

DFA unreliable in H1N1 testing in critically ill patients

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Direct Immunofluorescence Assay (DFA) testing for H1N1 influenza ("swine flu") is unreliable in ICU patients, according to a new study from Stanford University. Multiple methods exist for diagnosing influenza, but data on the utility and accuracy of these tests for H1N1 are still emerging, given the relatively recent onset of the epidemic.

"Our findings suggest that in patients with severe H1N1 influenza, in whom rapid and precise diagnosis would be most important, DFA unfortunately does not perform well. This is in contrast to less severely ill patients, where DFA appears to be quite reliable." said Chanu Rhee, M.D., a physician at Stanford University School of Medicine and lead author of the study.

The results will be presented at the ATS 2010 International Conference in New Orleans.

While PCR testing has emerged as the most sensitive and specific test for diagnosis of H1N1 influenza, availability of the test and turn-around time often limit its clinical usefulness. DFA testing is used at many institutions as an accurate and rapid means of diagnosing influenza. DFA for influenza uses a fluorescent dye attached to [antibodies](#) that bind to flu particles. If influenza is present, the antibodies will bind to viral antigens and a bright glow can be seen in the sample using a special microscope.

Several months after the H1N1 pandemic began, Dr. Rhee and

colleagues at Stanford University noticed a trend at their institution that critically ill patients with H1N1 influenza more commonly had negative DFA results than those who were less severely ill. To further investigate this observation, they reviewed the records of all patients who were admitted to the Stanford University Hospital between May 20, 2009 and January 30, 2010 with H1N1 influenza. All patients were confirmed for H1N1 influenza through either PCR or viral culture, and underwent DFA testing on a [respiratory tract](#) sample. During the research period, 19 patients were admitted to the ICU; 11 required mechanical ventilation and six died of respiratory failure.

To their surprise, Dr. Rhee and colleagues found that while DFA was a fairly accurate tool for diagnosing H1N1 in non-critical cases, it was not at all accurate for patients in the ICU. Just five of the 19 ICU patients (26 percent) had positive DFAs for H1N1 infection (four by nasopharyngeal swab, one by bronchoalveolar lavage), whereas 27 out of 33 non-ICU patients (82 percent) had a positive DFA test. The median time to first DFA was seven days in the ICU patients and three days in the non-ICU patients. Of the 31 respiratory tract samples in the ICU patients that were positive as determined by PCR, only 10 were concomitantly positive by DFA.

"For the non-ICU patients, the sensitivity of DFA was fairly good and correlated with previously published values. However, we found DFA to be significantly less sensitive in critically ill patients—those with severe respiratory distress requiring mechanical ventilation or a high degree of respiratory support in an ICU setting," said Dr. Rhee. "Interestingly, none of the DFA samples taken from the 18 endotracheal aspirates (secretions taken from the breathing tube on patients on a mechanical ventilator) were positive, despite the presence of virus detected by PCR or by bronchoalveolar lavage."

Dr. Rhee and colleagues were surprised by their findings, as they

expected that severely ill patients would have a higher burden of viral disease, leading to easier detection. "We would have also expected that samples taken from endotracheal aspirates, where the secretions are coming from lower down the respiratory tract, would have a higher likelihood of being positive, but this was not the case," said Dr. Rhee.

One possible explanation for the poor performance of DFA in ICU patients is that it is an over-exuberant host inflammatory response, rather than high viral load, that is responsible for severe disease. However, it remains unclear why certain patients develop severe respiratory failure from H1N1 while others with similar risk factors develop only mild symptoms.

If confirmed by further research, these findings have important ramifications. "This study reinforces the fact that patients with suspected H1N1 [influenza](#) who are severely ill should be placed in respiratory isolation and receive antiviral treatment without delay, even if DFA testing is negative" said Dr. Rhee. "This includes patients with a negative DFA from lower respiratory tract samples. Furthermore, all critically ill patients with suspected H1N1 should have PCR testing done to confirm the diagnosis, as PCR is significantly more sensitive than DFA, though not perfect either."

"The next logical step would be analyzing data from a much larger pool of patients from different institutions to confirm these findings," said Dr. Rhee.

Provided by American Thoracic Society

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