determine the subtype of HRSV in each sample, we performed viral RNA extraction and qPCR. Additionally, we sequenced the hypervariable ectodomain of the glycoprotein (G) gene for both subtypes of HRSV. The G-gene is one of the targets of neutralising antibodies and is therefore under constant immunological pressure to incorporate mutations. The G-gene has been shown to be the most divergent between HRSV-A and HRSV-B subtypes. The annual HRSV epidemic in Belgium occurred every year in winter, with a median onset in November (week 44, SD 1.9) and median end in March (week 5, SD 2.4) of the following year. Out of 1,944 HRSV positive samples at the University Hospitals Leuven, 1,199 (62 per cent) were subtyped. The overall prevalence of both subtypes of HRSV is similar, with 530 (27 per cent) and 487 (25 per cent) positive samples for HRSV-A and HRSV-B respectively over the six years. Nineteen samples (1 per cent) were positive for both subtypes. Both subtypes of HRSV co-circulated, with HRSV-B dominance in the 2013–2014 season and HRSV-A dominance in the 2014–2015 season. With 81 per cent of all positive samples, children under the age of six were the most vulnerable group. Genotyping of the G-gene indicated that genotypes GA2 (HRSV-A) and GB13 (HRSV-B) were the dominant strains in seasons 2011–2012 until 2014–2015. Previous studies of the circulation of HRSV in Belgium showed co-circulation of both subtypes, which is confirmed in our data from 2011 to 2017. Apart from an HRSV-B dominance in 2013–2014 and an HRSV-A dominance in 2014–2015, the prevalence of both subtypes was similar. Young children (<6) were more likely to suffer from an infection with HRSV. However, age was not related with the infecting subtype. The genotype dominance has shifted over the past twenty years: from GA5 and GB13 between 1996 and 2006 to GA2 and GB13 between 2006 and 2011. The latter were the dominant strains in other parts of the world and were still the most prevalent circulating strains in Belgium between 2011 and 2015.

A37 Genome sequencing, genetic characterization, and vaccine-match analysis of influenza B virus detected in hematopoietic stem cell transplant recipients (HSCT), an overview from 2010 to 2017

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Due to its segmented genome, influenza viruses can exchange gene segments during replication within a host cell and form a huge diversity of strains, which constitute the basis for viral pathogen evolution and maintenance. Consequently, every year epidemiologists deal with new virus variants that cause seasonal outbreaks of influenza and represent a challenge to vaccine production and influenza control. Therefore, sequencing segments of the influenza virus is essential to anticipate the impact of infection on the community. In the last three years, an earlier pattern of influenza virus circulation was observed, starting in late summer and peaking in winter. In addition, an increase in influenza B episodes was noted. Hence, in this study, we aim to perform phylogenetic analysis on whole genome of influenza B virus detected in HSCT recipients at Amaral Carvalho Hospital (ACH) and follow the genetic evolutionary pattern and antigenic variability of viral strains to evaluate the occurrence of adaptive changes in the influenza B genome. Respiratory viruses (RV), including influenza B, are major causes of morbidity and mortality in HSCT recipients. At Amaral Carvalho Foundation, RV control policies include respiratory symptom surveillance followed by RV detection in nasal wash (NW) samples by indirect immunofluorescence assay. One aliquot of NW sample is routinely stored for back up or for further studies. Positive influenza B samples identified from 2010 to 2017 will be selected and subsequently subjected to whole-genome sequencing using next-generation sequencing. For whole-genome sequencing, total RNA will be extracted by using QIAamp Viral RNA Mini kit (Qiagen, Hilden, Germany). cDNA will be prepared using SuperScript method as manufacturers instructions (Invitrogen, Carlsbad, CA, USA) and finally processed by Illumina sequencing following four main steps: 1) library preparation; 2) cluster generation 3) sequencing, and 4) data analysis. This advanced molecular gene detection shows genetic variations among different influenza virus particles in a single sample and will allow us to characterize and understand how influenza B viruses are evolving and mutating. In addition, we will investigate the match between the viruses detected and the vaccine strain in the corresponding year of diagnosis.

A38 Prevalence and evolution of avian H1 subtype influenza A viruses in Southern China

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H1 subtype influenza A viruses were the causative agents for three major pandemics in the last century, and have been circulating in human and pigs for decades. However, the natural reservoirs of H1 subtype avian influenza viruses (AIVs) are aquatic birds, which could provide highly diversified candidates for avian-to-mammalian transmissions. Currently, most of the avian H1 sequences in public database are contributed by surveillance of North American wild waterfowl, with only a very small amount from Europe and Asia, implicating a surveillance gap. How H1 AIVs persist in the gene pool and interact with other lineages or subtypes of AIVs remains unclear. Based on long-term surveillance conducted in seven provinces in southern China during the 1970s and from 2001 to 2013, we identified 351 H1 AIVs out of 556,122 samples (0.06 per cent) from wild birds, domestic ducks, geese, chickens, and minor poultry. This only accounted for 0.77 per cent of the influenza positive samples. No H1 AIVs were isolated from terrestrial poultry and 83.5 per cent of the H1 AIVs were isolated from domestic ducks. About 252 avian H1 isolates were selected for NGS sequencing and 148 non-mixed H1 genomes were obtained. Preliminary phylogenetic analysis revealed that the majority of the H1 AIVs belonged to the large monophyletic Eurasian avian gene pool clade, with frequent reassortments with other subtypes of influenza viruses. Of the 148 H1 AIVs analyzed, 64 genotypes were identified. Despite the low prevalence of H1 AIVs, which may restrict their chance of interspecies transmission, we discovered a superior mammalian infectivity of H1 AIVs compared with other subtypes including H2 to H10, except H5 and H8. Taking the complicated regional poultry farming system and live poultry marketing into consideration, the H1 viruses might be able to reassort with other viruses and generate advantageous variants to cause outbreak in human and other mammals, as exemplified by the case of H7N9. Early identification of emerging H1 AIVs with inter-species transmission potential requires continuous surveillance and monitor on the gene flow and evolutionary patterns of these viruses.

A39 Genetic diversity of the hepatitis C virus NS5B gene during HIV/HCV co-infection

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