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Detection of Respiratory Syncytial (RSV) and Influenza Viruses in Children with WHO Defined Pneumonia and Controls from Oshikhandass Village, Gilgit-Baltistan, Northern Pakistan from 2012-2014; Sensitivity and Specificity of Rapid Tests vs. PCR

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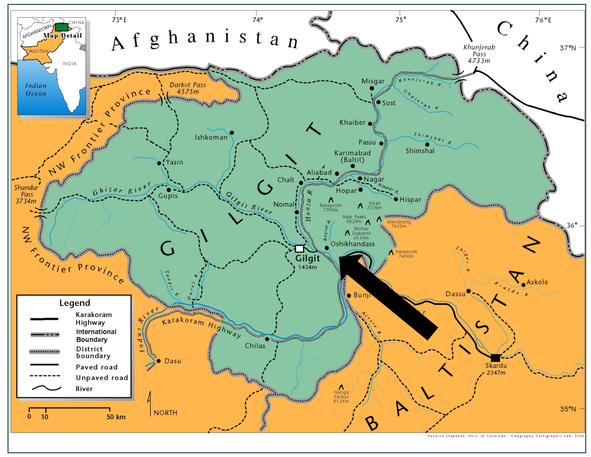


Introduction

Oshikhandass, a rural village in the Karakoram mountains of northeast Pakistan, was studied from 1989-1996 and 2011-2014. The village is in a region that experiences extreme winter and summer temperatures and is located 24 hours from reference labs.

Previous work demonstrated that pneumonia was the main cause of mortality among children under age 5 years in the village of Oshikhandass.[1] A follow-up study was completed in March 2014 to determine changes in pneumonia epidemiology, response to antibiotics and carriage of viral pathogens among the child population.

Methods and Materials

Female health workers trained in pneumonia management using WHO Integrated Management of Childhood Illness (IMCI) guidelines[2] conducted weekly surveillance of children under age 5 years. Each episode was classified by severity, based on WHO guidelines, treatment was given and referral done, if needed.

From 2012-2014, weekly surveillance of 1,176 children under age 5 years was conducted. Nasopharyngeal (NP) swabs (Copan® Flocked Ultra Mini Tip) were collected from 233 children with WHO-defined pneumonia and 66 controls for local rapid testing, followed by transport in PrimeStore® or viral transport medium for PCR testing at reference labs.

From Apr 2012 - Jan 2013, rapid tests were conducted using QuickVue® and from Jan 2013 - Mar 2014 using Sofia FIA®. For PCR testing, the Luminex® platform was used from Apr 2012 - Nov 2013 at AKU, Karachi, and the Taqman® Real Time PCR method from Dec 2013 - Mar 2014 at NIH, Islamabad. Reported sensitivity/specificity for QuickVue® and Sofia® are:

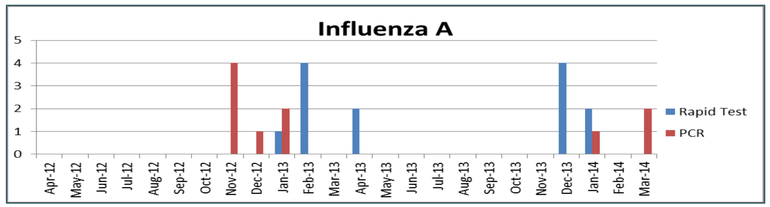
	QuickVue® vs. Cell Culture		Sofia FIA® vs. Cell Culture	
	Sensitivity	Specificity	Sensitivity	Specificity
Influenza A	83%	89%	97%	95%
Influenza B	62%	98%	90%	97%
RSV	92%	92%	100%	96%

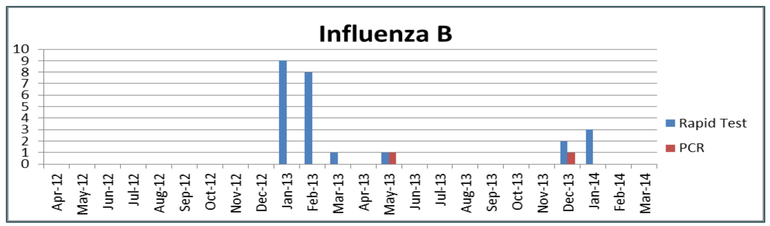
Results

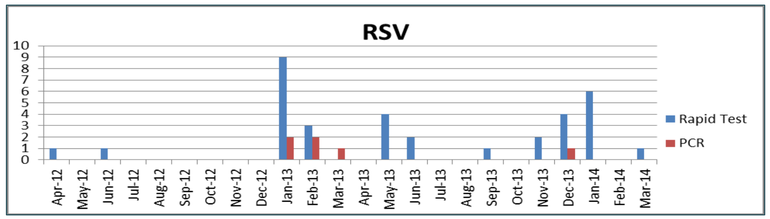
QuickVue® sensitivity was low for all 3 viruses. The number of influenza A and B positive results were too low to calculate sensitivity meaningfully. Sensitivity of the RSV Sofia® test was low when compared with Luminex® and Taqman®, while specificity for RSV was 92%, Influenza A 94%, and influenza B 91%.

Agreement between the PCR and Sofia-based methods was low for influenza (kappa≤0.152) and fair for RSV (kappa=0.273). Agreement between PCR and QuickVue was even poorer.

	QuickVue & Luminex N = 60		Sofia & PCR (Luminex+Taqman) N = 186+42=228	
	Sens(± 2*SD)	**Spec(± 2*SD)**	**Sens (± 2*SD)**	**Spec(± 2*SD)**
Inf A	0% (± 0)	100% (± 0)	50% (± 69)	94% (± 3)
	0/6	53/53	1/2	206/218
Inf B	N/A	100% (± 0)	100% (± 0)	91% (± 4)
	0/0	59/59	2/2	197/217
RSV	0% (± 0)	96% (± 6)	32% (± 13)	92% (± 4)
	0/11	47/49	16/50	163/178







Conclusions

Sensitivity for influenza was substantially lower than previously reported[3,4]. Possible reasons for poor sensitivity include low influenza circulation in the community and modest sample size. Other factors may be the inability to maintain controlled temperatures (15ºC - 30ºC) for the kits and equipment in winter and summer month field conditions, and remoteness of reference labs. Development of diagnostic tests that maintain stability and function over extended time periods and within a wide range of temperatures is essential to provide reliable results for appropriate and timely management of respiratory illness in resource poor environments.

Acknowledgements

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