Laboratory Confirmation by IgM Serology and Questions and Answers

http://www.cdc.gov/mumps/lab/overview-serology.html#st3

Laboratory confirmation of mumps in previously vaccinated or previously infected individuals is challenging, and failure to detect mumps IgM in previously vaccinated persons has been well documented.

People with a history of mumps vaccination may not have detectable mumps IgM antibody regardless of timing of specimen collection.

The ability to detect IgM varies by vaccination status and is highest in unvaccinated persons (80%–100%) (Sakata et al. 1985), intermediate in one-dose vaccine recipients (60%–80%) (Briss et al. 1994; Narita et al. 1998), and lowest in two-dose vaccine recipients (13%–14%) (Bitsko et al. 2008; Rota et al. 2009).

Mumps exposure	IgM	IgG	Comments
Unvaccinated; no history of mumps	+	+ or −	IgM may be detected for weeks to months; low levels of IgG may be present at symptom onset
1–dose vaccine history	+ or −	Likely +	50% of serum samples collected 1–10 days after symptom onset were IgM-positive; 50%–80% of serum samples collected >10 days after symptom onset were IgM-positive
2–dose vaccine history	+ or −	Likely +	13%–15% of serum samples collected 1–3 days after symptom onset were IgM-positive*

*30%–35% of buccal samples collected 1–3 days after symptom onset were positive by real-time RT-PCR among persons with 2 doses of MMR (Bitsko et al. 2008; Rota et al. 2009)

Q: What type of serologic assay is recommended for mumps IgM testing?
A: Both EIA and IFA assays can perform well for diagnosis of primary mumps infection. Acute-phase mumps specimens may contain significant levels of mumps IgG, especially among persons with a history of 1 or 2 doses of MMR. The IFA format is particularly susceptible to interference by high levels of mumps IgG. Treatment of serum with an agent to remove human IgG antibody, such as Gullsorb or a similar IgG inactivation reagent, is necessary to avoid false-positive IgM test results.

Q: What serologic tests are used at CDC to diagnose mumps?
A: A capture IgM EIA (non-quantitative) that incorporates a recombinant mumps nucleocapsid protein as the antigen is used to detect mumps IgM. A commercial, indirect EIA (non-quantitative) is used for detection of IgG.

Q: What does a positive mumps IgG test result mean?
A: A single serum sample tested for mumps-specific IgG is not useful for diagnosing acute mumps infections. The presence of mumps-specific IgG, as detected using a serologic assay (EIA or IFA), does not necessarily predict the presence of neutralizing antibodies or protection from mumps disease.

Q: What is the protective neutralizing antibody titer for mumps?
A: There is no known protective level of neutralizing antibody (antibody titer) for mumps, and there are no other immune parameters that correlate with protection from mumps disease.

Q: What etiologic agents are likely to interfere with serologic assays for mumps (i.e., produce false-positive results)?
A: Parainfluenza viruses 1, 2, and 3, Epstein-Barr virus, adenovirus, and human herpesvirus 6 have all been noted to interfere with mumps serologic assays (Davidkin et al. 2005).

Q: My patient specimen was positive by the monspot test (Epstein-Barr) but also gave a positive result for mumps IgM antibody. What is the explanation for this?
A: The initial immune response to Epstein-Barr produces a polyclonal B cell stimulation; the antibodies are broadly reactive and can result in a positive mumps IgM result. However, the monospot test should be considered less susceptible to a false-positive result with serum collected from a true case of mumps.

Q: If the IgM result is negative and IgG is positive, can mumps be ruled out?

A: Absence of a mumps IgM response in a vaccinated or previously infected individual presenting with clinically compatible mumps ***does not rule out mumps*** as a diagnosis. A positive IgG result is expected among previously vaccinated persons. Older persons or foreign nationals with no history of mumps illness or vaccination may have detectable mumps IgG due to a previous subclinical infection.

Q: Can serologic tests differentiate between a recent or prior exposure to mumps virus and a response to mumps vaccine?
A: The presence of mumps-specific IgG indicates a recent or a prior exposure to mumps virus or mumps vaccine. Serologic tests cannot differentiate between an exposure to vaccine and an exposure to wild-type mumps virus.

Q: The acute-phase serum was negative for mumps IgM. Is it appropriate to collect additional serology specimens?

A: Unvaccinated persons or unknown vaccination history
If an acute-phase serum sample collected ≤3 days after parotitis onset in an unvaccinated person is negative for IgM, testing a second sample collected 5–7 days after symptom onset is recommended since the IgM response may require more time to develop. A second negative IgM result does not rule out mumps unless the IgG result is also negative.

B: Previously vaccinated persons
There is some evidence that serum collected ≥10 days after parotitis onset may improve the ability to detect IgM among persons who have received only 1 or 2 doses of MMR (Krause et al. 2007; CDC, unpublished data). However, persons with a history of mumps vaccination may not have detectable mumps IgM antibody regardless of the timing of specimen collection.

Q: What does a positive IgM result mean?
A: Mumps is confirmed by detecting mumps IgM antibody in serum samples collected as soon as possible after symptom onset. A positive IgM test result indicates current or very recent infection or reinfection. A positive IgM test result may also be observed following mumps vaccination.

Q: What are the recommendations for collection of a convalescent-phase serum sample?
A: A second (convalescent-phase) serum sample is collected about 2–3 weeks after the first (acute-phase) sample. A 4-fold increase in IgG titer between acute- and convalescent-phase samples, as measured in plaque-reduction neutralization assays or similar quantitative assays, or a seroconversion from negative to positive from acute to convalescent phase, as determined by EIA, is considered a positive diagnostic result for mumps. ***Paired serum samples from vaccinated persons, even if appropriately timed, may not show a rise in IgG titer***.

Q: Why is it difficult to demonstrate a rise in titer (seroconversion) from persons with a history of vaccination?
A: In vaccinated persons, the existing IgG will begin to rise soon after exposure and infection. At the time of symptom onset and collection of the acute-phase serum sample, IgG may already be quite elevated, which would obviate the 4-fold rise in titer expected when comparing acute- and convalescent-phase titers. For this reason, it is recommended to obtain the acute-phase specimen as soon as mumps infection is suspected.

Q: What is the experience at CDC with paired serum samples from previously vaccinated persons with mumps?
A: A 4-fold rise in IgG titer is rarely demonstrated between paired serum samples from persons who have received one or two doses of MMR vaccine. In our experience using a plaque neutralization assay, we have only seen a 4-fold rise in neutralizing antibody titer between paired samples in persons who were also IgM positive.

Q: What method can be used to demonstrate a 4-fold rise in IgG titer between properly spaced serum samples?
A: In order to measure a 4-fold rise in titer, it is recommended that laboratories use an established assay or test protocol that provides a quantitative or semi-quantitative measure of antibody. Moreover, it is recommended that laboratories pay close attention to the parameters of the testing protocol to make certain that the acute- and convalescent-phase serum specimens fit the criteria of the protocol in terms of both timing of collection and parameters of data analysis. When a qualitative EIA is used for IgG titer determination, it is essential to do end-point titrations to determine if there is a 4-fold or greater difference in titers between acute- and convalescent-phase serum samples