



Characterization of viruses causing Human Respiratory Infections via Genomic Identification for in vitro diagnosis. CLINICAL ARRAYS/CLART® PneumoVir.

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Introduction

Acute respiratory infections are among the most frequent diseases in our area, and they are one of the main reasons for consultation and hospitalization. Due to a great variety of possible pathogenic agents and the high frequency of co-infections, it is necessary to use diagnostic methods that allow multiple, sensitive, and rapid identification of these viruses.

Objective

Evaluate the capacity of CLINICAL ARRAYS/CLART®PneumoVir (CA-PV) for simultaneous detection of the 18 most frequent types of human viruses causing respiratory infections: influenza A H1-N1 virus (Flu-A, H1-N1), Flu A H3-N2, Flu-B, Flu-C, parainfluenza virus 1 (PIV-1), PIV-2, PIV-3, PIV-4A, PIV-4B, respiratory syncytial virus type A (RSV-A), RSV-B, human metapneumovirus A (hMPV-A), hMPV-B, rhinovirus, enterovirus, coronavirus, adenovirus, and bocavirus. Several Hospitals from Spain and UK have taken part in this multicenter study.

Method

Virus detection is performed via multiplex RT-PCR for amplification, and a new technology platform based on low-density micro-arrays (ArrayTube) for visualization. This technology allows simultaneous detection of viruses and any necessary control in order to guarantee the reliability of the results obtained. In order to determine the diagnostic parameters of the kit, a comparative evaluation of CA-PV against clinical diagnostic's current methods (nested-multiplex PCR/agarose gel, culture) was made. A total of 374 clinical specimens were tested, being a true positive result judged according to the concordance between both methods. All the discrepancies were validated with sequencing, and homemade nested-PCR.

Results

100% analytical sensitivity was obtained in the detection of 14 recombinant plasmids between 10 and 1000 copies.

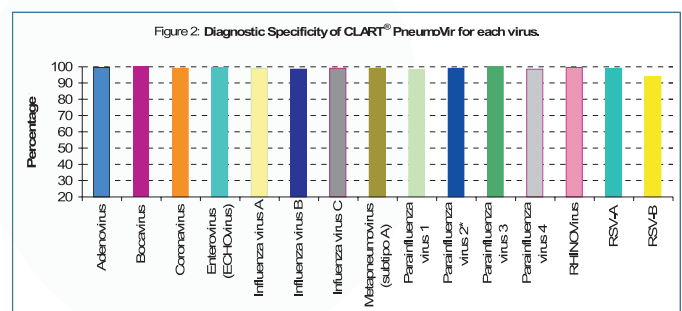
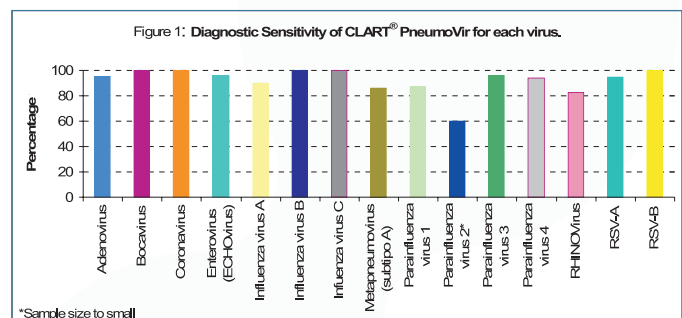
About the diagnostic sensitivity and specificity, the behaviour of each virus after the validation of 374 clinical specimens showed that most of virus has sensitivity higher than 90%, and specificity higher than 98%.

Conclusion

CA-PV is useful in the clinical setting for rapid screening and detection of a panel of respiratory viral pathogens based on the following facts: (i) excellent specificities and sensitivities; (ii) rapid and automatic procedure; (iii) simultaneous detection allowing the recognition of co-infections, (iv) hMPV-A, hMPV-B, Flu-AH1N1, Flu-AH3N2, PIV-4A, and PIV-4B subtypes can be differentially detected.

Identified Viruses N= 515 CHILDREN	ANTIGEN	CELL CULTURE	MULTIPLEX RT-PCR
RSV	91 (17,7%)	97 (18,8%)	122 (23,7%)
Adenovirus	18 (3,5%)	35 (6,8%)	35 (6,8%)
Parainfluenza I	3 (0,6%)	5 (1,0%)	9 (1,7%)
Parainfluenza 3	19 (3,7%)	24 (4,7%)	32 (4,7%)
Influenza Virus A	11 (2,1%)	19 (3,7%)	24 (4,7%)
Influenza Virus B	4 (0,8%)	6 (1,2%)	9 (1,7%)
Bocavirus	Indeterminate	Indeterminate	▶ 58
Rhinovirus	Indeterminate	Indeterminate	▶ 30 (5,8%)
Metapneumovirus	Indeterminate	Indeterminate	▶ 24 (4,7%)
Coronavirus	Indeterminate	Indeterminate	▶ 9 (1,8%)
TOTAL	146 (28,4%)	185 (36,2%)	▶ 312
Co-Infecciones ≥ 2 virus			▶ 36 (11,5%)

Table 1: Comparison of positive samples by method detection in children ≤ 5 years old with lower respiratory tract infection



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