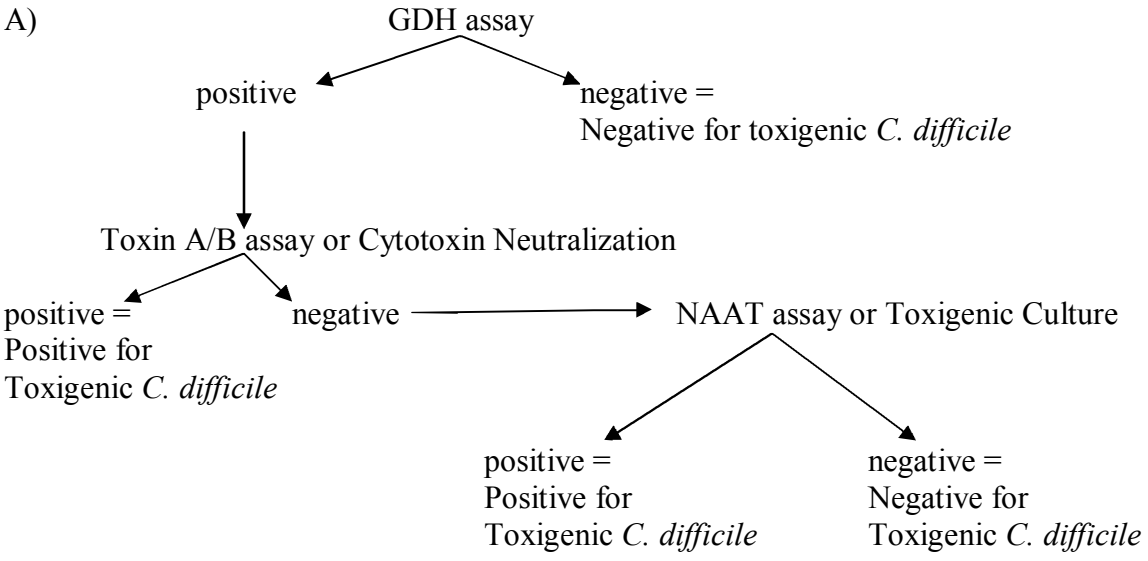
Diarrhea in hospitalized patients is common. However, as few as 10-20% of these patients have toxigenic *C. difficile* as the cause. In addition, the prevalence of asymptomatic carriage of toxigenic *C. difficile* is not known. Only patients with 3 or more watery, loose, or unformed stools (stools that take the form of the container) per day qualify for *C.difficile* toxin testing. An important exception is the very rare case where a patient has ileus (obstruction of the intestine due to paralysis of the intestinal muscles) without diarrhea. Any testing of formed stool specimens would need to be discussed with the clinician prior to testing to address the issue of ileus. Swab specimens are not appropriate samples for *C.difficile* testing.

Final reports of test results should be clear that the specimen is either positive or negative for toxigenic *C. difficile*. If a 2 or 3 step algorithm is used, preliminary reports indicating the result of the GDH and other sequential tests in the algorithm are likely to be misleading. Laboratory personnel should consult with appropriate medical staff to determine the most useful reporting procedure.

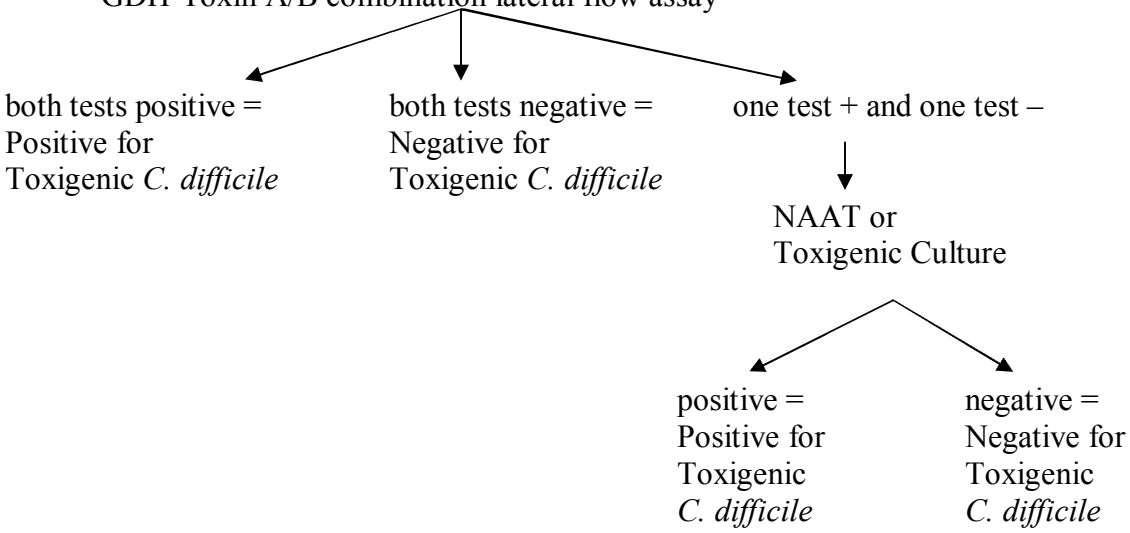
*This document updates the September 9, 2010 version. Future updates will be made available as new information is published.

THREE SAMPLE ALGORITHMS:

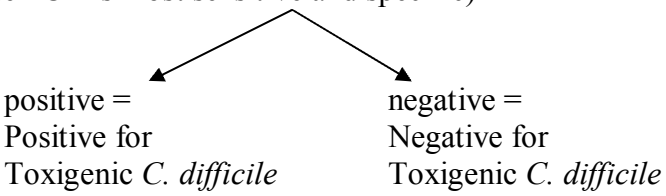
A)



B) GDH-Toxin A/B combination lateral flow assay



C) NAAT as stand alone test (To date PCR is most sensitive and specific)



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