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Dicht bij het leven

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POINT OF CARE TESTING FOR THE DETECTION OF INFLUENZA VIRUSES: DOES SPEED COMPROMISE ANALYTICAL PERFORMANCE?

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Background

Respiratory tract infection may be caused by a number of different viruses and bacteria but influenza (INF) A and B viruses and respiratory syncytial virus (RSV) are collectively responsible for a majority of respiratory illnesses and can lead to severe complications. Rapid diagnosis is highly desirable to start quickly antiviral therapy and to manage outbreak control.

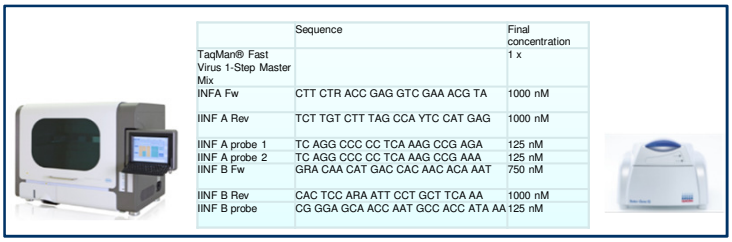
A new automated, qualitative point of care test (POCT), the cobas® Influenza A/B & RSV Nucleic acid test for use on the cobas® Liat® System, can detect INFA, INFB, and RSV RNA in nasopharyngeal swab specimens in about 20 minutes. The analytical performance was compared with our validated routine laboratory developed test (LDT), a reverse transcriptase real time PCR assay which targets the same genes (Ward *et al.*, J Clinical Virol, 2004).

Methods

33 external quality controls (INSTAND) were analysed following the manufacturer's instructions.
One INFA positive sample (H3N2) and one INFB were analysed during three days by three different operators to check the reproducibility of the system.

Fig. 1 Workflow Liat®:

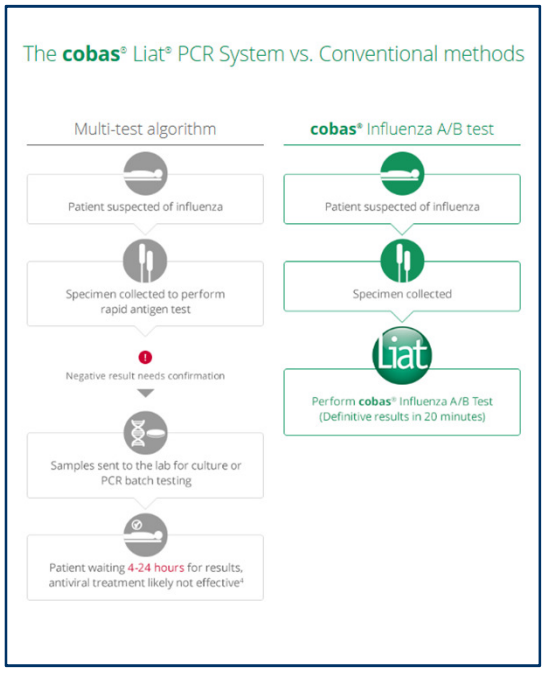
- ***E-swab medium is added to the assay tube using the transfer pipette***
- ***Assay tube is scannend***
- ***Assay tube is placed in the cobas® Liat ® System***
- ***Total hands-on time: < 1 min***



	Sequence	Final concentration
TaqMan® Fast Virus 1-Step Master Mix		1 x
INFA Fw	CTT CTR ACC GAG GTC GAA ACG TA	1000 nM
IINF A Rev	TCT TGT CTT TAG CCA YTC CAT GAG	1000 nM
IINF A probe 1	TC AGG CCC CC TCA AAG CCG AGA	125 nM
IINF A probe 2	TC AGG CCC CC TCA AAG CCG AAA	125 nM
IINF B Fw	GRA CAA CAT GAC CAC AAC ACA AAT	750 nM
IINF B Rev	CAC TCC ARA ATT CCT GCT TCA AA	1000 nM
IINF B probe	CG GGA GCA ACC AAT GCC ACC ATA AA	125 nM

Fig. 2 Workflow laboratory developed test method:

- ***Extraction on the MagNA Pure LC2 Extraction platform: 60 min***
- ***Preparing of the mastermix: 15 min***
- ***qPCR on the Rotor-Gene Q: 90 min***
- ***Total hands-on time: 30 min***



Results (2)

Accuracy

- The 33 external quality controls included six different strains of INFA (H1N1, (H1N1)pdm2009, H3N2, H3N2 (drift variant 2015), H5N8, H7N9) and 4 different strains of INFB. 20 samples were positive for INFA, 11 for INFB and 2 were negative.
- There was a 100% agreement with the reference method. No sample was inhibited.

Reproducibility

- The two positive samples gave the same result on all three days.

Conclusion

The need for batching the samples and a turnaround time of minimum one day are the main disadvantages of our current method. The high speed of the cobas® Influenza A/B & RSV Nucleic acid test does not compromise the analytical performance and is very easy to perform with almost no hands-on time (less than 1 minute). The system can easily be used as a POCT system but is less suitable for a high throughput laboratory as the test is performed one by one. The possibility to perform immediately bedside molecular diagnostics testing, will have an improved impact on clinical decision making and on the organisation of the health care institute.

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