KAISER PERMANENTE®
COLORADO LABORATORY

STAINING BLOOD, BODY FLUIDS AND SEMEN USING THE HEMATEK SLIDE STAINER

PRINCIPLE:

The automated self contained Hema-Tek slide stainer consists of two slide conveyor spirals, a stain platen and three controlled solution pumps. The spirals move slides from the slide loading area across the length of the platen. The slides undergo staining, buffering, rinsing and air drying and are delivered into a slide drawer as finished stained slides.

After processing of the peripheral blood or body fluid, smears may then be assessed for identification of WBC's, RBC morphology and platelet estimates. Semen specimens mixed with sperm diluting fluid are also stained and morphology is assessed at the Franklin lab using the Strict Kruger method.

SCOPE:

Medical Technologists and Medical Laboratory Technicians working in the Medical Office Laboratories

SPECIMEN REQUIREMENTS:

1. Peripheral blood smears prepared from an EDTA (purple) tube.
2. CSF obtained in sterile tube and cytocentrifuged processed slide.
3. Synovial fluid obtained in a plain (red top) tube or heparinized (green top) tube and cytocentrifuged processed slide.
4. Other body fluid obtained in a plain (red top) tube or EDTA tube and cytocentrifuged processed slide.
5. Semen specimens mixed with sperm diluting fluid and air-dried.

EQUIPMENT AND MATERIAL

1. Hema-Tek Wright Giemsa stain pak. Store at room temperature. Stable through expiration date noted on the kit. (KPID 0004 8044)
2. Methyl Alcohol, (reagent grade), 1 gallon. (KPID 3700 1047)
3. Glass microscope slides (KPID 0017 7861)
4. Hema-Tek slide stainer model #2000
5. Hema-Tek pump tube set. Siemens, Ref #00043344 (KPID 3600 0582)
6. Hema-Tek underplaten tubing, Siemens, Ref #05854863 (KPID 0005 7333)
7. Hema-Tek cannula set, Siemens, Ref #02502125 (KPID 0005 7332)
8. 4 X 4 Gauze, (KPID 0036 2439)

CALIBRATION
No calibration required

QUALITY CONTROL

1. The stain quality is checked once per day when patient manual differential is performed. Make two peripheral blood smears on the first manual differential of the day and label one slide as "QC" with the date and the other slide with the patient information according to the procedure. The following items are verified on the QC slide:
    a. WBC stained correctly
    b. RBCs stained correctly
    c. Platelets stained correctly
    d. Stain quality is appropriate for performing manual differential – not too light or dark.

    NOTE: Refer to the reference slide if adjustments to stain settings are necessary.
2. Record stain quality on the daily stainer maintenance log and mark with an "X" when there is no manual differential performed that day.
3. Once per quarter, review the reference slide and make the appropriate settings adjustments to match the stain. Document on the stainer maintenance log.
    a. The reference slide is obtained from RRL and will be provided to all MOLs twice per year.
    b. Keep the reference slide in a dark location close to the Hematek stainer. This can be placed in a coin envelope and taped to the side of the stainer or in an empty slide box or some similar process.

PROCEDURE

1. Prepare whole blood EDTA smears (See procedure)
2. Prepare body fluid cytocentrifuged slides (See procedure).
3. Prepare semen sample slides (See procedure).
4. Label the smears so that the accession numbers are on the opposite side and on the heel of the smears.
5. Place the slides in the grooves of the conveyor spirals. The side with the blood film or body fluid sample must face to the left of the operator. Place the slide so that the feathered edge of the blood film is toward the back of the instrument.
   NOTE: Do not put the accession number sticker towards the back of the instrument. The stickers might bend the reagent sensors.
6. The slides will now be conveyed along the platen where it will come in contact with the stain, buffer, and rinse solutions.
7. After rinsing, the slide is dried by a dryer fan and then delivered gently into the slide drawer.

CALIBRATIONS:
None

CALCULATIONS:
None

MAINTENANCE:

A. INSTRUMENT START UP

1. Check the reagent pak to ensure adequate volume. Replace pak as needed and note on the maintenance log.
2. Prime all the reagent lines
   a. Place all the cannulas back into the proper reagent container; they are marked A, B, and C.
   b. Hold the lever in the prime position until you see all reagents appear on the platen.
3. Clean the platen

NOTE: The platen requires cleaning at regular intervals to achieve optimum staining.

   a. Flood the platen with methanol.
   b. With a soft tissue or gauze, wipe the platen from **RIGHT** to **LEFT ONLY**.
   c. With a Q-tip swab, clean the grooves on the platen
   d. Before use, prime the stainer with four glass slides.

B. INSTRUMENT SHUT DOWN

1. Clean the reagent tubing.
   a. Remove and wipe the stain, buffer and rinse cannulas from the pak.
   b. Place the stain cannula in a container of methanol and the buffer and rinse cannulas in a container of DI H2O with the tubings attached. Lift the operating lever to “PRIME.” Holding the lever in the prime position, continue to pump liquids through the stain tubings until they are thoroughly rinsed and clear solution appears on the platen.
   c. After the tubings have been cleaned, remove the cannulas from the methanol and water and continue to prime until the tubings are emptied of all liquids. Leave the cannulas out overnight and do not put them back into the reagent reservoir.
2. Clean the platen.
   a. Flood the platen with methanol.
   b. With a soft tissue or gauze, wipe the platen from **RIGHT to LEFT** only.
   c. With a Q-tip swab, clean the grooves on the platen.
3. Release the pressure on the pump tubing overnight and on weekends by pressing the operating lever in the down position.
4. Turn the instrument OFF. The switch is located on the left rear of the instrument.

NOTE: See the Hematek slide stainer maintenance log for daily, weekly and periodic maintenance procedure. Refer to the Ames Hema Tek manual for other maintenance procedures.

C. REAGENT STAIN PACK

1. Once received, label the top of the pack as 1, 2, 3, etc.
2. After every third stain pack, all of the cannula, tubings and underplaten tubings are changed and documented on the maintenance log.

REPORTING RESULTS:

Refer to the Manual Differential procedure

LIMITATION OF THE PROCEDURE:

Abnormal Staining can be due to:

1. Smear was inadequately dried.
2. Slides were not clean.
3. Warped slides will stain unevenly
4. Slide stainer must be primed or poor staining will result.
5. Stain tubing, cannula, spouts and orifice may be plugged
6. Dirty platen or grooves result in uneven, low quality staining.
7. A bent platen hinders diffusion of stain solution, buffer and rinse

CRITERIA FOR REJECTION:

1. Clotted specimen.
2. Unlabeled or incorrectly labeled specimen.

DISTRIBUTION:

Medical Office Laboratories using the Hematek Slide Stainer

REFERENCES:

1. Henry, J.B., Clinical Diagnosis and Management by Laboratory Methods, 18th edition, pp. 583-585.
2. Aerospray hematology slide-stainer cytocentrifuge user manual. Wescor, Inc., 1992
3. O'Connor, Barbara H., A color atlas and instruction manual of peripheral blood cell morphology, Williams and Wilkins, Baltimore. 1984 p.1
4. Cytopro cytocentrifuged user manual, Wescor, Inc., 1993.
5. Kheldsber, C.R. and Knight, J.A., body fluids, 3rd edition ASCP Press, Chicago, IL, 1993.
6. Bibbs, marlice, M.D., Sc.D., Comprehensive Cytopathology, W.B., Saunders CO., Philadelphia, PA., 1991, p. 551.
7. Strasinger, Susan king, Urinalysis and Body Fluids, F.A. Daurs CO., Philadelphia, PA., pp. 139, 143, 167.
8. College of American pathologists, 1993 CAP survey manual, appendix 11A, 1993.
9. Hema-Tek slide stainer operating manual, Ames, CO., Division of Miles Laboratories.