Poster Presentation 4

P4-31

RNA virus diversity in *Aspergillus* species revealed by FLDS, a comprehensive non-retro RNA virus surveillance method

Yuto Chiba[1)], Takashi Yaguchi[2)], Syun-ichi Urayama[3,4)], Daisuke Hagiwara[2,3,4)]

[1)]Graduate school of Life and Environmental Sciences, University of Tsukuba, Japan
[2)]Medical Mycology Research Center, Chiba University, Japan
[3)]Faculty of Life and Environmental Sciences, University of Tsukuba, Japan
[4)]Microbiology Research Center for Sustainability, University of Tsukuba, Japan

Purpose: RNA virus diversity in filamentous fungi have been intensively investigated for these two decades. To find RNA viruses in fungi, detection of double-stranded RNA (dsRNA), a hallmark of RNA virus infection, and RNA sequencing (RNA-seq) were used. However, the sensitivity of dsRNA detection by electrophoresis was relatively low, and there were difficulties in capturing terminal sequences of the viral genome in RNA-seq analysis. Our knowledge of RNA virus diversity in filamentous fungi can be restricted by those methodological limitations. To overcome these limitations, we performed newly developed comprehensive virus detection method, fragmented and primer ligated dsRNA sequencing (FLDS).

Methods: We used clinical and environmental isolates of *Aspergillus fumigatus* and its related species for RNA virus identification. For high-sensitive detection of RNA virus and retrieval of its complete genome sequences, FLDS was used.

Result: We identified at least one RNA virus in 17 of 156 isolates. Among them, 8 isolates were infected by ssRNA viruses although they did not represent dsRNA bands by gel electrophoresis. This result suggested that ssRNA virus diversity had been underestimated by dsRNA electrophoresis. We also reconstructed complete RNA viral genomes based on terminal sequences of genome segments which are shared among segments in a single virus genome. Some sequences predicted to be one of the RNA viral segments did not show significant similarity to known RNA viral proteins, suggesting that these segments may have been missed by RNA-seq analysis.

Conclusion: We identified RNA viruses or RNA viral genes which could be overlooked by the limitations of conventional RNA virus detection methods. The RNA virus diversity in *Aspergillus* species was higher than previously expected. Our findings and the method will provide a deeper insight into RNA virus diversity in filamentous fungi.