

SEMINAR SERIES

8 OCTOBER 2008

INSTITUTE OF SYSTEMS BIOLOGY
(INBIOSIS)
UNIVERSITI KEBANGSAAN
MALAYSIA

PRESENTED BY:
DR. SUNGCHAI PAYUNGORN
CHULALONGKORN UNIVERSITY,
BANGKOK, THAILAND



TENTATIVE SCHEDULE 08/10/2008 (WEDNESDAY)

10:30 a.m. – 10:45 a.m.

Registration of Participants

11:00 a.m. – 12:00 p.m.

Presentation 1 : Molecular Epidemiology of Avian in Thailand 2004-2008

12:00 p.m. – 1:00 p.m.

Lunch Break

1:00 p.m. – 2:00 p.m.

Presentation 2 : Detection and Discrimination of WU/KI Polyomaviruses by Real-Time PCR with Melting Curve Analysis

Presentation 3: Prevalence and Molecular Characterization of WU/KI Polyomaviruses Isolated from Pediatric Patients with Respiratory Disease in Thailand

2:00 p.m. – 2:30 p.m.

Conclusion, questions and wrap-up
Tea Break



Biography

Dr Sunchai Payungporn from the Department of Biochemistry, Faculty of Medicine, Chulalongkorn University, Bangkok.

He worked as a guest researcher in Molecular Genetics Section, Influenza Branch, Division of Viral and Rickettsial Diseases, National Center for Infectious Diseases, Centers for Disease Control and Prevention, Atlanta.

His research is mainly focused on virus, especially in different strains of influenza A viruses.

Abstract

“Detection and Discrimination of WU/KI Polyomaviruses by Real-Time PCR with Melting Curve Analysis”

WU and KI polyomaviruses are novel viruses of the *Polyomaviridae* family, which have been identified recently in respiratory secretions from patients with acute respiratory tract infection. Their potential role in respiratory disease is still unclear and requires additional investigation. To facilitate further studies and diagnosis, a real-time PCR with melting curve analysis was optimized and evaluated to detect WU and KI polyomaviruses. Primers specific for the VP1 gene were designed from regions conserved among WU and KI polyomaviruses which provided amplification products of 198 and 231 bp corresponding to WU and KI, respectively and thus yielded a difference in melting temperature (T_m) between WU and KI polyomaviruses. The assay proved highly specific for WU and KI polyomaviruses as no cross amplification was detected with other respiratory viruses or human genomic DNA. The assay was also highly sensitive with a detection limit as low as 10 copies/ μ L for both WU and KI polyomaviruses. The performance of the real-time PCR assay was evaluated in terms of amplification efficiency (92%). Finally, the assay was validated using DNA extracted from clinical respiratory specimens for WU and KI polyomaviruses and the results were confirmed by direct nucleotide sequencing. The results obtained by melting curve analysis were in perfect agreement with nucleotide sequencing. In conclusion, this method is advantageous because it is rapid, specific, sensitive, reproducible, accurate, cost-effective and thus, would be feasible and attractive for large-scale analysis aimed at investigating the clinical role of WU and KI polyomaviruses.

“Prevalence and Molecular Characterization of WU/KI Polyomaviruses Isolated from Pediatric Patients with Respiratory Disease in Thailand”

WU and KI polyomaviruses represent novel viruses discovered in respiratory secretions from human patients with acute respiratory tract infection. However, the association between WU/KI polyomaviruses and human disease has remained unclear. In this study, the prevalence of these two novel viruses and occurrence of co-infection with other respiratory viruses were determined in Thai pediatric patients with respiratory disease. Previously described PCR assays were applied to detect WU/KI polyomaviruses as well as other respiratory viruses in 302 nasopharyngeal suction specimens collected from February 2006 through February 2007. Results revealed the annual prevalence of WU and KI polyomaviruses in the Thai population was 6.29% and 1.99 %, respectively. The frequency of co-detection of WU and KI polyomaviruses with other respiratory viral pathogens was 42.11% and 33.33%, respectively. Moreover, 2 each of the complete genome sequences of WU (CU_295 and CU_302) and KI (CU_255 and CU_258) polyomaviruses were genetically and phylogenetically characterized. Sequence analysis showed that they contained features common to those found in previous studies. However, there were several nucleotide variations within the non-coding regulatory regions and various non-synonymous mutations within the coding regions which may influence virulence and pathogenesis of these viruses. Nevertheless, it is still possible that these viruses are not the causative agents of clinical respiratory disease. Therefore, judging the association of WU/KI polyomavirus infections with a particular disease will be challenging and require more comprehensive case control investigations.