



VACCINIA VIRUS and VACCINIA VIRAL VECTORS

The Vaccinia virus is the most studied of the poxvirus family. These are the largest known DNA viruses and replicate entirely within the cytoplasm of infected cells. Vaccinia virus as a vector can accept as much as 25 kb of foreign DNA. Foreign genes are integrated stably into the viral genome.

Potential Health Hazards

Multiple strains of vaccinia virus exist with varying levels of virulence for humans and animals. Depending on the strain used, vaccinia virus presents different levels of health risk to laboratory personnel. Strains that are highly attenuated are typically unable to replicate or replicate poorly in human cells. However, non-highly attenuated strains of vaccinia have the ability to replicate in human cells and thus pose a risk to humans. Risks include localized skin infections, and more severe, disseminated reactions to which immunocompromized individuals may be more susceptible.

Vaccination is NOT recommended for those working with the following highly attenuated strains:

Highly Attenuated Strain	Biosafety Level	Derived From:
MVA	2	Vaccinia virus
NYVAC	1	Vaccinia virus (Copenhagen)
TROVAC	1	Fowlpox virus
ALVAC	1	Canarypox virus

Although there is no formal surveillance system in place, there have not been any reports of laboratory-acquired infection resulting from exposure to any of the above highly attenuated strains or recombinant vaccines derived from these strains in the literature or to the CDC.

Appropriate biosafety guidelines and infection control procedures should always be observed when working with viral material even if vaccination is not indicated.

Vaccinia vaccination is **recommended** (but not required) for laboratory workers who directly handle cultures or animals infected with:

- Non-highly attenuated vaccinia virus strains

- Recombinant vaccinia viruses from non-highly attenuated vaccinia strains
- Other orthopox viruses that can infect humans

Non-highly Attenuated Strains	Biosafety Level
WR (Western Reserve) NYCBOH (strain used in vaccinia vaccine) Copenhagen Temple of Heaven Lister Cowpox	2

Modes of Transmission

Vaccinia virus may be transmitted via surface contact with contaminated object(s) and subsequently spread to mucous membranes (eyes, nose, and mouth) and/or to open sores on skin. Accidental needlestick is a mode of transmission within research laboratories. Accidental ingestion of viral contaminated materials and inhalation are other routes of transmission. If working with infectious animal models, then bite wounds could transmit vaccinia virus infection.

Laboratory Acquired Infections

Recent reports of several laboratory-acquired infections of vaccinia virus featured in the Center for Disease Control's (CDC) Morbidity and Mortality Weekly Report (MMWR) provide convincing evidence for the importance of good laboratory biosafety practices. When appropriate, laboratorians must give serious consideration to vaccination to protect against potential occupational exposures to select vaccinia strains. Minimize or eliminate the use of needles, glass, and other sharps when handling vaccinia virus to avoid accidental needlesticks or cuts leading to possible infection.

Host Range

Humans and certain animals

Survival

Depending on the strain, vaccinia virus is known to be stable when dried.

Laboratory Practices

Biosafety Level 2 practices and facilities must be used for activities involving modified vaccinia vectors.

- Biohazard signs and labels must be displayed in areas and on equipment where vaccinia virus is used and stored. This includes, but is not limited to, laboratory entrance doors, biological safety cabinets, incubators, refrigerators, and freezers.

- Use a biological safety cabinet (BSC) (a.k.a. tissue culture hood) for manipulations that can generate aerosols, such as pipetting, harvesting, infecting cells, filling tubes/containers, and opening sealed centrifuge canisters.
- It is highly recommended that aerosol containment devices are used when centrifuging vaccinia virus, and must be used when using high concentrations or large volumes. These include sealed canisters that fit in the centrifuge bucket, covers for the centrifuge bucket, heat sealed tubes, or seal centrifuge rotors. Rotors should be removed and opened inside a BSC. Centrifuge tubes should always be filled and opened in a BSC.
- Vacuum lines must be protected with liquid disinfectant and micron filters.

Personal Protective Equipment

Work with vaccinia virus must be conducted using the following PPE within a BSL2 facility:

- Disposable gloves – consider the use of double gloves for enhanced protection
- Lab coat or back-closing disposable gown
- Eye protection (safety glasses or goggles) is recommended, but not required when working within a biosafety cabinet.

Precautions When Using Animals

All work with vaccinia virus in animals must be reviewed by the OHSU Institutional Animal Care and Use Committee (IACUC). See the IACUC webpage for more information: <http://www.ohsu.edu/xd/research/about/integrity/iacuc/index.cfm>

ABSL2 handling and containment may be required for your *in vivo* project with vaccinia. The ABSL2 designation will be determined by the IBC or Biosafety Officer during project review. If your work qualifies as ABSL2, then the Department of Comparative Medicine (DCM) will provide training to ensure safety requirements are met. The PI must contact DCM in advance to reserve ABSL2 lab space.

Recombinant and Wild-type Vaccinia Research

Protocols involving recombinant vaccinia virus must be approved by the Institutional Biosafety Committee (IBC); complete a Recombinant DNA Research Questionnaire (RDRQ). Work with wild-type vaccinia virus must also be approved by the IBC; complete an Infectious Agent/Toxin Questionnaire (IAQ).

RDRQ: <http://www.ohsu.edu/xd/about/services/integrity/policies/policy-detail.cfm?policyid=291376>

IAQ: <http://www.ohsu.edu/xd/about/services/integrity/policies/policy-detail.cfm?policyid=291366>

Employee Exposure

The occurrence of vaccinia infection and its course will vary depending on route of exposure, the strain of vaccinia, the dose of exposure, an individual's medical history and "smallpox" (vaccinia virus) vaccine immunization status. Immediate medical "first-aid" interventions may help prevent or lessen the severity of infection. If you know or suspect vaccinia virus exposure...

- **Irrigate the site of exposure**
 - If exposure was by needle stick or other route which breaks the skin, wash with soap and water for 5-15 minutes and cover with a bandage.
 - If exposure was by splash to eyes or mucus membranes, irrigate thoroughly for 15 minutes at an appropriate eye wash station.
- **Report to your Laboratory Supervisor and Biosafety Officer IMMEDIATELY**
- In the weeks following possible infection, take note of the following symptoms which may indicate a need for further medical attention:
 - Lesions or swelling at the site of exposure
 - Rash
 - Fever



Examples of vaccinia virus infections

Spill and Disposal Procedures

Refer to the OHSU BSL2 Manual Template spill response cue cards.

Disinfectants

- 70% Ethanol
- Sodium hypochlorite (1-10% dilution of fresh bleach). If using bleach within a biosafety cabinet, always follow up with a 70% ethanol rinse.

Decontamination

Autoclave cultures for 30 minutes at 121°C, 15 psi. Disinfect work surfaces using an agent above.

Transport Requirements

OHSU. Shipping infectious substances, diagnostic specimens, and/or dry ice off-campus requires training and certification. Individuals must take the Dangerous Goods Shipping Training available on Big Brain.

The use of vaccinia virus and vaccinia viral vectors may be subject to shipping regulations through outside monitoring agencies. Please consult the Biosafety Officer for any regulations that must be met if you are shipping vaccinia.

Questions/Concerns? Contact the Biosafety Officer at 503-494-0655 (Central Campus) or 503-690-5312 (West Campus).