Spinal Fluid Cell Count

Principle

The analysis of cerebrospinal fluid (CSF) for cell count and wbc differentiation is a useful and important diagnostic tool. Diseases from acute bacterial meningitis to tuberculosis meningitis, multiple sclerosis, polyneuritis spinal cord compression, brain tumor, cerebral atrophy and a simple bloody tap can be initially differentiated with this information.

Policy

Cell counts must be completed as soon as possible or within 1 hour of being received in the laboratory.

Safety

All specimens, reagents and controls should be handled as though capable of transmitting infectious diseases. Wear appropriate personal protective equipment when running patient samples or performing scheduled maintenance. Refer to: Policy and Procedures Safety Manual Infection Control and Procedures 11-085-01.

Materials and Reagents

10% Glacial Acetic Acid (Prepared by Sherman Way Regional Laboratory)
Microscope
In-Cyto disposable C-Chip hemocytometer
Sterile Transfer Pipet
Cytology Funnel and Caps
Cytology Clips
Cytospin Centrifuge

Specimen Collection

CSF collections are made by the physician and brought directly to the laboratory. If a complete CSF study is ordered, they should arrive in either three (3) or four (4) tubes, labeled 1-3/4 in order of collection.

If you received 3 tubes, analysis of samples should be:

Tube #1 - Chemistry Analysis
Tube #2 - Microbiology Analysis
Tube #3 - Hematology Analysis

If you received 4 tubes, analysis of samples should be:

Tube #1 - Hematology Analysis*****
Tube #2 - Chemistry Analysis
Tube #3 - Microbiology Analysis
Tube #4 - Hematology Analysis*****

Specimen Collection, continued

Erythrocyte and leukocyte counts should be performed as soon as possible after the specimen is obtained because cells lyse on prolonged standing and accurate counts become impossible. Furthermore, lysis of erythrocytes will cause artificial "xanthochromia". **If specimen is clotted do not perform cell count. Notify nursing staff or provider that specimen is clotted**.

*** If there's only one (1) cell count order, then the default tube to use is tube #4**.

Note: Always use sterile technique when handling specimens.

Procedure

GROSS EXAMINATION

Step	Action
1	**Note physical appearance**: normal CSF is crystal clear. Color should be evaluated by holding the sample beside a tube of distilled water and a clean white paper. Bloody specimens should be centrifuged to observe for xanthochromia (pale pink to pale orange or yellow color). Report the presence or absence of xanthochromia. **Report the appearance as**: bloody, clear, cloudy, clotted, hazy or slightly hazy.
2	Note the volume of the CSF and write the total volume on the log. If the volume is </= 2mL and additional tests (other than a complete CSF study) have been ordered, contact the Provider. Ask the Provider to list the tests desired in order of priority due to the limited amount of specimen submitted. Contact a supervisor if needed.

COMPLETING CELL COUNT

Step	Action
1	Load by capillary action both sides of hemocytometer chambers with 10uL of undiluted well mixed fluid specimen (more squares should be counted for low number of cells). **Note:** A dilution may be necessary for high counts.
2	Using 40x magnifications, count the RBC's and WBC's in the appropriate number of large squares in both chambers.
3	The counts from each chamber must agree within 10% or the count must be repeated. Use the average of the two sides for the calculation.

Procedure, continued

Step	Action
4	Use the following formula to calculate number of cells/mm3: $\frac{\text{Number of cells counted x depth (10) x dilution}}{\text{Number of large squares counted}} = \text{cells/mm3}$ Cell counts and calculation of cell counts can be difficult. It is imperative that counts be calculated correctly so when in doubt consult with your co-worker or supervisor. **Note:** for **Mod or High WBC Counts;** prepare 1:20 dilution using 10% Glacial Acetic Acid. Let dilution stand 3-5 minutes for complete hemolysis
5	Discard hemocytometer after use. **Note:** C-Chip Hemocytometer is for single use only, should be clean, and free of scratches.

Quality Control

If the specimen contains White Blood Cells, make a slide of the specimen by following the directions in Cytospin Slide Preparation procedure, Hematology P&P HEM.03-0110.

Performing Differential

Follow the steps below to complete the differential count on body fluids:

Step	Action
1.	If the total cell count is low, make a chamber differential count by identifying the cells in the counting chamber using the high power dry lens (40X).
2.	If the total cell count is high, make a smear using unspun CSF or the sediment from a centrifuged specimen. Stain it on the slide stainer. Cytospin procedure (04-190) can also be performed.
3.	Count 100 WBC's, differentiating between neutrophil, lymphocyte, monocyte, eosinophil and basophil.
4.	If blasts, atypical cells or unidentifiable cells are present, refer smear to Pathologists for review. Refer to Hematology P&P HEM.03-0250 (Smear for Pathology Review) for procedural specifics.
5.	The slides will be saved for a month.

NOTE: Cell and parasite identification can be difficult. If you have any doubt of the correct identification, you may consult your co-worker, supervisor, or pathologist for assistance.

Result Reporting

Follow the steps below to complete the reporting of spinal fluid counts:

Step	Action
1	Results to be entered are: • Total Volume (mL) • Xanthochromia: Yes or No • Color: Colorless, Yellow, Pink, Red • Appearance: Bloody, Clear, Cloudy, Clotted, Hazy or Slightly Hazy • RBC Total • WBC Total • WBC Differential: Perform 5 part differential; enter result percent Neutrophils, Lymphoytes, Monocytes, Eosinophils, Basophils. Report unidentifiable and immature cells under comments. • Comments: Unidentifiable cells and Immature cells • CSF Macrophage & Mesothelial (if applicable): Report as Occasional, Few, Moderate, and Many
2	Results are manually entered in Cerner under the Accession Result Entry (ARE) mode. For detailed instructions of entering results, please refer to Laboratory Informatics – Cerner Genlab Policies & Procedures Manual, "*Resulting in Cerner GenLab: Manual Entry*" LIS.SCPMG.041 document.

Reference Range

Appearance:	Clear
Red Blood Cells:	None
White Blood Cells:	0-8 Cells

Reference

Todd-Sanford. Clinical Diagnosis of Laboratory Methods 18th Edition. Davidson and Henry, M.D.

Brays Clinical Laboratory Methods 7th Edition. p. 512.

Document History Page

Change type: New, Major, Minor etc.	Changes Made to SOP – describe	Name of responsible person/date	Med. Dir. Reviewed/ Date	Lab Manager reviewed/ date	Date change Implemented
Minor	Updated format and revised index number.	Julius Salomon, 7/1/17			