Health Technology Assessment in India
Department of Health Research, MoHFW

Health Technology Assessment of various RT-PCR kits/methods for the diagnosis of Influenza A/H1N1pdm09 virus in all age group patients in India.

INTRODUCTION

Lower respiratory tract viral infections, including those due to influenza, are among the most common infectious diseases in humans and they are associated with significant morbidity and mortality. Seasonal influenza viruses infect 5–15% of the human population each year, resulting in ~500,000 deaths worldwide. A significant number of severely ill patients infected with H1N1pdm09 requiring intensive care and mechanical ventilation for severe viral pneumonia.

Nucleic acid tests are sensitive and specific and provide a rapid diagnosis, making them invaluable for patient and outbreak management. Real-time polymerase chain reaction (PCR) can be considered as gold standard for detection of influenza viruses due to its high sensitivity and specificity. Real time RT-PCR developed by WHO is considered as gold standard method for influenza A H1N1pdm09 diagnosis from Nasal/throat swabs.

Currently in India, suspected patients are screened by clinician and prescribing Oseltamavir drug without waiting for test report. Government of India recommended testing of sample from suspected patients of category C only. Indiscriminatory use of anti influenza anti-viral drug may develop resistance. Limited labs are testing H1N1pdm09 virus in country and have not been able to effectively implement at large programs due to lack of adequate infrastructure, trained manpower and limited resources and high costing of kits.

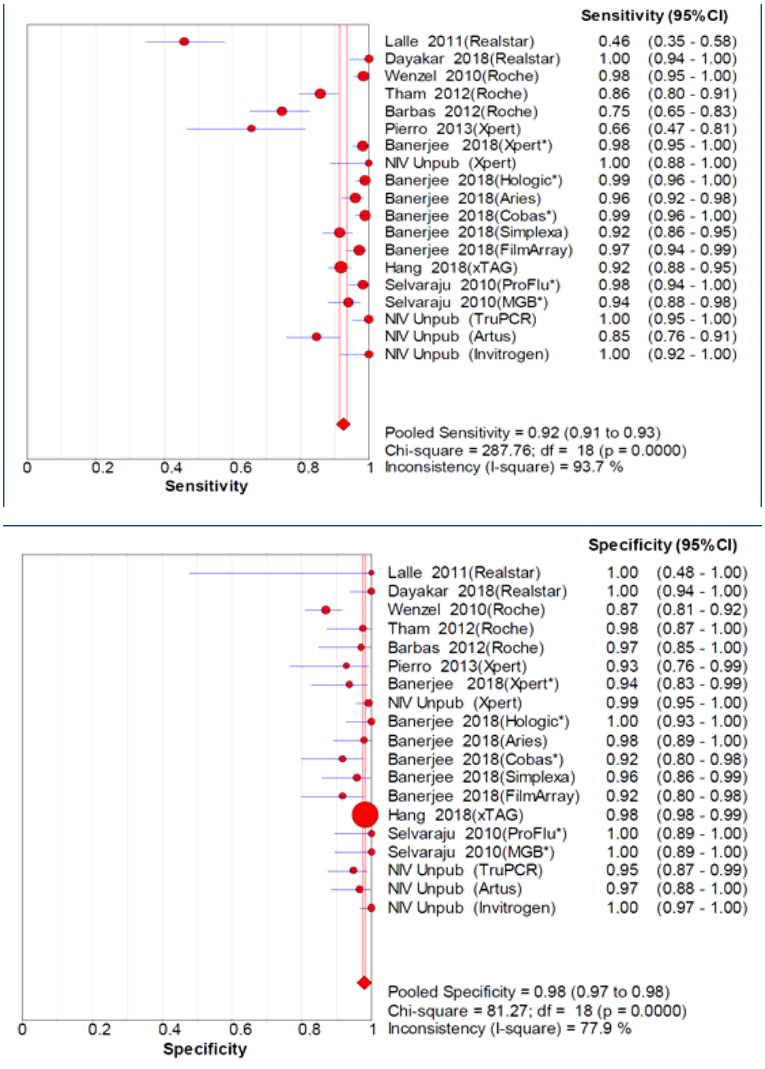

Figure 1: Forest plot for sensitivity (A) and specificity (B) of each individual study for overall kit, the pooled estimate are represented by diamond and the horizontal lines represent 95% confidence intervals (CI).

RECOMENDATIONS

1. In view of highest diagnostic accuracy (100% sensitivity and 100% specificity) among all the kits evaluated in this study, invitrogen kit is recommended for diagnosis of Category C patients for influenza A/H1N1pdm09 virus from clinical samples with an incremental cost of 355 Rs/test.

2. Diagnostics of H1N1in India, is currently being provided by ICMR-VRDL and NCDC network and all these centres are using real time PCR based technique with Invitrogen kits. The present study reconfirms the fact Invitrogen kit is most cost effective kit for H1N1 diagnostics with no additional burden to the healthcare system.

RATIONALE OF THE STUDY

Indiscriminatory use of anti influenza anti-viral drugs may develop resistance. Limited labs are testing H1N1pdm09 virus in country and have not been able to effectively implement at large programs due to lack of adequate infrastructure, trained manpower and limited resources and high costing of kits. Sensitivity, specificity and cost of different molecular tests exhibits huge variation. The purpose of this assessment was to appraise the current evidence for the clinical effectiveness (in terms of sensitivity and specificity) and cost-effectiveness of different RTPCR kits against CDC/WHO real time RT-PCR for diagnosis of influenza A/H1N1pdm09 in India.

Target*		Invitrogen Kit	Qiagen kit	TruPCR kit	Cepheid kit
Cost (Rs) Per Sample		2015	1902	1660	4342
Ease of doing		Easy	Easy	Easy	Easiest
Turnaround time		4 hours	4 hours	4 hours	2 hours
Samples in one go		29 Samples	34 or 46 samples, depending upon the rotor used.	29 Samples	1 to 4 samples, depending upon the machine module.
Operational Feasibility		Open system Existing labs equipped with RT-PCR machine	Open system Can be used in existing facilities.	Open system Can be used in existing facilities.	Closed system Health system will need to buy new RT-PCR machines, if this kit is introduced.
Influenza A	Sensitivity (95%CI)	100 (91-100)	84 (76-90)	100 (95-100)	100 (88-100)
	Specificity (95%CI)	100 (96-100)	96 (88-99)	94 (87-97)	99 (95-99)
H1N1	Sensitivity (95%CI)	100 (91-100)	94 (85-98)	94 (84-98)	93 (78-93)
	Specificity (95%CI)	100 (96-100)	98 (94-99)	100 (96-100)	100 (96-100)
H3N2	Sensitivity (95%CI)	100 (85-100)			63 (38-81)
	Specificity (95%CI)	100 (97-100)			100 (97-100)
B	Sensitivity (95%CI)				96 (83-99)
	Specificity (95%CI)				100 (96-100)

Table 1: Summary table for key findings of the study

METHODOLOGY

Clinical Effectiveness (Sensitivity & Specificity) Literature Review

The research methodology was designed using PRISMA-P (Preferred Reporting Items for Systematic reviews and Meta-analyses Protocols) statement guidelines including the preparation of a pre-specified protocol and analysis plan.

Primary Data Collection

Primary data were collected from in-house reports (NIV data), four (n=4) kits evaluated during the financial year 2018-2019.

Validation and costing Study

From four different companies' influenza diagnosis molecular kits [Thermo Fisher Scientific (Invitrogen); TRUPCR H1N1 kit (3BBlackBio Bhopal); Qiagen artus Infl./H1 LC/RG RT-PCR Kit; and Cepheid Xpert® Flu kit] were evalauetd and compared with CDC/WHO gold standard kit.

KEY FINDINGS

- **Literature Review:** Diagnostic accuracy of the kits for detection of H1N1 showed huge variation. The sensitivity of different kits varied from 76% to 100% whereas the specificity of different kits varied from 67% to 100% among the studies included in the review. Even for the same kit, there was huge variation between different studies both for sensitivity and specificity.
- **Validation Study**: Invitrogen kit exhibited the highest sensitivity and specificity for detection of H1N1 among 4 kits evaluated in validation study.
- **Costing Study**: Cephied kit was most expnsive with cost/test of Rs.4342, whereas TRUPCR was least expensive with cost/test of Rs. 1660.
- **Operational Feasibility**: Integrated Disease Surveillance Programme (IDSP) assisted lab network of 12 Laboratories are providing laboratory support in terms of testing, providing viral transport medium and diagnostic reagents. The laboratory network of ICMR-VRDL (30 labs) has also been activated to test for H1N1 cases. All these labs are currently using real time RT-PCR test for diagnosis of influenza H1N1 using Invitrogen kit. The present study reconfirms that Invitrogen kit is most cost effective kit for diagnosis of influenza H1N1 with an incremental cost of 355 Rs/test. As the kit uses an open system, it could be used in any real time PCR platform. Already being used in the system, the staff is trained in using the kit also. Therefore, there will be no addional burden in terms of procuring new platforms, or training of staff and cost of kits.
- **Cost Effectiveness**: If only H1N1 detection (not considering H3N2 and Influenza B) is considered for deciding the cost effectiveness, TRUPCR kit dominated over Qiagen and Cephied kit with least cost (1660 Rs/test) and highest accuracy (sensitivity 94%, specificity 100%) among these three kits, and thus Qiagen and Cephied were excluded from cost-effectiveness analysis. While comparing the TRUPCR kit and Invitrogen kit, later shows higher accuaray with an incremental cost of 355 Rs/test. If accuracy of overall kit is considered including Influenza A and Influenza B, Cephied kit can detect both subtypes in single reaction including the subtypes also. In addition to detecting both subtypes, the Cephied kit can provide results in least of the time. The kit is highly automated and thus easiest to be performed among the four kits. But at the same time, this kit works in a closed system model, that means the kit works only in a given platform and only 1-4 samples can be tested in one go, depending upon the machine module being used.

Acknowledgement:

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