LOCAL JOINT HEALTH AND SAFETY COMMITTEE
DEPARTMENT OF BIOMEDICAL SCIENCES
STANDARD OPERATING PROCEDURE

1. PHOTO DOCUMENTATION OF DNA BANDS IN AGAROSE GELS

Effective Date: March 2002
Author: Ann Maslen, Technician
Purpose: To promote safe and proper use of the Photo Doc. Centre.
Approvals required: Faculty Supervisor, Local JHSC, EHS

2. DEFINITIONS:

Mutagen: An agent that causes cellular DNA damage. Ethidium bromide is a **potent mutagen**. Always wear a protective lab coat and handle stock ethidium bromide and ethidium bromide gels wearing disposable gloves.
UV light: Light in the 180-390 nm wavelength range. Overexposure can cause pain and injury, particularly to the eyes, causing blindness. Always wear approved UV protective safety glasses.

3. REQUIREMENTS:

All persons must have WHMIS training and have completed a departmental safely orientation, and read **M**aterial **S**afety **D**ata **S**heet for ethidium bromide.

4. TASK:

Use of Documentation Centre in Room 3646: This Centre is used to visualize and document bands on agarose gels created by DNA intercalating with ethidium bromide in the gel.

a) Rm. 3646 is always kept locked. The key is on the orange foot key chain marked KING. You will need the key, your gel on the tray, and a clean scalpel blade and tubes if you are planning on excising the bands of interest. Always handle the gel wearing gloves, but leave one hand bare to open doors etc.
b) The Photo Documentation Centre is by the window opposite the door. Ensure that the light box within the curtained area is the FOTO/UV26. If not, exchange boxes.
c) If you are planning on excising bands, put the acrylic plate provided as a cutting surface over the surface of the light box before sliding the gel off the tray onto the light box surface.
d) Turn on the Gel Print 2000i and when the screen "settles", press Live Video.
e) Turn on the incandescent (regular) light with the switch on the light. The switch on the Gel Print (Lamps) doesn't work.
f) The camera lens and filter are located inside the curtained area above the light box. Rotate the filter out of the way of the lens.
g) Watching the screen, centre the gel below the lens and using the handle behind the lens on the right hand side of the vertical shaft, raise or lower the lens until the gel fills the screen.
h) You may have to control the amount of light to see the gel properly. Rotate the aperture (lowest "dial" on the lens) to the right to reduce light or to the left to increase light.
i) Adjust the focus ("dial" above the aperture) until the wells are clearly in focus. If this can't be done you may have to readjust the distance the lens is from the gel.
j) Swing the filter back into place and turn off the incandescent light.
k) Close the curtains around the platform. **UV light can cause blindness. Never look at it directly without wearing proper eye protection!**
l) Reach under the curtain and turn on the UV light using the switch on the front of the light box **without looking at the light.** Using the arrow up or down buttons on the Gel Print 2000i adjust the time to about 2 seconds. Push the CAPTURE IMAGE button. The image will appear on the screen at the end of the time period set. If the band(s) is to be excised minimize the exposure

time to the UV light as it can damage the DNA. Switch the light on only after the parameters are set and turn it off immediately after the image appears on the screen.

m) Adjust the exposure time using arrow keys until the bands of interest are easily visible. It may be necessary to readjust the aperture if exposure times are excessive or if entire gel is very bright. CAPTURE IMAGE button must be pressed every time you want to see the results of changes to settings.

n) Once a suitable image is on the screen, move the switch on the Video Copy Processor to PRINT and push PRINT HARDCOPY on the Gel Print 2000i. **Return the Video Copy Processor switch to "UP" or paper will be wasted!**

o) If you are going to cut out the bands of interest make sure appropriate eye protection is worn and the extra acrylic plate provided as a cutting surface is in place. Approved UV protective eye wear is available beside Centre. Turn on the UV light and as quickly as possible trim away the excess gel using a clean scalpel blade to isolate the individual bands. Turn off the UV light and place each band in a separate Eppendorf tube.

p) When finished, make sure the printer, lights and Gel Print 2000i are turned off and the surface of the light box is clean.

q) In the book provided beside the Centre record the date, your faculty supervisor's name, your initials and the number of photos printed.

r) Make sure the door to the room is locked when leaving.

s) Back in your own lab, dispose of the gel in the waste bag labelled ethidium gels. When full, this bag is to be sent for Hazardous Waste Disposal.

5. CONTINGENCY PLAN AND REPORTING:

If the gel containing ethidium bromide touches bare skin, the area of contact should be washed with soap and copious amounts of water.

6. WASTE MANAGEMENT AND ENVIRONMENTAL RESPONSIBILITY:

Gels containing ethidium should be placed in the plastic bag labelled ethidium waste located in the fume hood in your own lab. When the bag is full, it should be tagged and a "Hazardous Waste Disposal" form filled out and sent to Environmental Health and Safety requesting hazardous waste pick-up.

7. REFERENCES:

MSD Sheet for Ethidium bromide in Lab Safety
Manual for Photo Documentation Centre In Rm. 3646

8. DISTRIBUTION OF COPIES:

Technicians, Graduate Students, Project Students and other University of Guelph employees documenting bands in agarose gels.
Dr. , Faculty Supervisor
Environmental Health and Safety
Local JHSC

Written by: Ann Maslen Staff Technician
Authorization: Supervisor
Date: June 25, 2002