Standard Operating Procedure

Title: USE AND MAINTENANCE OF EVOS XL MICROSCOPE

Location: CBE Laboratories

1. PURPOSE

To describe the use and maintenance of the Evos xI microscope, located in the CL2 CBE laboratory H27

2. <u>SCOPE</u>

This SOP applies to CBE personnel using the Evos xl microscope for the visualisation and capture of cell culture images. The microscope is assigned to a single centre project, although it will be made available for general use, users must first contact the responsible person if they intend to use the microscope. Only authorised and trained personnel may use the microscope.

3. RESPONSIBILITES

3.1. Authorised laboratory staff shall:

- (i) Work in accordance with the instructions for operation and maintenance detailed in this SOP and the operator manual
- (ii) Carry out the routine inspection, cleaning and maintenance of the equipment, as required.
- (iii) Record any adverse events and alarms that indicate non-conformance or malfunction on the Maintenance Record and notify the Laboratory Manager/Responsible Person.

3.2. The Responsible Person/Laboratory Manager shall:

- (i) Ensure that authorised laboratory personnel are given suitable information, instruction, training and supervision in the correct use and maintenance of the equipment.
- (ii) Investigate any reported problems, adverse event, alarms or non-conformities associated with equipment usage.
- (iii) Organise the maintenance, repair or servicing of the equipment by trained and authorised contract / service personnel.
- (iv) Ensure the equipment undergoes annual PAT testing, in coordination with the responsible person / authorised laboratory staff
- (v) Ensure that prior to authorising the commencement of maintenance, repairs or servicing that the equipment is suitably disinfected and that a "Decontamination Certificate" is issued.

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4. EQUIPMENT AND MATERIALS

- 1. Evos xl microscope and accessories
- 2. Appropriate PPE (labcoat, gloves, eye protection where appropriate)
- 3. 1% Virkon
- 4. 70% IMS

5. PROCEDURE

5.1. Operating Environment and Portability

- (i) The Evos xl microscope is designed to be portable for use inside or outside a Biological Safety Cabinet.
- (ii) When moving the Evos ALWAYS lock the stage with the stage lock pin before moving. Refer to Operator Manual for instructions on moving / transporting the Evos to another bench, or in and out of a BSC, or to another facility.

NOTE: The stage pin is kept on mouse cable for safe keeping when not in use.

(iii) Place the microscope on a level surface away from vibrations (noting that image clarity may be reduced within a BSC due to vibration). Allow 5cm free space at the back of the microscope for proper ventilation and site away from direct light sources (e.g. windows) as this may affect the image.

NOTE: Operating temperature range is 4°–32°C and relative humidity range is 30–90%.

CAUTION: NEVER subject Evos to UV sterilisation as this can degrade the materials. Damage from UV exposure is not covered under the manufacturer's warranty.

5.2. Basic Operation

The EVOSxI microscope has two types of controls: mechanical and software. Mechanical controls include the stage X-Y axis knobs, focusing knobs, and the objective selection wheel. Software controls are located in the control bars at the bottom of the display screen.

- Refer to Figure 1 for Evos operational controls
- Refer to Figure 2 for Evos software control bar

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Figure 1. Evos Operational Controls

- 1. Power switch
- 2. Power input jack
- 3. USB and DVI ports
- 4. Coarse stage positioning knobs
- 5. Stage X-axis knob
- 6. Stage Y-axis knobs
- 7. Focusing knobs
- 8. Objective selection wheel
- 9. Phase annuli selector
- 10. Condenser slider slot



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Figure 2. Evos software control bar

- 1. LIGHT ON/OFF button
- 2. Illumination slider
- 3. Capture button
- 4. Save button
- 5. Selected objective



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- 1. Turn on the microscope with the power switch on the right side of the microscope base [Fig1; 2]. The microscope will take approximately 30s to load the software.
- 2. Plug the USB flash drive into one of the USB ports on the right side of the microscope arm [Fig1; 3]. With the exception of during image saving, the USB can be moved freely in and out of the port.
- 3. Place the sample on the stage, using a vessel holder if needed.

NOTE: Appropriate PPE must be worn when operating the microscope within the CL2 laboratory, and when using the microscope with biological and chemical materials.

- 4. Set magnification with the objective selection wheel on the front of the microscope [Fig 1; 4].
- 5. Turn on illumination with the LIGHT ON button located on the left side of the control bar [Fig 2; 1]. The Evos has a 3 megapixel colour camera and red pixels show when the image is saturated.
- 6. Turn the phase annuli selector to the position that corresponds to the selected objective and desired contrast method [Fig1; 9]. The software automatically updates to show the objective [Fig 2; 5]
- 7. If required, insert the *Diffuser slider* (brightfield, 10x, 20x, 40x) into the slot on the condenser assembly [Fig 1; 10]. This will cut down the light from the LED source. *Note:* Use a condenser slider as needed for optimal image quality during brightfield observations.
- 8. If ambient light interferes with phase contrast image quality, use the condenser shield [Fig 1; 10]
- 9. Adjust the illumination intensity as needed, using the slider on the software control bar or the mouse scroll wheel [Fig 2; 2]. Use this to reduce image saturation.
- 10. Focus the sample with the focusing knobs [Fig 1; 7].
- 11. To add a scale bar, select the scale bar option in the View section of the software control bar [Fig 2; 6]. Scale bars can be dragged to any point on the image. Double left click with the mouse on an image to digitally zoom in, scale bars will automatically change (Refer to operator manual for more information on using digital zoom).
- 12. Click the Capture button [Fig 2; 3]. The image is stored in a temporary memory buffer
- 13. Click the Save button to save the image to the USB drive [Fig 2; 4]. Refer to operator manual for further details on saving files, file types etc.
- 14. When work is complete, switch the microscope off at the power switch.

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5.3. Advanced Operation

Refer to operator manual for the following functions;

- User Account; saves user-specific settings including Quicksave [Fig 2; 7]
- Quicksave; saves to a specified file on the USB drive automatically [Fig2; 8]
- Timelapse Tool; minimal interval 10s, automatically creates avi. video files [Fig 2; 6]
- Cell Count Tool; applies grids to images to enable easy manual cell counting [Fig2; 6]
- Image Review Tool; review captured images from USB [Fig2; 6]
- Software updates

5.4. Preventative Maintenance of the Evos Microscope

5.4.1. Cleaning procedure (CAUTION: Never subject EVOS to UV sterilization)

- (i) Turn power OFF
- (ii) Clean the LCD display
 - o use a soft, dry, lint-free cloth to wipe off any dust from the screen
 - o clean the LCD display with a non-alcohol based cleaner made for flat-panel displays.
 - o <u>Do not</u> spray cleaning fluid directly onto the screen, as it may drip into the display or optics.
- (iii) Lightly wipe EVOS working surfaces (stage top, focusing knobs, objective selection wheel, housing) with paper towels or Kimwipes dampened with 70% IMS.
 - <u>Do not</u> allow solution to get into lubricated areas, such as the stage roller bearings, or any points of rotation such as axles for the stage knobs, condenser wheel, etc. Do not soak any surface in solution.
 - NEVER spray liquid anywhere on the EVOS. Always wipe surfaces with dampened paper towels instead.
- (iv) If it is necessary to clean the condenser, do not apply solution directly to the condenser assembly. Instead, select the desired phase ring, and then cover the condenser with clear plastic wrap and wipe the wrap with sterilization solution.

5.4.2. Objective Lens Care

- (i) Clean each objective periodically or when necessary with a lens paper and lens cleaning solution. To avoid scratching soft lens coatings, do not use non-optical materials.
 - To protect all optical components of the microscope, if possible use the dust cover when the microscope is not in use.

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5.4.3 Equipment Malfunction

- (i) If any part of the equipment fails or malfunctions, seek advice from the Laboratory Manager or Responsible Person. Never disassemble or service the microscope yourself. Unauthorized repairs may damage the microscope or alter its functionality, which may void the warranty. With permission of the Responsible Person, consult the operator's instruction manual to access fault finding and troubleshooting procedures.
- (ii) If any problem cannot be rectified according to the troubleshooting procedures a "do not use" notice must be put on the microscope and the Laboratory Manager informed. The manufacturer must be contacted for advice and external maintenance or servicing might need to be arranged.
- (iii) All problems and corrective actions must be recorded in the preventative maintenance log.
- (iv) If the equipment fails to work or malfunctions and cannot be rectified according to troubleshooting procedures detailed in the Operator and Users Manuals the Laboratory Manager must be informed and a "Do Not Use" notice should be posted on the equipment. Contact the manufacturer for advice and coordinate with the Lab Manager for external maintenance and servicing.
- (v) External maintenance and servicing of the equipment can only be performed after it has been suitably disinfected (refer to SOP003 for further details) and a 'Decontamination Certificate' has been issued (a pro-forma is available on the LEARN system for the CBE).

6. DOCUMENTATION

QS form - 009 Equipment decontamination certificate QS form – 018 Preventative Maintenance, Inspection and service record

These forms can be found on the Loughborough University LEARN module for the Centre of Biological Engineering under QS-Form templates.

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SOP Version History

Version Reviewed	Date Revised/ Reviewed	Revision Summary	New Version Number
1.0	01.10.12 P.Hourd	Annual Review – Revised format	2.0
2.0	04/02/21 K.Sikand	Reviewed SOP, minor changes made no change in version number.	2.0

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