Summary

Chapter 1 is a general introduction to this thesis. Respiratory syncytial virus (RSV) causes a substantial burden in particular in the very young and old. The spectrum of clinical disease ranges from mild upper respiratory tract illness, otitis media, croup to apnea in premature infants, pneumonia and bronchiolitis. In Europe, RSV accounts for 42-45% of hospitals admissions for lower respiratory tract infections in children younger than two years of age. At present there is no licensed RSV vaccine. Prophylactic use of neutralizing antibody is available. Monthly administration of palivizumab reduces the risk of hospitalisation in premature infants and infants with chronic lung disease or congenital heart disease. Currently, rapid and sensitive molecular techniques for RSV diagnosis are available. With surveillance of RSV, outbreaks can be rapidly detected. In addition, the longitudinal surveillance data can be used to measure the impact of palivizumab prophylaxis and effectiveness of a future RSV vaccine. In this thesis we aimed to improve surveillance and diagnostic methods of respiratory syncytial virus in Europe. In Chapter 2 we have assessed the comparability of virological data for the detection of influenza in countries in Europe and investigated which countries collect RSV data. The type of respiratory specimen and the transport conditions were similar. The diagnostic methods were diverse, and PCR was more often carried out in countries in Western Europe. In Chapter 3 we assessed whether data on RSV collected by a European influenza surveillance network could be used to build an RSV surveillance system in Europe. Data on RSV from France, the Netherlands, England and Scotland were used. The data were entered timely. RSV contributed noticeably to influenza-like illness. Recommendations for RSV surveillance were formulated: 1) Specimens collected as part of an influenza surveillance program should also be tested for RSV; 2) Both combined nose/throat swabs and nasopharyngeal aspirates are acceptable for RSV diagnostics; 3) The application of molecular techniques such as real-time PCR in the diagnosis of respiratory disease has been demonstrated and we advocate this technique for RSV detection; 4) Further develop standardized methods and laboratory techniques; 5) Consider the development of a sentinel approach of representative hospitals; 6) Integration of RSV surveillance in countries joining the surveillance system alongside influenza. Following the surveillance recommendations, we presented the progress over seven years (2001-2008) (Chapter 4). By 2008, progress was made for four out of six recommendations: the number of European countries testing specimens for RSV increased from six
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to fourteen; nose and/or throat swabs were generally used for detection of influenza and RSV; a total of 25 laboratories performed molecular testing for diagnosis and participated in a quality control assessment for RSV with an overall good performance; four of the ten countries that joined EISS in 2004 started reporting RSV detections in addition to influenza in the period 2004-8. Limited progress was achieved for standardising methods and the development of a sentinel surveillance system of representative hospitals. In Chapter 5 we have described the seasonal variation in RSV activity and investigated which meteorological variables are related to RSV outbreaks for different time lags. Time lags up to 4 weeks were included to assess a possible delayed weather effect in relation to RSV activity. We have found that the onset of RSV activity occurred around week 44 and activity peaked around week 52. Relative humidity was positively associated with RSV activity for all time lags, indicating more RSV when relative humidity increased. Minimum temperature was negatively associated with RSV activity and cloud cover was positively related with RSV activity. Relative humidity, minimum temperature, and cloud cover are important predictors of RSV activity in the Netherlands, with the effect of relative humidity being most consistent. In Chapter 6 the laboratory performance of nucleic acid amplification techniques for respiratory syncytial virus (RSV) diagnosis was investigated in 25 laboratories across Europe. In addition we explored what factors were related to the diagnostic performance. The overall sensitivity for all laboratories was 88% (n=25; range 50-100). A correct score of 93% (range 70-100) was observed for laboratories performing in-house real-time PCR or nested PCR. Multilevel analysis showed that the type of assay (nested or real-time PCR vs. commercial test) was a significant factor (OR=8.39; CI1-36.78) in predicting a correct result. The results for this external quality control study showed that the overall performance of laboratories for RSV diagnosis in Europe is good and that real-time PCR is preferably used for RSV diagnostics. In Chapter 7 we present the detection of respiratory viruses in infants during primary respiratory illness, investigate the sensitivity of nasal swabs and nasopharyngeal aspirates and assess whether patient characteristics and viral load played a role in the sensitivity. Paired nasopharyngeal aspirates and nasal swabs were collected in 98 infants. Rhinovirus (n=67) and respiratory syncytial virus (n=39) were most frequently detected. Co-infection occurred in 48% (n=45) of the infants. The sensitivity of the nasal swab was lower than the nasopharyngeal aspirate in particular for respiratory syncytial virus (51% vs. 100%) and rhinovirus (75% vs. 97%). Sensitivity of the nasal swab was strongly determined by the cycle threshold
value (p<0.001). Sensitivity of the swab for respiratory syncytial virus, but not rhinovirus, was 100% in children with severe symptoms (score≥11). It is concluded that for community based studies and surveillance purposes the nasal swab can be used, though the sensitivity is lower than the aspirate in particular for the detection of mild cases of RSV infection. Finally, in Chapter 8, the results of the earlier chapters were summarised and discussed and the implications and recommendations for future research were formulated. RSV surveillance may be used to support clinical decision making for prophylaxis in premature infants and the data obtained through RSV surveillance has been useful in understanding the seasonal and geographical RSV trends. The findings of this thesis will provide reference to further establish surveillance of RSV on both a regional and European level. RSV surveillance is relevant for providing knowledge on “who” is infected and “when”, and will provide useful information for the timing of administration of palivizumab. In addition, in the light of a future vaccine, fully integrated surveillance data - from clinical diagnosis to hospitalisation - is important and may be used to assess vaccine efficacy.