

## Abstract

8<sup>th</sup> meeting of the WHO Advisory Committee on Variola Virus Research, 16-17 November  
2006, Geneva, Switzerland

### **An update on variola virus stocks. Conservation of genetic material of different variola virus strains from the Russian collection at the WHO Collaborating Centre for Orthopoxvirus Diagnosis and Repository for Variola Virus Strains and DNA**

**I.G.Drozdov, A.N.Sergeev, S.N.Shchelkunov, I.N.Babkina, P.F.Safronov**  
FSRI State Research Center of Virology and Biotechnology VECTOR, Rospotrebnadzor,  
630559 Koltsovo, Novosibirsk region, Russian Federation

To permit more robust conservation of variola virus DNA (in comparison to full-length viral DNA that needs to be stored in a solution without being frozen), an effort was undertaken to create collections of long PCR amplicons embracing the entire viral genome whereas the amplicons could be stored in the form of alcohol precipitates, which significantly extends the period of reliable conservation of DNA fragments.

Amplicons of predicted lengths (from 2.4 to 11.6 kbp) were produced in LPCR using preparations of 17 VARV strains. All amplicons, carrying variola virus genomic fragments, were characterized in 0.8% agarose gel electrophoresis and deposited for long-term storage within a certified repository. Each variola virus strain has its own storage compartment in the repository. The collections of LPCR amplicons of DNA of each variola virus strain consist of a triply repeated set of fragments.

Based on the amplicon collection, we began creating clone libraries of hybrid plasmids containing DNA fragments of complete genomes of variola virus strains from the Russian collection. This method of conservation of variola virus genomic fragments is the most reliable because plasmids keep well under appropriate conditions and, if necessary, they can be produced easily in bacterial cells in a BSL-2 laboratory. The variola virus DNA amplicons were hydrolyzed by certain restriction enzymes and then inserted into bacterial plasmids. Hybrid plasmid collections with DNA fragments of 13 variola virus strains have been produced.