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**Abstract no: 221**  
**Presentation at ESCV 2016: Poster 191**

Microarray-based molecular detection of viral pathogens associated with respiratory infections

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**Objective:** Acute respiratory infections are one of the most common infectious diseases. Clinical course of acute respiratory infections in young children and the elderly may be serious and life-threatening. It has been emphasized that 20–60% of etiologic agents are viruses. Respiratory syncytial virus (RSV) is the most common viral pathogen in children. On the other hand, influenza virus is the most commonly identified agent in adults. The aim of this study is to identify the viral pathogens in hospitalized patients with respiratory tract infection by using multiplex PCR method.

**Methods:** Nasopharyngeal swab samples obtained from hospitalized patients with acute respiratory infections were enrolled between January 2013 and December 2015. The identification of influenza virus type A and B, human rhinovirus (HRV), respiratory syncytial virus A and B (RSV A-B), parainfluenza virus type 1, 2, 3, and 4, adenovirus, bocavirus, human coronavirus, human enterovirus and human metapneumovirus (HMPV) in nasopharyngeal samples was investigated by using CLART® Pneumovir kit based on clinical array technology (Genomica, Spain).

**Results:** Of the 1290 patients included, 1110 (86%) were children and 180 (14%) were adults. The number of samples in which only one virus was identified was 600. In additional 150 specimens, co-infections of multiple viruses were detected. The total of positive samples was 750 (58%). The majority of these positive specimens were children's samples (694 versus 56). RSV was the most common viral agent (35%) followed by HRV (13%), and influenza (10%). The rate of co-existence of viral pathogens was 20%. The multiplex PCR results were shown in Table 1.

**Conclusion:** While RSV was the most common viral pathogen detected in respiratory infections, other emerging agents, such as human metapneumovirus, bocavirus, and HRV were detected in considerably high rates, suggesting these emerging agents should not be underestimated in the etiology of respiratory infections. In conclusion, in this study, it was shown that microarray-based multiplex PCR method is an easy, rapid, and sensitive diagnostic tool for diagnosis of viral respiratory infections and that utility of this method makes it essential among routine diagnostic tools in clinical microbiology laboratories.

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| Table 1 Distribution of respiratory viruses in positive samples. |
|---|
| **Viral agents** | **% (n)** |
| RSV | 35 (266) |
| HRV | 13 (94) |
| Influenza type A–B | 10 (78) |
| Bocavirus | 6 (45) |
| Parainfluenza virus | 6 (44) |
| Metapneumovirus | 5 (35) |
| Adenovirus | 4 (30) |
| Enterovirus | 0.7 (6) |
| Coronavirus | 0.3 (2) |
| Co-existence | 20 (150) |
| Total | 100 (750) |

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Detection of influenza viruses from patients in university hospital

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**Objective:** Influenza epidemics occur almost every winter and are associated with considerable morbidity, mortality and economical lost. All age groups are susceptible, but increasing age, certain chronic medical conditions, immunity deficiency, pregnancy increase the risk of complications and death. Influenzae A and B are responsible for annual epidemics. Quick diagnosis of influenza by laboratory methods are important for treatment of flu and prevention of epidemics.

The aim of this study is to detect prevalence of influenza virus and its subtypes in patients who admitted to University Hospital in Konya province with flu like symptoms.

**Material and methods:** Nasal swap samples of patients with flu like symptoms who admitted to various clinics in Necmettin Erbakan University Meram Medical Faculty in Konya; Turkey between January 1, 2013 and May 25, 2016 were tested for Influenza A and Influenza B by CLART® PneumoVir and Seeplex® RV12 ACE Detection multiplex PCR (Seegene, S. Korea).

**Results:** Results of total 2041 samples are analyzed retrospectively. 258 (12.6%) samples were positive for influenza virus, 97 (4.7%) were found to be positive for influenza A and 161 (7.8%) for influenza B.

**Conclusion:** Influenza B was found to be the predominant subtype in patients who admitted to hospital with flu-like symptoms in Konya province. In our study January to March were the months with the highest percentage of testing positive for influenza virus infection. Influenza virus can be detected from respiratory samples with high sensitivity by molecular methods such as microarray method and multiplex PCR. Observing seasonal activity and epidemic strains and starting early treatment and taking isolation measures are important for preventing rapid spread and progression of virus and has critical role for public health.

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An overview of human parainfluenza virus 1-4 infections in northeastern Slovenia based on molecular detection

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**Introduction:** Human Parainfluenza viruses (HPIVs) are one of the most common causes of upper and lower respiratory tract illnesses, and an important cause of hospitalizations among children under 5 years old. There are currently no published data on HPIV infections in Slovenia.
Methods: In this retrospective study we examined a total of 2637 samples (nasopharyngeal, nose and throat swabs). They were taken from patients with symptoms of respiratory infection admitted to University Medical Centre Maribor during the years of 2014 and 2015. HPIV RNAs were detected with a commercial automated multiplex PCR system (FilmArray, Biofire).

Results: Out of 2637 samples, 173 (6.56%) tested positive for HPIVs. Nearly half of the HPIV-positive patients were infected with HPIV-3 (49.71%, 86), followed by HPIV-4 (21.39%, 37), HPIV-1 (16.76%, 29) and HPIV-2 (15.03%, 26), respectively. Most frequently identified type was HPIV-3, with regular activity throughout 2014 and 2015, including a substantial increase in both autumn-winter seasons with peaks in November of 2014 and 2015. It was also the predominant HPIV type represented in summer months of both years alongside minute occurrences of type 2 and 4. An apparent outbreak of HPIV-4 infections starting in summer, and progressing in autumn of 2015 with a peak in September, was observed. At the same time, HPIV-3 and HPIV-2 were in decline. Also, type 1 and 3 started to increase as HPIV-4 decreased. Type 2 was completely absent in spring 2014 but had a slight peak in October 2014 and was subsequently present in smaller numbers for the rest of 2015. The median age of HPIV-tested patients was 5, and ranged from less than a year to 96 years old. The majority (82.08%, 142) of infected patients were children under the age of 5. Among the elderly (>65 years old) 12.75% (13/102) tested positive for one of HPIVs, the oldest being 87 years old. The male to female ratio of patients infected with HPIV was 1:1. HPIV was detected as the only cause of infection in 60.11% (107) of cases and 5 of them tested positive for two types of HPIV. In forty-eight (28.97%) HPIV-positive samples one co-infection with other respiratory pathogen was detected, 18 (10.11%) had two co-infections and 5 (2.81%) had three or more co-infections. The prevalent (50.00%) pathogen of co-infection was rhinovirus, followed by adenovirus in 18.00%, enterovirus in 12.00% and respiratory syncytial virus in 10.00% of samples. Coronavirus (HKU1 and OC43), Mycoplasma pneumoniae, human metapneumovirus and Bordetella pertussis accounted for the remaining 10.00%.

Conclusions: Overall, the analysed data suggests HPIV-3 as the most prevalent type of HPIV infections in NE Slovenia. Both HPIV-2 and HPIV-3 showed continual presence in the studied 2-year period with the latter greatly outnumbered the former. A similar biennial distribution pattern for HPIV-1 and HPIV-4 was noted, which could mean that they tend to occur in odd-numbered years. We also observed an epidemic of HPIV-4 which is rarely reported in literature. From previously published reports it appears that seasonal trends vary in different parts of the world and that the distribution of HPIV types is also affected by environmental conditions. Additional data from following years is needed for a more clear understanding of HPIV seasonal trends and interactions between all four types in NE Slovenia.

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Seasonality of respiratory syncytial virus infection in the EU/EEA, 2010–2016
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Background: Respiratory syncytial virus (RSV) is considered the most common pathogen causing severe lower respiratory tract infections among infants and children. RSV vaccine candidates are in development and the World Health Organization is preparing global RSV surveillance to estimate the impact of future RSV vaccines. One of the surveillance objectives is to monitor RSV seasonality and intensity. A subset of EU/EEA Member States (MS) are already testing clinical specimens for influenza and RSV as part of their routine influenza surveillance. In this study, we are describing the seasonality of RSV infection in these countries.

Methods: We performed a retrospective descriptive study of laboratory-confirmed RSV detections reported weekly through the European Influenza Surveillance Network based on influenza-like illness (ILI) or acute respiratory infection case definitions between weeks 40/2010 and 14/2016. We compared findings between systematically sampled primary-care-based sentinel specimens tested according to a standard protocol and convenience sampled primary and hospital-care-based non-sentinel specimens. We also studied the correlation between the median week of peak RSV detections and the latitude of each reporting country’s capital by Pearson’s correlation. RSV seasons were defined as the number of weeks when detections exceeded 5% of total detections per season per country.

Results: Seventeen MS reported RSV detections during the study period: seven MS reported 4399 sentinel detections and fifteen MS reported 156,698 non-sentinel detections. Two MS contributed 60% of sentinel and 61% of non-sentinel detections. Seasonality was observed within both surveillance systems. The median length of RSV season estimated based on sentinel and non-sentinel surveillance was 11 (with country range 6–28) and 10 (range 6–18) weeks, respectively. The median peak week for sentinel detections was week 6 (range 48–18), and for non-sentinel detections week 5 (range 49–17). RSV was detected by non-sentinel surveillance throughout the year but in sentinel system only during weeks 45–13 with consistent reporting. RSV detections peaked later with increasing latitude (r = 0.41 for sentinel and 0.46 for non-sentinel).

Conclusions: RSV detections in 17EU/EEA MS followed a seasonal pattern, peaking regularly early February and lasting around 10 weeks. Our data confirm the moderate correlation between the timing of the epidemic peak and increasing latitude that has been shown earlier. Our study suggests that RSV seasonality can be assessed through both sentinel and non-sentinel influenza surveillance systems but more sensitively in the latter one. Overall, the number of sentinel RSV detections were vastly lower compared to non-sentinel specimens which is a reflection of different surveillance systems and number of participating countries. We do not have RSV-specific denominator data and can therefore not calculate proportions. Further limitations of the data include that large detection volumes originate from only two MS. Despite the limitations, this study supports the use of influenza surveillance systems for monitoring RSV seasonality with consideration to adjust the ILI case definition to establish an RSV-specific surveillance system. Further-