BioMaterial Fact Sheet: *Adenovirus and Adenoviral Vectors*

Adenoviruses are a frequent cause of acute upper respiratory tract infections such as the common cold, but have also been associated with other forms of illness such as conjunctivitis (pink eye), gastrointestinal illness and urinary tract infections. There are at least 52 immunologically distinct types that can cause human infections. The most common one used in research is Adenovirus-type 5 (Ad-5, Ad5, Adeno5). Adenoviruses are composed of a single double-stranded DNA molecule and have an icosahedral structure. They are described as a non-enveloped virus, which makes them extremely stable in the environment. Certain types of adenovirus have been shown to survive up to 10 days on paper under ambient conditions, while others have survived 3-8 weeks on environmental surfaces at room temperature. Adenoviral infections are thus easily spread through indirect contact, as well as through exposure to aerosols, ingestion, accidental injection and close personal contact. Individuals with compromised immune systems are especially susceptible to severe complications of adenoviral infections. Adenoviruses can infect a wide variety of cell types and tissues in both dividing and non-dividing cells. This characteristic, together with their relative ease of preparation and purification, has led to their extensive use as gene vectors.

Adenoviral vectors are used by scientists as a means to transfer genetic material into a cell (this process is also known as transduction). Adeno-vectors are typically based on less pathogenic (or attenuated) strains. The viral vector is engineered to be deficient in certain genes so that when the vector infects a cell, it cannot replicate. The vectors are rendered replication-incompetent through the deletion of E1(a) (early gene) or E2, E3, and E4 (early genes). During production of the vector, cultured cells that express the gene product will supply the missing gene components. Adenoviral vectors are used in research because they are not known to integrate into the host's chromosome; can infect a broad range of cells; can accept large amounts of foreign DNA; are easily produced at high titers; and can achieve a high level of expression.

NOTE: Replication-incompetent adenoviral vectors could be complemented *in vivo*, thus resulting in virus dissemination *or* replication-competent virus, if there has been a recombination event.

Containment Level: Adenovirus and adenoviral vectors will be manipulated following Biosafety Level 2 (BSL2) criteria and animals infected with the virus or viral vector should be handled and housed according to Animal Biosafety Level 2 criteria (ABSL2). *Concurrent approvals are needed from the Institutional Biosafety Committee (IBC) and the Institutional Animal Care and Use Committee (IACUC) prior to initiating work.*

Required Training: To work with adenovirus or adenoviral vectors, laboratory and animal staff must have completed the Bloodborne Pathogen (BBP) training offered by EHS as well as the Viral Vector Safety Training coordinated by the IBC Administrator. In addition, staff should receive 'hands-on' training from their laboratory supervisor or animal facility manager prior to manipulating the agent. Training should cover the hazards associated with the work, required practices and procedures and, if manipulating infected animals, proper handling of bedding, cage washing, and all other husbandry materials associated with the experiment.

Personal Protective Equipment (PPE): Laboratory coat (or if in animal facility, PPE per building entry requirements); gloves; goggles and surgical mask OR face shield must be worn. A respirator is not required. **If working with concentrated titers and highly aerosolizing procedures contact the Biosafety Officer at 301-846-5038.**

Engineering Controls: All manipulations/injections/dissections with adenovirus or adenoviral vectors must be conducted within a Biological Safety Cabinet (BSC). ***No work on the open bench!*** If used in animal studies, animals must be placed in ventilated caging racks maintained under negative pressure with HEPA-filtered supply and exhaust post-infection for a minimum of 10 days. Alternative caging options may be determined and implemented on a case-by-case basis by the Animal Facility Manager in conjunction with EHS concurrence.

Additional Safety Practices (Laboratory):

- All vacuum lines must be fitted with a HEPA filter (an example is the "Vacushield ™" in-line hydrophobic filter, Product # 4402 from Gelman Science - http://www.labfilters.com/catalog/924_20077.asp).
- Centrifugation must be done in closed containers and accompanied by an intact aerosol ring using sealed rotor buckets with safety caps and samples in screw-cap tubes. Centrifuge containers must only be loaded and unloaded within a BSC **not** on the open bench.
- Aerosol-resistant tips must be used when pipetting.

Additional Safety Practices (Animal) in addition to those listed above:

- Infected animals may excrete Adenovirus (especially in the first 72 hours after infection). It is recommended by EHS that the lab personnel or animal husbandry technicians, specifically trained on the handling of adenovirus infected animals, be responsible for all animal husbandry practices during the first 72 hours following infection of the animal.
- Precautions must be taken not to create aerosols when emptying animal waste material and when washing down cages, or cleaning the room with pressure hoses.
- All sharps (one-time use) should be immediately disposed of into sharps container (located *within* the BSC) and disposed of as hazardous waste. When a sharps container is full, it must be closed and wrapped with autoclave tape (the tape will ensure the cap does not pop off if dropped).
- Cage bedding and excreta should be bagged, immediately autoclaved and disposed of according to facility requirements.
- Post-infection, all tissue harvests / necropsies must be conducted within a BSC.

Disinfection (Laboratory and Animal Facility): Disinfect all work surfaces and materials *both* prior to and immediately following all work practices and procedures. (also see *Biosafety Technical Bulletin: Decontamination and Virus Inactivation*)

- Surfaces: Dispatch™ or 10% (1:10) bleach solution (made daily) with a minimum of 10 minutes contact time. Rinsing of surfaces with water is recommended after use of any chlorine-based disinfectant on metal surfaces to mitigate corrosion. ***Cavicide™ and/or ethanol are not effective disinfectants against non-enveloped viruses, such as Adenovirus***.

- Liquid Waste: Bring liquid waste to a final concentration of 10% bleach. If aspirating into a 1L flask, flask should first be filled with ~100mL bleach and diluted down to 10% (1:10) with liquid waste. Contact time should be a minimum of 30 minutes prior to drain disposal (while tap water is running).

- Solid Waste: All solid wastes should be treated as hazardous waste according to NCI-Frederick handling procedures.

Employee Exposure:

- Eye Exposure from splash or aerosols: Flush eyes for a *minimum* of 15 minutes in eyewash and then report to OHS immediately afterwards. Follow the NCI-Frederick Exposure Control Plan (http://home.ncifcrf.gov/ehs/ehs.asp?id=12) procedure for reporting occupational exposures to potentially infectious material. Dial 911 after-hours to report exposure and obtain assistance.
- Needlesticks and/or non-intact skin exposure: Wash contaminated skin for 15 minutes using a 10% povidone iodine solution (such as Betadine), a chlorhexadine scrub kit, or soap and copious amounts of water. Report to OHS immediately after scrub. If the exposure occurs during after normal business hours contact the 911 emergency number. Follow NCI-Frederick Exposure Control Plan.

References:
www.cdc.gov
http://www.phac-aspc.gc.ca/lab-bio/res/psds-ftss/index-eng.php
http://webeye.ophth.uiowa.edu/eyeforum/cases/case28.htm