

Performance of rapid SOFIA Influenza A+B test compared to Luminex x-TAG respiratory viral panel assay in the diagnosis of influenza A, B, and subtype H3

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ABSTRACT

Influenza is an acute respiratory illness caused by influenza A or B viruses that occur in outbreaks, mainly during the winter season. Rapid laboratory diagnosis of influenza can help guide the clinical management of suspected patients effectively. Clinical sensitivities and specificities of the rapid influenza diagnostic tests have varied considerably in the literature. Most of these studies are evaluated using previously frozen or stored specimens that had previously tested positive. This study compares the performance of the rapid SOFIA Influenza A+B test to nucleic acid multiplex test x-TAG respiratory viral panel (RVP) assay in freshly collected nasal aspirates and measured simultaneously by both assays. Retrospective data from 1649 nasal aspirates (September 2014 to May 2015) collected from adults as well as from children tested simultaneously by both rapid SOFIA Influenza A+B FIA immunofluorescence (Quidel, San Diego, CA) and qualitative nucleic acid multiplex RVP assay X-TAG Luminex technology (Luminex, Austin, Texas, USA) were analyzed. Concordance, and analytical sensitivity and specificity were evaluated for influenza A, subtypes H1 and H3, and influenza B. Prevalence for influenza A by RVP was 15%, for subtype H3 it was 11.2%, and for influenza B, 2.9%. None of the aspirates were positive for influenza A subtype H1. SOFIA Influenza rapid test demonstrated good specificity and low sensitivity compared with a nucleic acid test for influenza A, subtype H3, and for influenza B. SOFIA Influenza A + B test performed well in providing a rapid diagnosis, however, confirmatory molecular testing is recommended for negative test results. Re-evaluation of test performance should be periodically carried out during outbreaks with the emergence and circulation of new influenza strains.

INTRODUCTION

An average of 200,000 hospitalizations and 35,000 deaths are attributed to influenza every year in the USA,¹ with annual direct medical costs estimated by the Centers for Disease Control at \$8.3 billion. With such a sizable toll in terms of morbidity, mortality, and monetary cost, it is little wonder that prevention, early diagnosis, and treatment of influenza are heavily prioritized in this country. Early

detection and treatment of influenza has been proven to improve outcomes in influenza patients. Further, early detection prevents unnecessary testing and treatment, and it helps prevent the unwarranted use of antibiotics in cases of viral respiratory illness, reducing healthcare expenses and development of antimicrobial resistance.²⁻³ For these reasons, a number of rapid influenza detection tests have been developed in recent years. Many of these tests are CLIA waived, conferring the added benefit of convenient, inexpensive, quick results in the physician's office. Unfortunately, though most of these tests show strong specificity, they are generally notorious for poor sensitivity. One meta-analysis of 159 RIDT (Rapid Influenza Diagnostic test) studies evaluating 26 RIDTs showed pooled specificities of 98% but pooled sensitivities of only 62.3%.⁴ With its SOFIA Influenza A + B test developed in 2011, Quidel aimed to address these accuracy issues by employing europium based immunofluorescence to boost sensitivity and a digital detection system to reduce subjectivity in interpretation of results. Results boasted by the company of, respectively, 99%/88% sensitivity for influenza A and B, and 96%/96% specificity, sound quite promising, and several studies have independently borne out these results.⁵⁻⁷ However, most of these previous studies have incorporated restrictive criteria for patient selection to control variables such as patient age and symptomatology. We suspected these stringent criteria might not accurately reflect actual clinical use both in terms of patient population and specimen handling. Therefore, in this study, we sought to examine SOFIA test performance in real world conditions to evaluate the sensitivity and specificity of the SOFIA test as actually utilized in current clinical practice. We evaluated the performance of the SOFIA Influenza A+B fluorescence immunoassay, using retrospective analyses on all fresh specimens received, which were confirmed by RT-PCR for the presence of influenza A and B viruses, in a tertiary care medical center.

METHODS

Retrospective data of 1649 nasal aspirates collected from children as well as adults, who had



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been tested for influenza between September 2014 and May 2015, were analyzed. There were 829 (50%) samples from females and 820 (50%) from males. Median age of patients was 57 years, with 252 (15%) under 21 and 1397 (85%) over 21 years of age. Because we sought to establish real-world accuracy of these tests, no clinical inclusion or exclusion criteria were applied. All of the specimens had been concurrently tested by both SOFIA A+B immunofluorescence (Quidel, San Diego, California, USA) and by qualitative nucleic acid multiplex RVP assay X-TAG Luminex technology (Luminex, Austin, Texas, USA). We assessed the accuracy of the SOFIA RIDT in detection of influenza A, influenza A subtype H3 and influenza B, using the PCR-based XTAG RVP test as the gold standard. Results were then analyzed for precision statistics, using EP Evaluator (Data Innovations, LLC).

RESULTS

Sensitivity, specificity, positive predictive value (PPV), and negative predictive value (NPV) of SOFIA A+B were calculated for each virus in comparison to RVP. For influenza A, SOFIA A showed overall sensitivity of 41.5% and specificity of 99.2% (table 1). For influenza A, subtype H3, SOFIA showed a sensitivity of 51.1% and a specificity of 99.0% (table 2). For influenza B, SOFIA showed a sensitivity of 37.5% and a specificity of 99.5% (table 3). Sensitivity of SOFIA A for patients under 21 years was 53.8%. There was no statistically significant difference in sensitivity between SOFIA RIDT for influenza A and SOFIA RIDT for influenza B (p=0.8824) nor between SOFIA influenza A and influenza A subtype H3 (p=0.2595) by Fisher exact test. Additionally, SOFIA influenza A sensitivity did not significantly differ between patients older than 21 years and patients under 21 years of age (p=0.4603).

DISCUSSION

The SOFIA RIDT test showed excellent specificity but rather poor sensitivity for detection of influenza A, influenza subtype H3, and influenza B. In fact, the sensitivity observed for each of these influenza variants was significantly lower than that observed in most previous studies of SOFIA A+B. One important factor contributing to this reduced sensitivity may be our substantially older patient population (median age 57 years). Most previous studies⁶⁻⁸ have involved a preponderance of pediatric patients. For instance, Lewandrowski *et al*⁶ investigated a patient

Table 2 Performance characteristics of SOFIA Influenza A RIDT (Rapid Influenza Diagnostic test) and respiratory viral panel molecular assay (subtype H3)

Influenza A (subtype H3)	Percentage	95% CI
Sensitivity	51.1	43.6 to 58.5
Specificity	99.0	98.4 to 99.5
Positive predictive value	87.0	79.2 to 92.7
Negative predictive value	94.1	92.8 to 95.2
Agreement	93.7	
Prevalence	11.2	

population of which 92% were under the age of 21 years. By contrast, only 15% of our participants were under 21 years of age. This may be an important factor since children have been proven to shed influenza virus more plentifully and for a longer time than adults.⁹ Further, other rapid influenza tests have also demonstrated relatively poor sensitivity in older adults.¹⁰

Still, the older age of our patient population likely does not entirely account for the discrepancy between our demonstrated sensitivities for SOFIA and those of previous investigations. After all, in our study, though there was higher sensitivity of SOFIA for influenza A in patients under 21 years (53.8%) versus over 21 years of age (40%), this difference was not statistically significant (p=0.4603). Moreover, Hazelton *et al*¹¹ also examined a predominantly older population (median age 56 years) and showed 72.4% sensitivity for influenza A by SOFIA—significantly higher than ours. In this study season, we also could not find any positive test result for influenza A subtype H1. In the Massachusetts department of public health weekly’s influenza report it shows that in the same period there were only four cases of subtype H1 reported in the State laboratory.¹²

Aside from patient age, another important factor distinguishing our study from previous studies is the lack of control for patient selection. Previous studies have required documented symptoms of flu, such as high fever or cough, as inclusion criteria.⁶⁻⁷ Our study did not specify the need for particular symptoms nor did it specify duration of illness. This is likely to be a significant factor because viral shedding is known to be markedly higher in early influenza, when symptoms first begin to manifest, than in later phases.¹³⁻¹⁴ In our study, the decision to have the SOFIA

Table 1 Performance characteristics of SOFIA Influenza A RIDT (Rapid Influenza Diagnostic test) and respiratory viral panel molecular assay

Influenza A	Percentage	95% CI
Sensitivity	41.5	35.2 to 47.9
Specificity	99.2	98.6 to 99.6
Positive predictive value	90.3	83.2 to 95.0
Negative predictive value	90.6	89.0 to 92.0
Agreement	91.0	
Prevalence	15.0	

Table 3 Performance characteristics of SOFIA Influenza B RIDT (Rapid Influenza Diagnostic test) and respiratory viral panel molecular assay

Influenza B	Percentage	95% CI
Sensitivity	37.5	24.0 to 52.7
Specificity	99.6	99.1 to 99.8
Positive predictive value	72.0	50.6 to 87.9
Negative predictive value	98.1	97.3 to 98.7
Agreement	97.7	
Prevalence	2.9	

test performed was entirely at the discretion of the submitting clinician, so it is likely that some of our specimens came from patients in the late phases of illness, when RVP would still detect virus, but shedding would be too low for detection by SOFIA.

Based on the results of our study, it appears that the low sensitivity of SOFIA for detection of influenza points out two important issues. First, SOFIA may not be as effective for diagnosing influenza in adult patients as it is in children. Second, at least with regard to the clinical clientele our laboratory serves, the testing may be improperly utilized. Healthcare professionals cannot solely rely on SOFIA RIDT test in adults and should follow-up with more sensitive molecular methods, when clinical findings are not concordant.

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Contributors LVR designed the project, retrieved retrospective data from IS database, contributed and reviewed the article. WS performed the analyses on the data, wrote the manuscript and collected relevant literature.

Competing interests None declared.

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