Toolbox for serological detection of influenza virus infection

Mathias Ackermann[1], Cornel Fraefel[1], Andrea Laimbacher[1], Silke Heidemeyer[1]
[1]Institute of Virology, University of Zurich, Winterthurerstrasse 266a, 8057 Zurich

Key words
Influenza; MS-RT-PCR; ELISA; Serologie; Array

Aim of the study
Optimized identification, cloning, and sequencing of novel Influenza virus strains.
Expression of Influenza virus antigens (H and N) in selected vectors.
Preparation and characterization of ELISA antigens for serological survey studies.

Material and methods
Multi-segment amplification (MS-RT-PCR) was established to quickly and reliably amplify and clone all 8 segments from either historic or current Swiss Swine influenza viruses (SSIV). Some cloned H and N genes were made available from IVI. Cloned genes were transferred to the Gateway system in order to be expressed as fusion proteins with different C-terminal tag markers (EYFP or V5). Expressed proteins were characterized by transient expression and Western immunobloting.

Results and significance
A total of 49 historic and current SSIV samples were subjected to MS-RT-PCR. The obtained sequences revealed that all available SSIV were H1N1 and carried mutations to make them resistant against amantadine as well as against Oseltamivir (Tamiflu). For the ELISA purpose, we yielded the following 6 expression clones within the Gateway amplicon expression vectors: H3-EYFP, H3-V5, H5-EYFP, H5-V5, N1-EYFP, N1-V5. From transient expression and Western immunoblotting, it may be concluded that the expressed proteins conform to our expectations, both in terms of subcellular localization and apparent mobility on Western blots. The antigens are ready for further use by ELISA arrays.

Publications, posters and presentations
Heidemeyer, S. (in preparation) The first full set of Swiss swine influenza virus sequences (Genbank submission and accompanying publication are in progress).

Project *1.07.18*

Project duration *März 2008 – Juni 2010*