# Working Safely with Vaccinia Virus (VV): Biosafety Information & Procedures Note

**Virus classification and characteristics:** Vaccinia Virus (VV) is a member of the *Poxviridae* family, Genus *Orthopox*. It Is microscopic in size approx 350 by 300 nm. It is a double stranded DNA virus that unusually replicates in the cytoplasm of a cell, rather than in the nucleus *(Image 1).* Certain modified VV strains were (and are still) utilised as the vaccine of choice protecting against smallpox exposure.

**Disease / illness caused:** Localised epidermal lesion/s at site of entry *(Image 2A)* accompanied by an acute fever (temperature spike) and localised rash. Fatigue, headaches, myalgia (muscle pain) may occur to some. Eczema, encephalitis (inflammation of the brain), myopericarditis (inflammation of the heart muscle) and (localised) vaccinia necrosum (skin gangrene) can be caused in more serious infections. Entry of VV into the eye can cause ocular vaccinia lesion/s and conjunctivitis (*Image 2B)* and potentially affect eyesight.

**Host range:** Several mammalian species including humans, rabbits, cows and river buffalo. Rodents are used as experimental infection models. VV can enter most vertebrate cells although replication may be constrained depending on strain variations.

**Route/s of VV Exposure: Close Contact** with the virus and entry through broken skin (contact with VV contaminated samples, equipment, (scratching of) wound sites, bodily fluids (including those from human sexual contact; contact with contaminated cattle teats), dressings, clothing, infected animals, waste); **splash** of VV containing droplets into eye, mouth and other mucous membranes; **inhalational exposure** to aerosols released during VV manipulation or handling (e.g. centrifugation) or spills during transport; **inoculation** via VV contaminated needle/s and sharps.

**Infectious dose**: A (smallpox) vaccine titre is $10^8$ pock-forming VV units per ml. Evidenced lesion formation by VV strains can be as small as $10^4$ pock-forming VV units.

**Individuals at greater risk from VV infection:** Immunocompromised individuals, those with certain skin (e.g. eczema, psoriasis) and cardiac diseases; new and expectant mothers (including the foetus and new born child).

Serious adverse effects from the smallpox vaccination history indicate one death per million doses; 38 cases of eczema vaccinatum, 3 cases of vaccinia necrosum and 12 cases of CNS diseases per million doses. *Pathogen safety data sheet, Public Health Agency of Canada* (https://www.canada.ca/en/public-health/services/laboratory-biosafety-biosecurity/pathogen-safety-data-sheets-risk-assessment/vaccinia-virus.html)

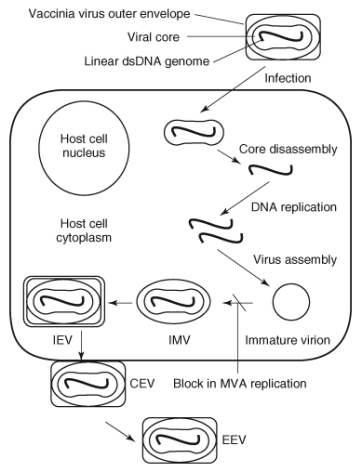
**Incubation period:** Epidermal lesions can appear 3-6 days after exposure, lesions would normally scab over and heal in about 10-15 days.

**Stability of VV:** The dried virus can survive for weeks on many surfaces / samples (up to 39 weeks at 4°C at low moisture content); virus can also survive in aqueous solutions for a few weeks.

**Inactivation:** VV is susceptible to autoclaving (121°C for 15 min holding time) and high temperature treatment (95°C for 2 hours). A number of disinfectants can inactivate the virus but efficacy data must be obtained from supplier or in house tests conducted.

**Image 1** - Vaccinia Virus Expression System and Lifecycle.

IMV – Intracellular mature virus; IEV – Intracellular enveloped virus; CEV – Cell associated extracellular virus; EEV – Extracellular enveloped virus.

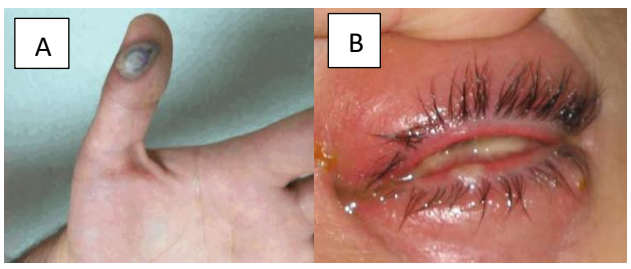



Hall, Yper, and Carroll, Miles W(Jan 2018). In: eLS. John Wiley & Sons Ltd, Chichester. http://www.els.net [doi: 10.1002/9780470015902.a0002659.pub4]

**Image 2** – (A) Localised epidermal lesion at site of VV loaded needle entry and (B) conjunctivitis and ocular lesion after eye exposure.

**(A) CDC.** Laboratory-acquired vaccinia virus infection in a recently immunized person—Massachusetts, 2013. MMWR 2015 May 1;64(16):435-8

**(B)** Lewis F, Chernak E, Goldman E, et al. Ocular Vaccinia Infection in Laboratory Worker, Philadelphia, 2004. *Emerging Infectious Diseases*. 2006;12(1):134-137. doi:10.3201/eid1201.051126.




**Virus risk level:** Wild type non-attenuated VV is classified as **Hazard Group 2** by the Advisory Committee on Dangerous Pathogens (ACDP), which identifies that the virus (i) may cause human disease / illness so is a risk to those who work with it but is (ii) unlikely* to spread to others and (iii) effective prophylaxis and treatment is available.

** In laboratory work involving VV, shedding and spread of virus is possible if biosafety / infection control procedures are not fully identified / followed.*

**VV strains** always considered as Hazard Group 2 by ACDP / SACGM *(see references opposite)*:

- Wild type / unknown strain isolates, Western Reserve.

VV strains that maybe considered as Hazard Group 2 by ACDP / SACGM but with potential for derogation according to peer reviewed / approved risk assessment:

- Lister (Elstree)
  *(the VVL15 strain with thymidine kinase (tk) and viral growth factor (vgf) double deletion is conditional replicative in tumour cells).*
- WYETH (Dryvax)*; Copenhagen.
- ACAM2000 is a derivative of WYETH.

VV strains that are considered as Hazard Group 1 by SACGM:

- MVA, NYVAC.

Genetic modifications that would likely to increase the virulence / risk of a VV strain (see SACGM ref):

- Alteration of 'host range' genes that would alter / widen tissue tropism
- Insertion / expression of immune-evasion (e.g. soluble IFN α/β receptor) or immune-modulatory (e.g. IL-4) gene/s
- Homologous recombination events with other VV strains / pox viruses
- Expression of harmful inserts (e.g. oncogene, toxin)

The risk level assignment for a genetically modified VV strain **must** be peer reviewed by the QMUL BGMSC and GM Class allocated.

**References:**

Approved List of Biological Agents (ACDP 2013; Crown Copyright). http://www.hse.gov.uk/pubns/misc208.pdf

SACGM Compendium of Guidance, Part 2 (HSE 2007; Crown Copyright). Pages 104-115. http://www.hse.gov.uk/biosafety/gmo/acgm/acgmcomp/part2.pdf

QMUL GMO topic page: http://www.hsd.qmul.ac.uk/a-z/genetically-modified-organisms/

QMUL BGMSC page: http://www.hsd.qmul.ac.uk/a-z/health-and-safety-advisory-group/health-and-safety-adviosry-group/bgmsc/

**Health surveillance and post exposure incident management at QMUL:**

**Health surveillance:** The QMUL pre-employment health questionnaire / supplementary health questionnaire are to be completed by the staff member / PGR student with their line manager / supervisor. It is the responsibility of the recruiting manager to complete: 'The Post Details and Potential Hazards' section of the pre-employment health questionnaire. **QMUL OH Service** will then provide appropriate health surveillance advice and support for all relevant staff / PGR students working with VV.

**Vaccination (**with the designated Lister strain vaccine issued by the UK Dept of Health) is possible for the following laboratory personnel, on a case-by-case basis, utilising the information contained in a peer reviewed risk assessment for the

**References:**

HSE, (1990) *Vaccination of laboratory workers handling vaccinia and related poxviruses infectious for humans* http://www.hse.gov.uk/pUbns/priced/acdp-acgm-vaccine.pdf

DoH (2013) Smallpox and Vaccinia - 'Green Book' Chapter 29 https://www.gov.uk/government/publications/smallpox-and-vaccinia-the-green-book-chapter-29

QMUL Occupational Health: Occupational Health Protocol for those working with vaccinia virus (2018) http://hr.qmul.ac.uk/about-us/

work; All current and new laboratory personnel who work with or have (significant) indirect contact with VV will receive counselling and full information by **QMUL OH Service** on the possibility of an adverse outcome in a minority of subjects.

- Work involving the genetic modification of VV infectious for humans or where the risk assessment identifies enhancement of virulence/pathogenicity or gene insert presents a hazard to the health of the worker.
- Scaling up of laboratory work
- Inoculation and work with experimental models

**Contra-indications for vaccination** for certain personnel are noted in the 'Occupational Health Protocol for those working with vaccinia virus' document (reference opposite).

**Post exposure incident management:**

- Laboratory personnel must adhere to the **QMUL procedure for contamination incidents** requiring first aiders to attend so that the correct procedure is then followed ***before*** contacting OHS.
- In the event of an exposure, contact OH Service immediately on 0207 882 8700/7207 requesting an urgent same day appointment.
- The laboratory personnel will be seen in OHS at the earliest opportunity where a strict 10 day surveillance programme will commence. A health assessment form will be completed and Public Health England (PHE), North East and North Central London Health Protection Team will be notified.
- OHS will notify the Royal London Hospital of the exposure to ensure that out of hours and weekend is provided. The designated hospital is The Royal London Hospital at Whitechapel.

See full detail in the in the 'Occupational Health Protocol for those working with vaccinia virus' document.

QMUL procedure for contamination incidents
http://www.hsd.qmul.ac.uk/media/hsd/documents/QMUL_HS_124_Procedure-in-the-event-of-a-contamination-incident_V4_10.10.2018.pdf

**Laboratory tasks that may increase the risk of exposure to VV:**

- Use of needles and sharps containing VV (e.g. experimental animal inoculations and/or preparation of syringes)
- Processes that provide energy for particle dissemination / aerosol or droplet release (e.g. centrifugation, mixing, vortexing)
- High titres or scale up cultures of VV
- Transport between rooms / buildings
- Column or other purification or concentration of the virus
- Freeze-thawing or sonication
- Removal from storage of dried virus samples

**Laboratory risk control measures:**

- Handle live virus within a Class II microbiological safety cabinet or HEPA filtered downdraft table giving adequate user protection.
- Always practice safer sharp handling including eliminating the use of sharps where possible, keeping fingers behind the needle at all times, never re-sheathing and disposal directly into sharps bin. See further precautions at http://www.hsd.qmul.ac.uk/a-z/safer-sharps/
- All contaminated equipment and surfaces to be disinfected according to agreed lab disinfection protocol before it is removed from the primary containment.
- Infected experimental models are kept in approved designated room/s and within HEPA filtered Independently Ventilated Cages on the exhaust.
- Where virus shedding is possible from experimental models, all bedding material is bagged within a HEPA filtered downdraft table giving adequate user protection.
- Containment Level 2 standard facilities and procedures are utilised unless specific derogation to Containment Level 1 is identified for a specified VV strain / GM construct through a peer reviewed risk assessment http://www.hsd.qmul.ac.uk/media/hsd/documents/checklists/QMUL_HS_097_May-2016_Containment-Level-2-Inspection-Checklist-V3.docx
- Risk awareness training is provided (and recorded) to those who work / will work with VV and to those who may be adversely impacted (e.g. others working in multi-use laboratory where VV is handled, staff handling infected experimental models and their wastes). This should include training to recognise vaccinia virus infection; awareness of the possibility of human-to-human transmission; and awareness of the increased risk to those with eczema, those who are immuno-compromised, or those who are pregnant.
- Training includes at minimum the significant findings of the risk assessment; risk assessment user training to be recorded.

**For further advice at QMUL contact** –
QMUL Health & Safety Directorate
http://hsd.qmul.ac.uk/Contact%20Us/index.html
Lead contact - Dr Mark Ariyanayagam (H&S Manager and QMUL Biosafety Adviser)

QMUL Occupational Health Service
http://hr.qmul.ac.uk/about-us/
Lead Contact - Marcia Bennett-Pompey (OH Manager)

**Further Information** (references above and)
Health & Safety Executive (HSE) Biosafety Webpages http://www.hse.gov.uk/biosafety/