

WHO Recommendations concerning the distribution, handling and synthesis of Variola virus DNA

**Based upon recommendations made to WHO by
the WHO *Ad Hoc* Committee on Orthopoxvirus Infections (1990 & 1994)
AND
the WHO Advisory Committee on Variola Virus Research (2003, 2004 & 2007))**

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Preamble

The only known stocks of Variola virus are held at the Centers for Disease Control and Prevention (CDC), Atlanta, Georgia, United States of America, and the Russian State Centre on Virology and Biotechnology (Vector), Koltsovo, Novosibirsk Region, Russian Federation, both of which are WHO (World Health Organization) Collaborating Centres. Any research using live Variola virus has to be performed in the maximum containment laboratories of these institutions and requires permission from WHO. Genetic engineering of Variola virus and attempts to produce live virus from DNA are strictly prohibited.

Scientists wishing to perform research on diagnostics or treatment of smallpox, or vaccines against smallpox, may obtain parts of the Variola virus genome, which in its naked form is not infectious, from one of the WHO Collaborating Centres. WHO or the Collaborating Centres will advise scientists on the procedure to follow in order to obtain permission to receive viral DNA. Scientists should be aware that the amount of DNA they request or hold must not exceed 20% of the total viral genome (see also below).

The scientific community may not be fully aware that the distribution, synthesis and handling of Variola virus DNA is governed by a series of recommendations made by the WHO Ad Hoc Committee on Orthopoxvirus Infections and by the WHO Advisory Committee on Variola Virus Research, which have been endorsed by WHO. Scientists wishing to obtain, handle or synthesize Variola virus DNA must therefore comply with these recommendations. The present document gives an overview of these recommendations, which are reproduced in their original wording as found in the various WHO meeting reports (<http://www.who.int/csr/disease/smallpox/research/en/index.html>).

Distribution of Variola virus DNA

The two WHO Collaborating Centres acting as repositories for Variola virus may distribute Variola virus DNA fragments to appropriate research laboratories that request them provided that:

- a) The request has been submitted to the international repository through WHO/Headquarters (1, 2).

- b) The receiving laboratory agrees that the DNA will not be distributed to third parties, unless authorization by WHO has been obtained. This should be controlled through a Material Transfer Agreements between the distributing and receiving laboratories (with copy to WHO) (1, 2, 3).
- c) An annual report on the status of the Variola virus DNA will be made to the international repository and to WHO (2).

No laboratory (except the International repositories) shall be permitted to hold clones representing more than 20% of the Variola virus genome at any one time (2).

Fragments of Variola virus DNA, not exceeding 500 base pairs in length, may be freely distributed between identified laboratories for use as positive controls or standards in diagnostic kits, providing collectively they do not exceed 20% of the total genome size (4, 5).

Handling of Variola virus DNA

Studies on Variola virus DNA are permitted on conditions that:

- a) The DNA will not be used for insertion into vaccinia virus or related poxviruses (2).
- b) All work with Variola virus DNA (greater than 100 nucleotides long) is done following a written risk assessment and in accordance with locally agreed national guidelines (2).
- c) No other orthopoxviruses are handled in the laboratory rooms where Variola virus DNA is studied (2).
- d) All by-products containing Variola virus DNA are disposed of by autoclaving at 120°C for 30 minutes (6).

Synthesis of Variola virus DNA

- a) Attempts to synthesize full-length Variola virus genomes or infectious variola viruses from smaller DNA fragments are strictly forbidden (7).
- b) In vitro synthesis of Variola virus DNA, or any DNA encoding a Variola virus polypeptide, where the length of the DNA exceeds 500 base pairs requires approval from WHO. Similarly, mutagenesis of orthopoxvirus DNA of larger than 500 base pairs, with the aim of producing the corresponding Variola virus DNA sequence again requires permission from WHO. Under no circumstances can laboratories, other than the WHO Collaborating Centres hosting the Variola virus repositories, hold DNA comprising more than 20% of the total genome (4, 7).
- c) Production of DNA microarrays, on which small oligonucleotides (less than 80 base pairs) are covalently bound to a matrix and which, in aggregate, may span the entire genome, does not require permission from WHO (4, 5).

Reporting obligations

Variola virus DNA is distributed to scientists on the understanding that an annual report on the status of Variola virus-specific DNA clones will be made to the international repository (see above: distribution of Variola virus DNA, paragraph c). This reporting obligation also

applies to scientists who have obtained permission from WHO to synthesize Variola virus DNA larger than 500 base pairs, or generate Variola virus-like DNA by site-directed mutagenesis of other orthopoxvirus DNA.

References

- 1 Report of the *Ad Hoc* Committee on Orthopoxvirus Infections, 1990, page 5.
- 2 Report of the *Ad Hoc* Committee on Orthopoxvirus Infections, 1994, page 8.
- 3 Report of the WHO Advisory Committee on Variola virus Research, 2007, 23.4
- 4 Report of the WHO Advisory Committee on Variola virus Research, 2003, 11.7
- 5 Report of the WHO Advisory Committee on Variola virus Research, 2004, 8.2.
- 6 Report of the Ad Hoc Committee on Orthopoxvirus Infections, 1994, page 9.
- 7 Report of the WHO Advisory Committee on Variola virus Research, 2004, 8.4.