Molecular characterization of respiratory syncytial virus during 2015-2016 season in Portugal


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Background
Respiratory syncytial virus (RSV) is one of the most frequent and important respiratory viral agent that causes respiratory infection complications in younger children and elderly. RSV has an autumn / winter seasonality. Genetic diversity in both of RSV A and B subtypes increased in last years with the spread of new genotypes. This study aims to describe the genetic variability of RSV during 2015/2016 season in Portugal and correlate the circulating genotypes with detected ones in previous seasons. Will also be evaluated the association between genotype, clinical diagnosis and age.

Materials and Methods
- During 2015/16 winter season, between November/2015 and February/2016, 45 RSV were genetically characterized.
- RSV positive respiratory samples were collected in two settings: children under 5 years old diagnosed by 5 hospital laboratories from the Portuguese Laboratory Network for the Diagnosis of Influenza Infection, and all age Influenza-like illness (ILI) patients reported by primary care health services diagnosed by the National Influenza Reference Laboratory.
- All samples were irreversibly anonymized.
- Demographic and clinical data were collected.
- RSV detection was performed by real-time PCR and other biomolecular methods.
- RSV genotype was assigned by the nucleotide sequence of the hypervariable C-terminal region of the G protein gene and the phylogenetic analysis was performed in MEGA 6.0.

Results
- From 45 RSV genetically characterized, 31 (69%) were reported by hospitals, patients age ranged from newborn to 4 years old. From these, 25 (81%;25/31) patients were hospitalized, being the bronchiolitis the most frequent diagnosis.
- 4 (31%) RSV cases came from primary care health services, patients age ranged from 3 to 83 and had a clinical diagnosis of ILI.
- Were included patients from both genders in equal proportions.
- RSV A and B co-circulated during 2015/2016 season.
- Were genetically characterized 21 (47%) RSV A and 24 (53%) RSV B.
- 90% (19/21) of RSV A clustered in ON1 genotype, the others 2 clustered with NA1 genotype (Fig 1).
- All RSV B present a BA-like genotype, 70% (17/24) were similar to BA9 and 30% (7/24) clustered with BA10 genotype (Fig 2).

Conclusions
- During 2015/2016 season was observed a co-circulation of RSV A and RSV B. In present study ON1genotype was predominant in circulation among RSVA, this was also detected as the major RSV A genotype at the global level. Only two RSVA belonged to NA1 genotype. In Portugal, NA1 was in circulation during 2010-2012 period. Undetected since 2012, it seems to reappear during 2015/16 season.
- All RSV B characterized belonged to BA genotypes, the majority clustered within BA9 genotype. BA10 genotype was also identified in circulation at low frequency. BA9 and BA10 were being found in co-circulation since 2011/12.
- No association was found between age, clinical diagnosis and RSV A and B genotypes.
- RSV has an important impact in children in high-risk groups highlighting the need off a continuous RSV surveillance each winter.

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