



TRAINING UPDATE

Lab Location: GEC
Department: Core

Date Distributed: 8/1/2012
Due Date: 8/31/2012

DESCRIPTION OF PROCEDURE REVISION

Name of procedure:
Cytospin CSF / Body Fluid Slide Preparation GEC.H03 v001
Description of change(s):
<p>Section 5: Add sample chamber size; update slide labeling and Cytospin programming, add manual staining</p> <p>Section 6: Add documents</p> <p>Section 9: Add Maintenance Log</p>

Document your compliance with this training update by taking the quiz in the MTS system.

Approved draft for training (version 001)

Non-Technical SOP

Title	Cytospin CSF / Body Fluid Slide Preparation	
Prepared by	Leslie Barrett	Date: 12/22/2009
Owner	Cynthia Reidenauer, Robert SanLuis	Date: 6/16/2012

Laboratory Approval		
Print Name and Title	Signature	Date
<i>Refer to the electronic signature page for approval and approval dates.</i>		
Local Issue Date:		Local Effective Date:

12 month (or new) management review and approval: Signature acknowledges SOP version remains in effect with NO revisions.		
Print Name	Signature	Date

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1. PURPOSE

Cyto centrifugation forcefully sediments cells from suspension onto a vertical micro slide as the suspension medium is absorbed by a blotter. This technique allows preparation of Body fluid/CSF slides, which are superior to slides made the conventional way even with low cell counts.

2. SCOPE

This procedure applies to all personnel at the Germantown Emergency Center.

3. RESPONSIBILITY

The Technical Supervisor of the Core Laboratory is responsible for the maintenance and the annual review process of this procedure.

4. DEFINITIONS

None

5. PROCEDURE

A. Material Required:

- Cyto centrifuge
- Slide/Sample chamber holder
- Disposable sample chamber (1mL), caps and white filter cards
- Microscope slides, frosted end
- Plastic transfer pipettes
- 0.9% NaCl
- 30% Albumin

Form revised 3/31/00

B. Specimen Preparation: (Prepare duplicate)

1. Label (2) slides with **patient last name first name**, accession number, **date**, and **source**.
If not enough sample, use blank cyto well for balance.
2. Prepare sample chambers:
 - a. Place a slide into the Cyto-Tek specimen chamber holder.
 - b. Place Cyto-Tek specimen chamber into the holder and snap it in place.
Note: Refer to the [Cyto-Tek operator guide for step by step instructions \(with pictures\)](#) for assembling the specimen chamber.
3. Place sample chamber into rotor with the funnel-facing center. Ensure that centrifuge is balanced.
4. To ensure that a monolayer of cells is produced the fluid should have less than 300 WBC/uL and less than 5,000 RBC/uL when five drops of fluid are placed in chambers. To arrive at these counts, refer to dilution chart below.

Dilution Chart

WBC Count	Dilution factor	Drops fluid	Drops 0.9% NaCl	Drops in chamber
0-300	None	5	0	5
301-600	1:2	5	5	5
601-900	1:3	5	10	5
901-1200	1:4	3	9	5
1201-2000	1:6	3	15	5
2001-3000	1:10	1	9	5

5. If WBC count is greater than 3000/uL, dilute to 3,000 and then use above chart.
6. If RBC count is greater than 5,000/uL in diluted sample, then continue to dilute until less than 5,000/uL.
7. For viscous fluids make 1:1 dilution with saline and skip step #8.
8. Place one drop 30% Albumin into chamber first then add 5 drops of fluid or diluted fluid.
9. Press caps onto chamber slowly so as not to push prematurely onto slide.

C. Cyto-Tek Centrifuge Procedure:

1. Turn on machine. Switch is located on front lower left side of machine.
2. Open lid to machine by depressing button on top left side.
3. Place rotor with top in place into machine.

4. Close lid and listen for audible click indicating lid is locked.
5. Program Cytospin as follows:
 - a. Press: **Set time**, then **5**, then **Enter**
 - b. Press: **mode** key and use arrow key to set speed to 20
 - c. Press: **Set Speed**, then **20**, then **Enter**
 - d. Press: **mode** key again and use arrow key to set time 5
 - e. Press **Start**
 - f. When Cytospin stops an alarm will be heard
6. Disassembly/Staining:
 - a. Remove rotor from machine
 - b. Remove sample carriage
 - c. Open sample carriage, then pull sample chamber away from microslide. DO NOT brush the two against one another as this will smudge the cell preparation.
 - d. Air dry slide before staining. Stain with Wright's stain. Refer to manual [Wright Stain procedure \(GEC.H05 Diff-Quik Stain Kit\)](#) to stain slide.

D. Differential

1. Perform differential under oil immersion. If possible, count 100 cells; if not, count and report percentage of each cell type.

Note: If the patient is a known tumor patient or the physician suspects tumor cells in the fluid, always examine the slide under low power then proceed to oil immersion to perform the differential.

2. Technical Note:
Recovery –

Numbers of WBC's counted on chamber	Number of cells counted on cyto slide
0	0 – 40
1 – 5	20 – 100
6 – 10	60 – 150
11 – 20	150 – 250
20	250

3. If the slide and chamber counts do not agree according to the chart:
 - a. Too many cells on the cytocentrifuge slide – recount the white cell count.
 - b. No cells or not enough cells on the cytocentrifuge slide as compared to the chamber count – prepare a new slide making sure that the specimen is well mixed.

4. After the fluid is counted it is saved in the refrigerator for 30 days, after which it is discarded.

6. RELATED DOCUMENTS

[Diff-Quik Stain Kit, Hematology procedure](#)
[Shandon Cytospin User's Manual](#)

7. REFERENCES

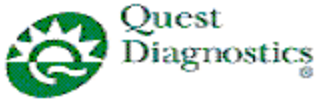
- Shandon Cytospin User's Manual.
- Nicky Sherwood and Joanne Combleet Stanford University Medical Center

8. REVISION HISTORY

Version	Date	Reason for Revision	Revised By	Approved By
		Supersedes GEC H022.001		
000	6/16/12	Update owner Section 5: Add sample chamber size; update slide labeling and Cytospin programming, add manual staining Section 6: Add documents Section 9: Add Maintenance Log	D. Patel	R. SanLuis

9. ADDENDA AND APPENDICES

[Cyto-Tek Maintenance Log](#) (see Attachment tab of Infocard)



- Germantown Emergency Center
- Shady Grove Adventist Hospital
- Washington Adventist Hospital

Cyto-Tek Centrifuge Maintenance Log

Make / Model #: _____

Serial number: _____

Month: _____

Year: _____

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31
Daily – when there is patient testing																															
As needed																															

Instructions:

Daily

1. Wipe the external surfaces with a clean cloth moistened with neutral detergent, pat dry.
2. Wipe the rotor bowl with a disinfecting solution, then wipe with a clean cloth moistened with neutral detergent.
3. Disconnect the drain hose at the rear of the instrument, and place a drain pan under the drain port of the instrument. Thoroughly rinse the rotor bowl with clear water. Completely dry the rotor bowl with clean, lint-free cloths.

Periodically

Soak the rotor in 5% sodium hypochlorite solution, then soak in a neutral detergent solution. Rinse with clear water, then thoroughly dry.

Weekly review:	Weekly review:	Weekly review:
Weekly review:	Weekly review:	Monthly review: