COMPARISON of Alere™i, Xpert® Flu+RSV Xpress, and Sofia Influenza A+B FIA FOR INFLUENZA A & B DETECTION IN POINT OF CARE SETTING

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ABSTRACT

INTRODUCTION
Three point of care (POC) waived Flu A/B testing platforms were evaluated for use at our outreach clinic sites. Methods evaluated were: Alere™i Influenza A & B rapid molecular isothermal NAAT, Xpert® Flu+RSV Xpress automated RT-PCR, and Quidel Sofia® Influenza A+B FIA. Comparison included accuracy and hands-on time.

METHODS
68 samples were evaluated. All samples were nasopharyngeal swabs (NPSW) collected in BD viral transport media (VTM), stored at 4C. Primary patient testing was performed on either the BioFire (BF) FilmArray Respiratory Panel or the Cepheid Xpert FLU/AB/RSV assay. The maximum time from collection to evaluation was 72h. Testing on all three platforms was performed on the same day. Alere is CLIA waived for direct nasal swabs, CLIA moderate for swabs in VTM. Sofia is CLIA waived for nasal swabs and NPSW tested directly, and NPSW in VTM. Xpress is CLIA waived for NPSW in VTM. All three assays were run according to manufacturers' instructions.

•Evaluation Criteria: Samples had to be positive with at least two systems (BF, Alere, Xpert, Sofia) to be included in the study. Several operators performed testing and evaluated hands-on time for each assay.

RESULTS
•Ten samples were negative for Flu A,B,RSV; 4 samples were Flu A-, B-, RSV+; and 6 samples were Flu A,B,RSV- and positive for other targets. Twenty-seven samples were Flu A+; 21 samples were Flu B+. Of the 68 samples, 22 were discrepant on at least one of the 3 systems. Four of the discrepant samples positive for Flu A in BF and negative with all three comparison assays were excluded from further evaluation. Forty-six samples agreed across all systems: 20 Flu A/B-, 16 Flu A+/ B-, and 10 Flu B+/ A-.

•Sensitivity: Alere 82.6% A+, 81% B+; Xpress 100% Flu A,B; Sofia 69.6% A+, 47.6% B+

•Specificity: 100% for all platforms

•Hands-on/testing times for each platform are: Sofia 2.5 min set up time, 15 min incubation and read totaling18 min. Alere 15 minutes directly from swab and 18 min if testing swab in VTM; about 2 min hands-on time. Xpress 1-2 min. set up time, 60 min incubation.

CONCLUSION
Timely testing at the POC with a rapid accurate influenza test will directly impact appropriate treatment. The Xpert Xpress platform had the highest sensitivity (100%) with results in 1 hr. The sensitivity of Alere was 82.6% and 81% for Flu A and B respectively, results in 18 min. Sofia had the lowest sensitivity with 69.6% and 47.6% for Flu A and B respectively; results in about 18 min. When considering rapid systems, accuracy, hands-on time, incubation time, and interface capability should all be considered for the final system selection.

INTRODUCTION

Three POC waived Flu A/B testing platforms were evaluated for use at our outreach clinic sites.
Methods evaluated were:
Alere™ Influenza A & B rapid molecular isothermal NAAT,
Xpert® Flu+RSV Xpress automated RT-PCR,
Quidel Sofia® Influenza A+B FIA.
Comparison included accuracy and hands-on time.

POC PLATFORMS

Alere i ™ Influenza A & B
•Technology: NEAR: Nicking Endonucleases Amplification Reaction
•Method: Molecular
•Approved specimen: Nasal swab, NP swab (with or w/o VTM)
•Assay time: about 18 mins
•CLIA Waived: **Yes**

Xpert® Flu+RSV Xpress
•Technology: RT-PCR
•Method: Molecular
•Approved specimen: NP swab, nasal wash in VTM
•Assay time: about 60 mins
•CLIA Waived: **Yes**

Quidel Sofia® Influenza A+B
•Technology: Fluorescent Immunoassay
•Method: Lateral flow
•Approved specimen: Nasal swab, NP swab, aspirate/wash
•Assay time: 15 mins incubation
•Assays performed in Walk Away mode
•CLIA Waived: **Yes**

METHODS

60 samples were to be compared with three POC flu assays: 20 A/B neg, 20 A+, and 20 B+. Specimen type: Fresh N/P swab specimens submitted in Universal Transport Media for routine patient testing on either BioFire RP Panel or Cepheid Xpert® Flu A/B, RSV panel. Some negative samples were to include samples positive for other non Flu A/B targets.

68 samples were evaluated. All samples were nasopharyngeal swabs (NPSW) collected in BD viral transport media (VTM), stored at 4°C.
The maximum time from collection to evaluation was 72h. Testing on all three POC platforms was performed on the same day.
All three assays were run according to manufacturers' instructions. Several operators performed testing and evaluated hands-on time for each assay.

Evaluation Criteria: Samples had to be positive with at least two systems (BF, Alere, Xpert, Sofia) to be included in the study.

RESULTS

•10 samples were negative for Flu A,B,RSV
• 4 samples were Flu A-, B-, RSV+
• 6 samples were Flu A,B,RSV- and positive for other targets.
•27 samples were Flu A+
•21 samples were Flu B+.

•Of the 68 samples, 22 were discrepant on at least one of the 3 systems. Four of the discrepant samples positive for Flu A in BF and negative with all three comparison assays were excluded from further evaluation.

• **Forty-six samples agreed across all systems: 20 Flu A/B-, 16 Flu A+/ B-, and 10 Flu B+/ A-.**

Total Results

n=64	Alere		Xpress		Sofia	
	A+	B+	A+	B+	A+	B+
TP	19	17	23	21	16	10
TN	41	43	41	43	41	43
FP	0	0	0	0	0	0
FN	4	4	0	0	7	11
TOTAL	64	64	64	64	64	64

RESULTS

Sensitivity / Specificity

	Alere		Xpress		Sofia	
	A+	B+	A+	B+	A+	B+
Sensitivity	82.6%	81%	100%	100%	69.6%	47.6%
Specificity	100%	100%	100%	100%	100%	100%
PPV	100%	100%	100%	100%	100%	100%
NPV	91.1%	91.5%	100%	100%	85.4%	79.6%

Discrepant Samples

	Alere i	Xpress	Sofia
Total discrepant	12	4	22
% Discrepant	17.7%	5.9%	32.4%
Four samples were positive with BioFire FilmArray RP for Flu A and negative on all 3 POC platforms. Samples excluded. N=64			
% Discrepant after resolution	8 12.5%	0 0%	18 28.1%

DISCUSSION/CONCLUSION

Timely testing at the POC with a rapid accurate influenza test will directly impact appropriate treatment. The Xpert Xpress platform had the highest sensitivity (100%) with results in 1 hr. The sensitivity of Alere was 82.6% and 81% for Flu A and B respectively, results in 18 min. Sofia had the lowest sensitivity with 69.6% and 47.6% for Flu A and B respectively; results in about 18 min. It is important to note that antigen detection tests such as the Sofia FIA that do not have an amplification step are affected by the approx. 1:50 dilution that occurs when the N/P swab is placed into 3 mL of transport medium. Additional studies performed directly from N/P swabs at the Point of Care would likely improve the performance of the Sofia assay.
When considering rapid systems, accuracy, hands-on time, incubation time, and interface capability should all be considered for the final system selection.

ACKNOWLEDGEMENTS

We thank Alere, Cepheid, and Quidel for providing kits for this study. Also, thanks to my wonderful coworkers for helping me with this project. ☺