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DETECTION OF RESPIRATORY INFECTION VIRUSES IN SYMPTOMATIC PATIENTS BY MULTIPLEX PCR - A PORTUGUESE STUDY

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INTRODUCTION

Acute respiratory tract infections (ARI) are the most widespread types of infections in adults and children and are responsible for considerable morbidity and mortality.

The aetiology of ARI is mostly viral, particularly severe in children, often requiring hospitalization. Clinical presentation is not indicative of the causal agent since clinical features are nearly indistinguishable, as well as does not suggest co-infections, involving either bacteria and virus or more than one virus.

A rapid diagnosis with a broad panel of respiratory virus is a helpful tool for clarifying the aetiology of infections and support therapeutic decision.

AIM

The aim of this study was to evaluate the prevalence of respiratory viral agents in patients presenting respiratory symptoms, by multiplex PCR.

METHODS

- ✓ From December 2009 to May 2011, a total of 65 respiratory samples (respiratory secretions, nasopharyngeal aspirate and bronchio-alveolar lavage) from patients presenting respiratory symptoms were analysed. Patients included 54 children aged between 15 days to 21 months and 1 with 10 years old; 10 adults, aged between 19 and 59 years old.
- ✓ Viral RNA or DNA was isolated using the automated NucliSens® EasyMAG™ (bioMérieux, France).
- ✓ The viral nucleic acids detection was performed by using multiplex PCR assays that detect and differentiate up to 9 viral agents and its 8 subtypes, including New FluA H1N1 (CLART® Pneumovir, GENOMICA; RespiFinder® SMART22, Patho Finder).

RESULTS

Overall, 47 samples (72.3 %) were positive for at least one viral agent (Table 1).

Virus	Samples	
	Nº	%
Bocavirus	8	17.0
RSVA	6	12.8
hRV	6	12.8
MPV	3	6.4
RSVB	2	4.3
PIV 3	2	4.3
ADV	1	2.1
Inf B	1	2.1
New Inf A (H1N1)	1	2.1
PIV 1	1	2.1
Corona 229E	1	2.1
RSVA/RSVB	3	6.4
hRV/RSVA	2	4.3
hRV/Bocavirus	2	4.3
Bocavirus/RSVA	1	2.1
Bocavirus/PIF 3	1	2.1
RSVA/Inf B	1	2.1
RSVB/hRV	1	2.1
MPV/hRV	1	2.1
ADV/PIF 3	1	2.1
ADV/Inf B/RSVA	1	2.1
ADV/PIF 3/hRV	1	2.1
Total	47	100.0

Table 1 – Frequency of respiratory viral agents in the analysed samples.

✓ There were 42 positive samples among children, and 5 among adults (Figure 1), being the virus prevalence shown in Figure 2.

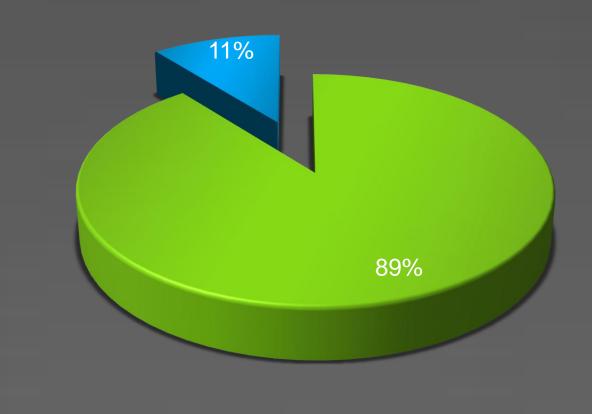


Figure 1 – Proportion of respiratory virus in children and adults.

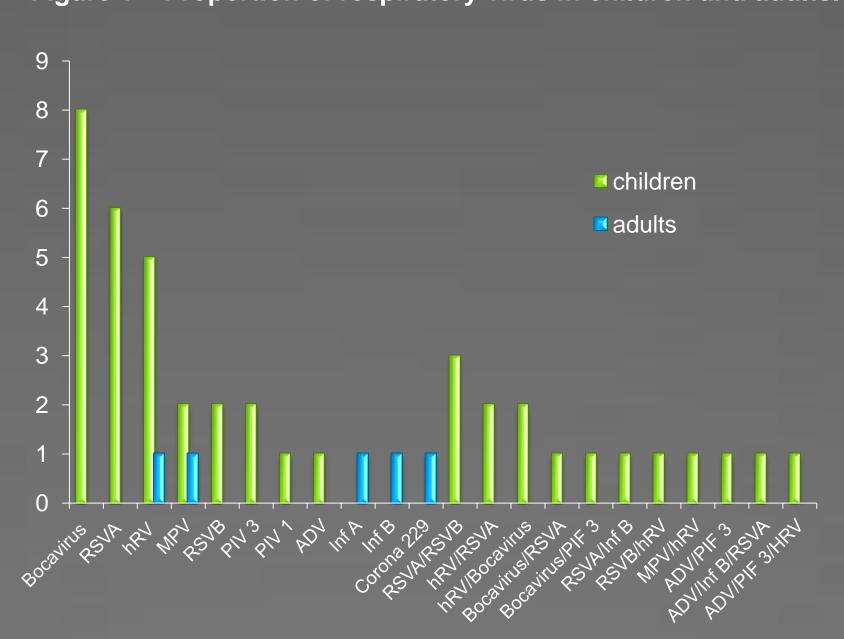


Figure 2 – Prevalence of respiratory virus in children and adults.

- ✓ Among the positive samples, 15 (32%) presented mixed infections and were all isolated from children (Figure 3).
- ✓ Eleven different combinations of mixed infections were observed, being the most frequent one RSV A and RSV B (n=3, 20%). In two samples, three viral agents were detected simultaneously (Figure 4).

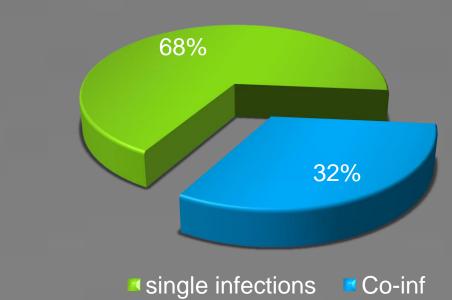


Figure 3 – Prevalence of single and mixed infections.

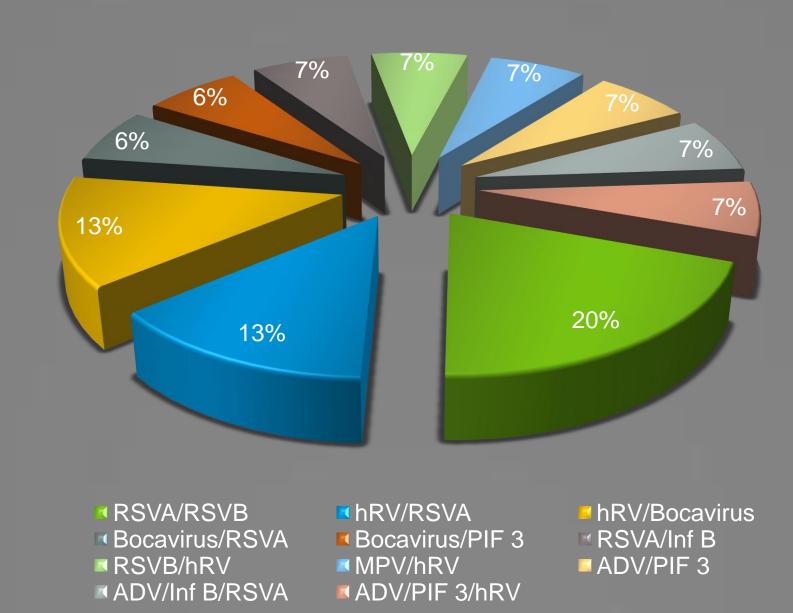


Figure 4 – Composition and distribution of the viral respiratory mixed infections.

- ✓ Mixed infections were observed mostly at age group under 2 years old, except for 1 children with 10 years old, presenting with severe asthma condition, infected simultaneously with hRV and RSVA.
- ✓ Bronchiolitis was the most frequent clinical presentation among children (n=23, 41.8%), and all these samples, except 3 were positive for viral respiratory agents. Among these samples, RSVA and hRV were the most frequently found (Figure 6).

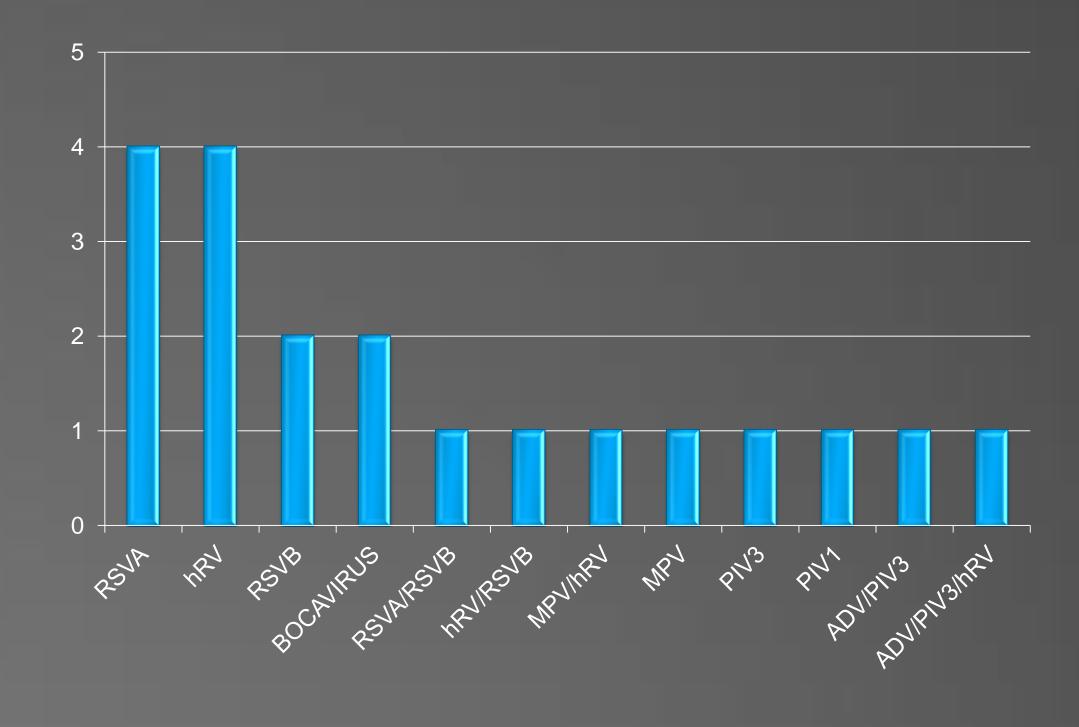


Figure 6 – Prevalence of respiratory virus in children with bronchiolitis.

CONCLUSIONS

The results obtained in this study emphasize:

- Respiratory virus are common among patients with ARI.
- Viral respiratory infections with more than one agent are also common.
- The use of the multiplex assay provides a userfriendly and high-throughput tool for an accurate diagnosis of ARI.

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