## **Technical Procedure for Microscopes**

Version 2

Effective Date: 10/18/2013

- **1.0 Purpose** This technical procedure shall be followed for the operation of the microscopes within the Trace Evidence Section.
- **Scope** This procedure applies to all microscopes located within the Trace Evidence Section.
- **3.0 Definitions** -N/A

### 4.0 Equipment, Materials, and Reagents

- Stereomicroscopes
- Polarized light microscopes
- Reflected light microscopes
- Comparison microscopes
- Optical micrometer

### 5.0 Procedure

### 5.1 New Instrumentation

**5.1.1** New microscopes shall be installed by a certified engineer according to the manufacturer's instructions.

# 5.2 Köhler Illumination

- **5.2.1** Setting up Köhler illumination is one of the most widely accepted techniques of ensuring that a microscope is properly aligned, exhibits high intensity and uniform illumination and is correctly focused. Köhler illumination adjustments shall be performed as needed.
- 5.2.2 Depending on a particular microscope and its design, setting up Köhler illumination may be possible in full or only in part. This procedure shall not apply to stereomicroscopes.

#### 5.2.3 Procedure:

- **5.2.3.1** Using the highest magnification dry objective (fixed objective) and looking through only one ocular, focus on a specimen.
- 5.2.3.2 Now looking only through the other ocular, focus the specimen by adjusting the diopter ring on the ocular.
- **5.2.3.3** Completely open the aperture diaphragm.
- **5.2.3.4** Remove one ocular or insert the Bertrand lens and focus and center the filament in the back focal plane. Replace the ocular.
- **5.2.3.5** Close the field diaphragm.

**5.2.3.6** Focus the substage condenser by bringing the leaves of the closed field diaphragm into focus.

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- **5.2.3.7** Using the substage condenser centering screws, center the substage condenser.
- **5.2.3.8** Open the field diaphragm so that the leaves are just outside the field of view.
- **5.2.3.9** Using the stage centering screws, center the stage.
- **5.2.3.10** Remove an ocular or insert the Bertrand lens and adjust the aperture diaphragm so that it is open at least two-thirds of the diameter of the back focal plane. Replace the ocular.
- **5.2.3.11** Rotate the nosepiece to the next objective and center the objective using the centering screws on the side of each objective. Do not move the stage or the substage condenser.
- **5.2.3.12** Repeat **5.2.3.11** for each additional objective.

### **5.3** Calibrations – N/A

### 5.4 Instrument Maintenance

- **5.4.1** All of the microscopes used within the Trace Evidence Section shall be cleaned and serviced annually by an outside vendor. A record of this cleaning and service shall be maintained.
- **5.4.2** Lenses (including condenser lenses, objective lenses and oculars) shall be cleaned during use as needed.

#### 5.5 Standards and Controls

- **5.5.1** An optical micrometer shall be used to determine the spacing of the ocular measuring graticule.
- **5.5.2** No measurements made on the microscopes in the Trace Evidence Section shall be considered critical measurements.
- **5.6** Sampling and Sample Selection N/A
- 5.7 Calculations N/A
- **5.8** Uncertainty of Measurement N/A
- **6.0** Limitations N/A
- 7.0 Safety

- **7.1** Avoid looking directly into the fluorescence source.
- 7.2 Do not spend long periods of time making observations at high light intensities.
- 7.3 When changing bulbs, ensure that the power cord is disconnected.

### 8.0 References

Abramowitz, M. Microscope Basics and Beyond. Lake Success: Olympus Corporation, 1985.

Birk, G. *Instrumentation and Techniques for Fluorescence Microscopy*. Sydney: Wild Leitz Pty Limited, 1984.

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Delly, J.G. Essentials of Polarized Light Microscopy. Westmont, IL: College of Microscopy, 2009.

McCrone, W.C., L.B. McCrone and J.G. Delly. *Polarized Light Microscopy*. Chicago: McCrone Research Institute, 1984.

Saferstein, R. Forensic Science Handbook. Volume II. Englewood Cliffs: Prentice Hall, 1988.

### 9.0 Records

• Microscope annual cleaning/maintenance records.

## 10.0 Attachments - N/A

Revision History		
Effective Date	Version Number	Reason
09/17/2012	1	Original ISO Document
10/18/2013	2	Added issuing authority to header

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