

# INSTRUMENT OPERATION

## JEOL 100CX II

### GENERAL OPERATING INSTRUCTIONS

**Note:** Never try to use the scope unless green READY light is on (located next to HT button).

The small dewar should be filled with liquid nitrogen. To fill dewar with liquid nitrogen, cover microscope viewing window with foil wrapped shield. Obtain liquid N<sub>2</sub> from large tank in room 164 using small dewar flask. **BE CAREFUL.** Place funnel in liquid N<sub>2</sub> trap on microscope and fill - holds about 1 pint.

#### Pre-check:

The HT button should be out and NOT lit up. The filament emission should be turned fully counterclockwise. The accelerating voltage should be set at 80 KeV (80 button depressed and lit). The film advance button should be depressed for single (NOT AUTO).

#### A. Inserting specimen holder in column:

The specimen holder will accommodate 2 grids. The grid nearest the tip of the holder is #1, the other is #2. It is possible to go from one grid to another by using the round dial located opposite the specimen holder on the left of column. The dial is marked #1 and #2 which corresponds to #1 and #2 positions on specimen holder.

Match small brass screw on specimen rod with small notch on black plate to the right of column on the specimen exchange mechanism.

Push rod in and hold firmly, pump will come on. Wait until pump ceases and red light found on the back of the specimen exchange mechanism goes out. The "C" indicator on the rod will be facing the operator. Turn specimen rod clockwise (to the right) while holding it firmly; allow the vacuum of the column to pull the rod into the column. **DO NOT RELEASE ROD UNTIL IT IS ALL THE WAY IN.** The "O" indicator will be facing the operator. (If you let go of the rod, you will damage the tip.)

#### B. High tension and condenser focus:

1. Push HT button on (button on extreme right of left panel marked accelerating voltage).
2. Turn filament emission up to the stationary lock (located on knob).
3. The counterclockwise movement of the condenser control will spread the beam; clockwise movement will condense the beam. The outer of the two knobs is the course control, the inner is the fine adjustment.

To obtain proper illumination, turn condenser controls clockwise until crossover is achieved, i.e. the beam forms a spot image on the screen.

Center crossover spot with trans-alignment controls only. There are two, one on each side of the panel.

4. Spread beam by turning condenser control counterclock-wise. If the beam is left at crossover, it will severely damage specimen.

5. If objective aperture is not in, rotate the aperture insertion rod to the #1 setting. With the beam spread and magnification set to 1-2 K, center the aperture using the X and Y centering knobs on the aperture insertion rod. Continue in this manner until the desired aperture size is reached and centered.

6. Adjust magnification accordingly. When going down in magnification, always reduce beam intensity first by use of the condenser; otherwise, the sections will be damaged.

**Switching Off Microscope:**

1. Turn filament emission off (full counterclockwise).
2. Push HT button off.
3. Remove specimen holder by pulling rod out while turning the rod to the left (counterclockwise).

**DO NOT ATTEMPT TO TURN THE ROD WHILE IT IS FULLY INSERTED IN THE COLUMN!**

4. Place specimen holder into the black wooden case.

## TAKING MICROGRAPHS ON THE JEOL 100CX II

**Note:** The exposure settings are preset. They should read sensitivity of 6, and a shutter speed of 2. If they have been changed, seek assistance; otherwise, please do not change.

1. Press screen button to tilt, focusing screen at 45° angle. Adjust the external binocular microscope to sharpen focus on screen. Using the medium and fine focus controls of the objective lens, focus the image.
2. After you have attained best possible focus, press screen button to lower focusing screen.
3. With condenser knob, adjust beam intensity until green light appears over the exposure-sensitivity knob. Red light to the left indicates under-exposure, red light to the right indicates over-exposure.
4. After setting the proper illumination, raise the viewing screen and watch the specimen for drifting. It is helpful to use the viewing screen dot as a marker to determine if the image is shifting.
5. Once the image has stabilized, push the green Camera Film Advance button. After a few seconds, this button should light up showing “single”, indicating that a plate is now in position and ready for recording an image.
6. To take photograph, press PICTURE button and release. If the microscope is set for automatic film advance, there is no need to press the Film Advance button. The exposure will be made and the film will automatically be advanced to the box receiver.

### Removing Photographic Plates:

1. Turn filament emission and HT off.
2. Turn red safelight on. Make sure overhead lights are out.
3. Vent plate holder drawer by turning handle down to OPEN position. Door will pop open when ready. Pull drawer out. Take both empty and full plate boxes into darkroom. There will be one empty box sitting on the counter for use when reloading as below.
4. Open desiccator (located in button left panel of microscope) by turning knob up to where the word AIR is even with small dot on desiccator door (counterclockwise). WAIT UNTIL DOOR OPENS. Take loaded film box from desiccator and empty holder from darkroom counter. Place in microscope plate holder drawer carefully.
5. Slide drawer in as far as it will go. Close door, push and hold. Turn handle up to CLOSE position.
6. Refill individual plate holders for a total of 40 and place into box containing unexposed plates. Reset the unused film number by depressing the reset button until the proper number is reached (e.g. 40).
7. Place full unexposed box into desiccator carefully. Close door and turn knob down clockwise. The word PUMP will line up with small dot on desiccator door.

## JEOL FILM DARKROOM PROCEDURES

### Take the following into the darkroom:

1. empty plate holder - removed from microscope
2. full plate collector - removed from microscope
3. box of unexposed plates - kept in hallway refrigerator

Check the temperature of the Developer to be sure it is within the proper range (see chart on wall).

Turn safe light ON, conventional lights OFF, close and lock the door.

Open plate collector and remove plates.

Remove plates from the metal cassette and place them in the plate hanger, flip clip. If you are unable to develop, place exposed plates in yellow box, marked 100CX PLATES and cover.

Place all four plate hangers (12 plates) in the nitrogen burst rack.

Flip nitrogen burst switch to TIMED.

Place N-burst rack into developer tank (far left), set timer according to developer temperature, activate timer by depressing the white button in the middle of the timer.

While plates are developing, reload cassette and the empty plate holder. Load with emulsion side UP. The plate holder is spring loaded, so press down on the mechanism and place plates into the holder from the top. When all plates are in the holder, replace top, and secure lid.

When timer sounds, press black button below timer, transfer N-burst rack to the second tank from the left, wash for 90 secs.

Transfer N-burst rack to the third tank from the left, fix for 5 mins.

Transfer N-burst rack to wash water (far right), turn water on to slight overflow. Flip N-burst switch OFF. Turn blower and heater ON. Wash plates for 30 mins.

Transfer N-burst rack to dryer, dry completely. Once dry, turn blower and heater OFF.

Remove plates from hangers and place in plastic envelopes to protect emulsion.

### **CAUTION:**

To avoid damaging the emulsion, always handle plates and negatives by the edges. Always place unexposed plates in the cassette with the emulsion facing UP. If in doubt, the plates are packed in the boxes with two plates back to back with the emulsion side OUT, and the non-emulsion side facing IN.

## JEOL TEM FILM AND PLATE PROCESSING

The following procedure will be used to develop JEOL photographic plates:

1. **DEVELOPMENT**- Use full strength D-19 20 C for 3.5 min(BEST)

18 C for 4-5 min

19 C for 4.0 min

21 C for 3.5 min

22 C for 3.0 min

2. **RINSE** - to stop (neutralize) development, rinse in H<sub>2</sub>O  
5-10 sec.

3. **FIXING** - to dissolve the unactivated, undeveloped silver halide grains and harden the emulsion, use rapid fixer 5 min.

TURN NITROGEN BURST TANK OFF.

4. **WASHING** - to remove excess silver halide grains dissolved by the "hypo", use running water wash 30 min.

5. **RINSE** - for uniform drying, use Photoflo wetting agent - dip briefly

6. **DRYING** - in dryer, on holder racks  
2-3 hours

7. **STORAGE** - place negatives in plastic envelopes. Observation is made with a hand lens without touching the emulsion.

8. **SIGN SHEET** - please sign sheet on the door indicating the number of plates developed, plate numbers, date and name. This lets the lab personnel know when the developer should be changed.

**NOTE:** It is helpful to face the film or plates with the emulsion towards the front in processing so that more plates behind will not scrape the emulsion side. Uniformity is important.