



## Work Instruction

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Title:	<b>Use of the Class II Biosafety cabinet (BSC)</b>		
Approver	Document No:		
Name:	Chris Rollinson	Version No:	2.0
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### 1. Purpose

The purpose of this Work Instruction (WI) is to detail the safe operation of the Biological Safety Cabinet (BSC), in the James Lind Research Centre (JLRC) and to ensure adequate containment of biological materials. The JLRC has a Class II BSC to provide the microbe-free work environment necessary for cell culture propagation and handling of infectious organisms.

### 2. Scope

This WI outlines the safety policies and work practices to provide optimum contamination control and safety when working in a BSC.

### 3. Responsibilities

#### Designated person (Research Governance Manager [RGM])

Will oversee the correct operation and maintenance of the BSC by:

- Coordinating yearly BSC certification by the certified vendor. The dates of the most recent certification must be posted on the front of the BSC.
- Label and quarantine the BSC when it is not operating correctly.
- Make any necessary arrangements for repairs and annual certification of the BSC.
- Periodically inspect the BSC to ensure its operational performance.
- Report to PI unsafe practices by BSC users.

#### BSC users

- BSC users must receive training on the safe and proper use of the BSC prior to use.
- BSC users must follow correct procedures.
- BSC users shall report any injuries, accidents or spills to the designated person.
- BSCs users shall report any defects or malfunctions of the BSC to the designated person.

## 4. Documents needed for this WI

- Study protocol, risk assessment and the BSC operating manual.

## 5. Related documents

- Genetically Modified Organisms (Contained Use) Regulations 2014
- The Scientific Advisory Committee on Genetic Modification (SACGM) (Contained Use) Compendium of Guidance. Part 1: Introduction to the legislation and general health and safety issues. Part 3: Containment and control of contained uses involving genetically modified micro-organisms. Part 6: Guidance on the use of genetically modified microorganisms in a clinical setting.
- Pan UK Pharmacy Working Group for ATMPs. Gene Therapy Medicinal Products. Governance and Preparation Requirements

## 6. Definitions

**BSC:** Biological Safety Cabinet: A piece of equipment designed to protect the operator, the laboratory environment and work materials from exposure to infectious aerosols and splashes that may be generated when manipulating substances containing infectious agents, such as viruses, bacteria and primary tissue cultures.

**Designated person:** Research Governance Manager (may be deputised by the Assistant Clinical Trials Manager).

**HEPA:** High Efficiency Particulate Air Filter: Filter that traps 99.9% of particles of 0.3 µm in diameter and 99.9% of particles of greater or smaller size, thus capturing all infectious agents and ensuring only microbe-free air is exhausted from the cabinet or directed to the work surface.

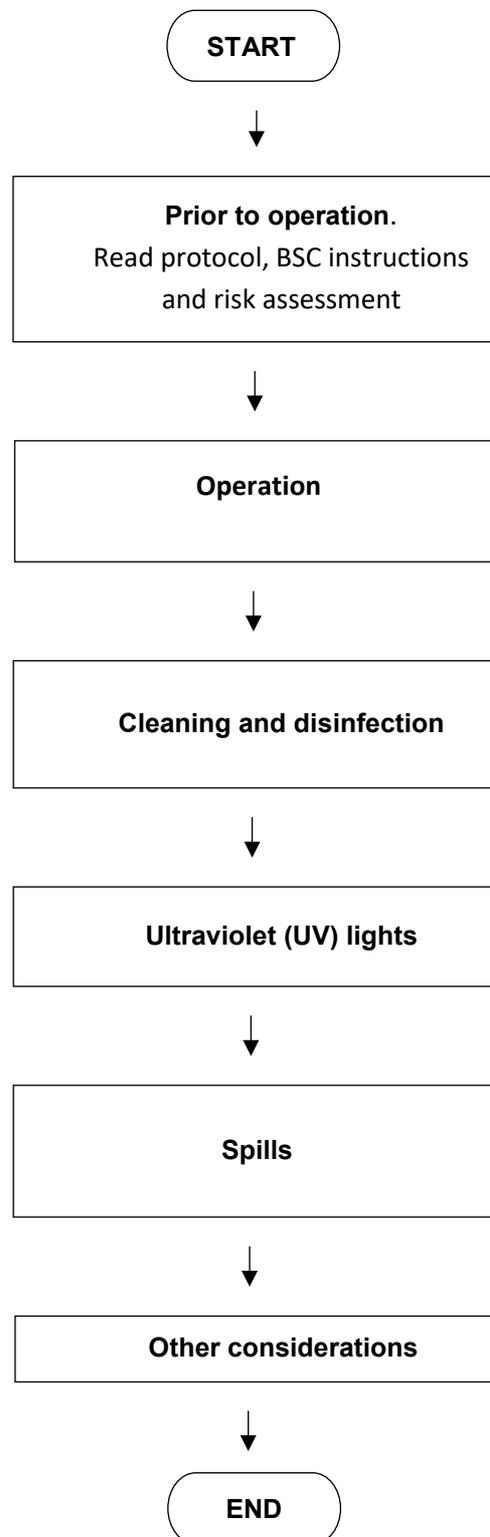
**RD&I:** Research Development and Innovation.

**RGM:** Research Governance Manager

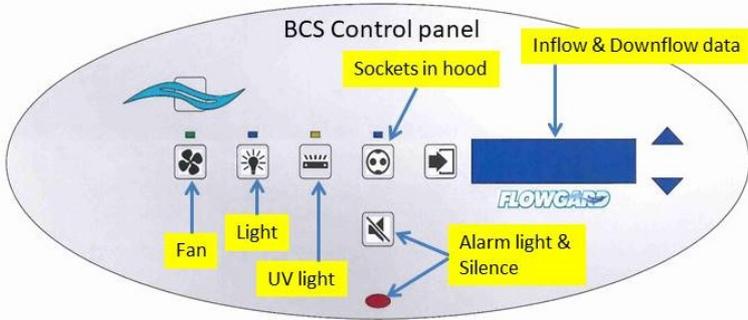
**SOP:** Standard Operating Procedure.

**UHPNT:** University Hospitals Plymouth NHS Trust.

## 7. Process map(s)/ flow chart(s)



## 8. Procedure

Step	Action	Responsibility
1	<p><b>Prior to operation</b></p> <ol style="list-style-type: none"> <li>1. Know the specific hazards associated with the biological samples you are using before you start work. Note any precautions regarding the use of the chemical or microorganisms in the BSC. Incorrect use of cabinet raises the possibility of infection with microbial pathogens or release of GMOs to the environment. To prevent this, ensure you read and understand the Safety Data Sheets, the project risk assessment for materials and organisms being used. Importantly:               <ol style="list-style-type: none"> <li>a. Know your "safe working area" in BSC. The safe working area is basically the work tray or depressed area. All work should be performed on or above the work tray. The area on or above the front grill is a non-safe working area.</li> <li>b. Minimize disruption of "air curtain"</li> <li>c. Minimize room activity</li> <li>d. Utilize unidirectional air flow</li> <li>e. Employ aseptic techniques</li> </ol> </li> <li>2. On entering the James Lind Research Centre (JLRC) BSC room, via swipe card, turn on the lights and if setting up to use the cabinet turn on the biohazard warning light (switch labelled to the right of the light switch).</li> </ol>	BSC operator
	 <p>The diagram shows the BCS Control panel with the following components and labels:</p> <ul style="list-style-type: none"> <li><b>Inflow &amp; Downflow data</b>: A yellow label pointing to a blue rectangular display on the right.</li> <li><b>Sockets in hood</b>: A yellow label pointing to a power outlet icon.</li> <li><b>Fan</b>: A yellow label pointing to a fan icon.</li> <li><b>Light</b>: A yellow label pointing to a light bulb icon.</li> <li><b>UV light</b>: A yellow label pointing to a UV light icon.</li> <li><b>Alarm light &amp; Silence</b>: A yellow label pointing to a bell icon with a slash through it.</li> <li><b>FLOWCAR</b>: A blue logo with a circular arrow.</li> </ul>	
	<ol style="list-style-type: none"> <li>3. Turn on UV light for at least 20 minutes before using the cabinet to sterilise prior to use. Ensure UV light is OFF before starting work. UV is damaging, especially to eyes.</li> <li>4. Lift the sash to the recommended height.</li> <li>5. Turn on the BSC fan 10 minutes before beginning work to allow adequate air filtration.</li> <li>6. If the BSC alarms, check the air intake and exhaust grilles for obstructions (see section 3.4), check the pressure gauge reading indicating HEPA filter load (Inflow 0.51 to 61m/s, Downflow 28 to 33 m/s). If still alarming consult the manufacturer's manual, error indicator &amp; troubleshooting section (page 54). If the problems continue contact the designated person (int Tel: 31045 / 32195).</li> <li>7. If you are concerned that the cabinet is not drawing in air from the room you can confirm the inward airflow by holding</li> </ol>	

	<p>a piece of tissue at the middle of the edge of the viewing panel and ensure it is drawn in.</p> <ol style="list-style-type: none"> <li>8. Decontaminate the cabinet surface and surface-decontaminate all materials to be placed inside the BSC with 70% ethanol (EtOH) or equivalent. If you are working with agents that will not be killed by ethanol, use an alternative disinfectant e.g. Viraclean, Virkon™ or Biozidal FF®. Please note: <b>USE OF CHLORINATED OR HALOGEN MATERIALS IN THE CABINET MAY DAMAGE STAINLESS STEEL</b> if used, should be followed with a wipe down of sterile water or 70% EtOH.</li> <li>9. Bulky items, such as waste containers and suction collection flasks, should be placed to one side of the interior of the cabinet.</li> <li>10. Keep the work area of the BSC free of unnecessary equipment or supplies. Clutter inside the BSC may impede proper airflow and the level of protection provided. Consider using wire racks or shelving to increase airflow around front and rear grills.</li> <li>11. If hair is long ensure it is tied back. Wear a lab gown, closed-toe shoes and disposable nitrile or latex gloves (gloves should be of a standard for work with viruses) to protect yourself as well as your samples from contamination.</li> </ol>	
<b>2</b>	<p><b>Operation</b></p> <ol style="list-style-type: none"> <li>1. Active work should flow from clean to contaminated areas across the work surface.</li> <li>2. Always use mechanical pipetting aids. Mouth pipetting is not allowed.</li> <li>3. Arms should be moved in and out slowly, perpendicular to the front opening to minimize disruption of the air curtain and laminar flow.</li> <li>4. Do not work in a BSC while a warning light or alarm is signalling.</li> <li>5. Heat sources such as Bunsen burners are strictly prohibited inside the BSC as they significantly disrupt the laminar flow of air and present a significant fire hazard.</li> <li>6. Locate liquid waste traps inside cabinet and use an in-line HEPA filter to protect the vacuum line. If traps must be located on the floor, place them in a secondary container such as a durable plastic tray or box to prevent breakage.</li> <li>7. Work as far to the back as possible, but within comfortable reach.</li> </ol>	BSC operator
<b>3</b>	<p><b>Cleaning and disinfection</b></p> <ol style="list-style-type: none"> <li>1. When work is completed, all equipment and supplies from the BSC should be decontaminated and removed from the cabinet.</li> <li>2. The interior surfaces should also be wiped with an appropriate disinfectant that would kill any microorganisms that could be found in the cabinet. Corrosive chemicals such as bleach should be avoided, but if used, should be followed with a wipe down of sterile water or 70% EtOH.</li> <li>3. Allow the cabinet to run for 10 minutes.</li> </ol>	BSC operator

	<ol style="list-style-type: none"> <li>4. Turn on the UV for 20 minutes as you vacate the room. Ensure someone is tasked to turn the UV light off after 20 – 30mins.</li> <li>5. All used PPE i.e. gloves, gowns, masks etc. and, used consumables after preparation must be placed into the yellow biohazard bins for incineration. Ring the designated person (int Tel: 31045 / 32195) to arrange the collection of the full biohazard bin.</li> </ol>	
<b>4</b>	<p><b>Ultraviolet (UV) lights</b></p> <ol style="list-style-type: none"> <li>1. The UV lamp must be cleaned fortnightly to remove any dust and dirt that may block the germicidal effectiveness of the ultraviolet light.</li> <li>2. When operating the UV lamp ensure the cabinet sliding sash is closed (will not work if open).</li> <li>3. UV lamps must be turned off when the room is occupied to protect eyes and skin from UV exposure, which can burn the cornea and cause skin cancer.</li> </ol>	BSC operator
<b>5</b>	<p><b>Spills</b></p> <p>In the event of a spill within a BSC, follow these steps:</p> <ol style="list-style-type: none"> <li>1. Alert the others in the laboratory and vicinity.</li> <li>2. Leave the cabinet turned on.</li> <li>3. While wearing gloves, spray or wipe cabinet walls, work surfaces and equipment with disinfectant equivalent to 1:10 bleach solution. If necessary, flood the work surface, as well as drain-pans and catch basins below the work surface, with disinfectant for a contact time of at least 20 minutes.</li> <li>4. Report the spill to the designated person, who will report the spill to the Health &amp; Safety and Occupational Health if a GMO, select agent or toxin is involved.</li> <li>5. Soak up disinfectant and spill with paper towels. Drain catch basin into a container. Lift front exhaust grill and tray and wipe all surfaces. Ensure that no paper towels or solid debris are blown into the area beneath the grill.</li> <li>6. All clean-up materials for disposal must be placed in the biohazard waste container.</li> <li>7. Wash hands and any exposed surfaces thoroughly after the clean-up procedure.</li> </ol>	BSC operator & designated person
<b>6</b>	<p><b>Other considerations</b></p> <ol style="list-style-type: none"> <li>1. All repairs to the BSC must be performed by a qualified technician. Any malfunction of the BSC must be reported and repaired before the BSC is used again.</li> <li>2. The BSC must be decontaminated before filter changes and before being moved. The most common decontamination method is by fumigation with formaldehyde gas. BSC decontamination may only be performed by a certified contractor.</li> </ol> <p>Contact the designated person if you have any questions. The designate person is the Research Governance Manager (int Tel: 31045 / 32195).</p>	BSC operator & designated person

## **8. Changes from last revision**

Added control panel diagram. BSC Inflow and Downflow ranges added.