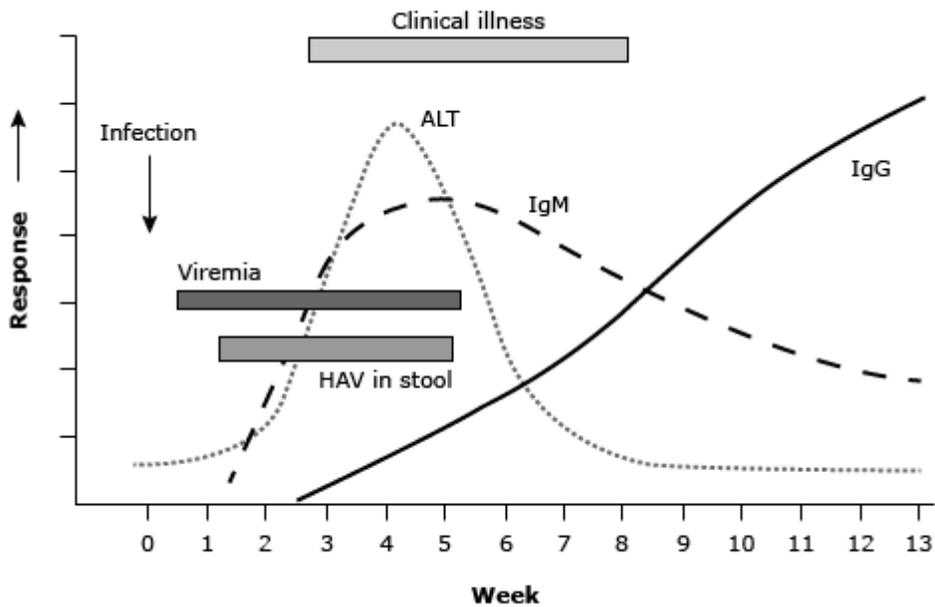


Hepatitis A Virus Outbreak Antibody Testing: Frequently Asked Questions

Background: Hepatitis A Virus (HAV) IgM antibodies in patient stool and sera demonstrate a recent exposure to hepatitis A virus or vaccine response. In most infected individuals, HAV IgM antibody is detectable 5 to 10 days before the onset of symptoms and declines to undetectable levels within 6 months. However, sensitive immunoassays will occasionally detect anti-HAV IgM for up to one year after acute hepatitis infection. At the onset of symptoms, the presence of anti-HAV IgG rises with the presence of anti-HAV IgM. Since anti-HAV IgG persists lifelong after acute infection, detection of anti-HAV IgG alone indicates past infection or vaccine response. IgG antibodies typically provide life-long immunity against future infection. HAV IgM testing is not recommended to determine infection before the administration of PEP due to the possibility of false positive test results and early antibody waning post infection.

Mayo Medical Laboratories and possibly other commercial laboratories offer reportable HAV PCR testing, which may detect virus before the IgM anti-HAV appears.

Course of hepatitis A:



Timeline for hepatitis A manifestations.

ALT: alanine transaminase; HAV: hepatitis A virus; Ig: immunoglobulin.

Reproduced from: American Medical Association; American Nurses Association-American Nurses Foundation; Centers for Disease Control and Prevention; Center for Food Safety and Applied Nutrition, Food and Drug Administration; Food Safety and Inspection Service, US Department of Agriculture. Diagnosis and management of foodborne illnesses: a primer for physicians and other health care professionals. MMWR Recomm Rep 2004; 53(RR-4):1. Graphic 57931 Version 4.0

Are false positive IgM results possible?

IgM testing may lead to false positives if a patient has been vaccinated prior to having blood drawn for serology. The Bureau of Laboratories is unable to offer vaccine status testing for patients at this time. Work is underway to determine if there are appropriate assays to assist in determining vaccine efficacy for high risk populations (i.e. HIV populations).

There is no indication that specific assays have problems with biological false positives more than others at this time. All assays that have gone through FDA approval processes have sensitivity and specificity information that may be acquired from the manufacturers. Generally speaking, the sensitivities and specificities of HAV IgM assay are high; indicating a low likelihood of biological false positives. There are some published data regarding some substances that may lead to false positives or false negatives. The Bureau of Laboratories has not been able to perform a comprehensive literature review or assessment on these published claims.

The Bureau uses the MONOLISA kit for HAV IgM testing. The manufacturer's performance characteristics of this test showed positive and negative percent agreement with the reference test at >96% with a 95% confidence interval. The cross reactivity study showed no other disease states causing false positive results, including other hepatitis viruses and conditions that typically produce cross reacting antibodies. Interferences in absorbance readings may be caused by heterophilic antibodies present in samples from individuals regularly exposed to animals or animal serum products and samples with bacterial contamination. Other test methodologies have demonstrated false positive results in patients with other viral diseases or underlying illnesses and in those with Non-Hodgkin's Lymphoma. Dietary supplements containing biotin or vitamin B7 taken within 24 hours of blood collection may also cause false positive reactions with *chemiluminescent* methodologies.

Does the Michigan Department of Health and Human Services Bureau of Laboratories have a Limit of Detection for molecular testing?

At this time, Bureau of Laboratories does not have a Limit of Detection (LOD) for the molecular testing that is performed for genotyping. The laboratory performs repeat IgM testing on any specimens that are "negative" on the nested PCR/genotyping analysis. Approximately 50% of negative genotyping results are HAV IgM negative when tested at BOL.

It is important to note that it is unclear if result differences are due to minor changes in genetic sequences for specimens that may be positive by IgM but negative for genotyping, or if the differences are due to limit of detection differences.

Please keep in mind that genotyping is not a diagnostic assay.

What is the impact of storage conditions, shipment or time between testing on the IgM assay?

It is not known what impact storage conditions, shipment, or length of time from specimen collection to date of testing has on IgM assay differences even when the same assay is used between laboratories. It has been suggested by some studies that storage conditions and shipment temperatures may impact test sensitivities.

The Bureau of Laboratories is working closely with the San Diego County Laboratory and State of California's laboratory to help inform all parties of differences and similarities that are seen between testing and results obtained for the respective outbreaks.

How should a hepatitis A antibody assay resulting as "indeterminate" be interpreted and handled?

Indeterminate results do not indicate the IgM is negative. A test resulting as indeterminate should be repeated immediately. If the test results are indeterminate for a second time, the sample should be repeated with a different assay, or the patient should be retested in 2-4 weeks.

A follow-up total anti-HAV (both IgM and IgG) may be ordered if it is likely that the patient is identified and tested while the IgM antibody is declining. Some hospital and commercial laboratories may offer this testing, since the MDHHS Bureau of Laboratories does not currently offer total HAV antibody testing at this time.

Are all Anti-HAV IgM positive samples sequenced by the MDHHS Bureau of Laboratories (BOL)?

Yes. All positive Anti-HAV IgM samples should be forwarded to BOL for sequencing.

MDHHS currently does not offer official reports directly to submitters, but rather genotyping is performed to determine viral genotype and strain identification for epidemiologic purposes in effort to stop the spread of disease.

How should patients that are asymptomatic with positive antibody results be investigated?

Hepatitis A antibody assays are considered diagnostic and the clinician should treat the patient as they deem appropriate. The sample may be sent to the MDHHS Bureau of Laboratories for genotyping and confirmation of virus presence.