

Cytospin Slide Preparation

Principle Cytocentrifugation force sediments cells from suspension onto a vertical micro slide as the specimen's suspension medium is adsorbed by a blotter. Cytocentrifugation is performed in a bench-top centrifuge with a specially designed rotor and sample chambers that together constitute a cytocentrifuge. The rotor can accommodate up to 12 sample chambers that are each aligned with blotters and micro slides, and held together as units by the spring-loaded assemblers. The centrifugal force constructively flattens individual cells to enhance the display of their chromatin distribution patterns.

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	Cytospin Centrifuge	Cytology Clips
	Cytology Funnel and Caps	Sterile Transfer Pipet
	Microscope	Slides
	22 % Bovine Albumin Solution	Saline
	Hematek Slide stainer	

Specimen Collection Specimens from all body sites are eligible for cytocentrifugation but a uniform cell suspension is a prerequisite for satisfactory results. CSF specimens should be submitted in a standard CSF collection tube. All other specimens should be submitted in an EDTA tube to prevent clotting.

Step	Action
1	Label a slide with the patient's last name, first initial, date, and source.
2	Carefully place the disposable sample chamber (funnel), blotter and labeled slide in the assembler and release the spring. Check that: <ul style="list-style-type: none"> a) The exit port of the sample chamber- is centered within the hole in the assembler. b) The slide and sample chamber are aligned along their edges and seated against the lower lip of the assembler. c) The label on the slide faces the sample chamber.

**Procedure,
 continued**

Step	Action																									
3	Load the assembled cell collection devices into the support plate of the Cytospin rotor with the rotor out of the Cytospin. Check that each sample chamber ensemble is correctly assembled, securely seated and freely tiltable. The sample chamber ensembles should be symmetrically distributed around the support plate to permit balanced cytocentrifugation.																									
4	Add 1 drop of 22% bovine albumin to each sample chamber before adding the specimen or saline.																									
5	<p>Use the following dilution chart to figure the needed dilution for the WBC count and the RBC count. If either cell count exceeds the count on the dilution chart make a dilution of the specimen with saline which will contain less than 150 WBC/uL and less than 4000 RBC/uL. Add 8 drops of the dilution to the sample chamber.</p> <table border="1"> <thead> <tr> <th>WBC COUNT</th> <th>DILUTION FACTOR</th> <th>DROPS (FLUID)</th> <th>DROPS (SALINE)</th> <th>DROPS IN SAMPLE CHAMBER</th> </tr> </thead> <tbody> <tr> <td>0-101</td> <td>1:1</td> <td>8</td> <td>0</td> <td>8</td> </tr> <tr> <td>101-300</td> <td>1:2</td> <td>4</td> <td>4</td> <td>8</td> </tr> <tr> <td>301-700</td> <td>1:4</td> <td>2</td> <td>6</td> <td>8</td> </tr> <tr> <td>701-1500</td> <td>1:8</td> <td>1</td> <td>7</td> <td>8</td> </tr> </tbody> </table>	WBC COUNT	DILUTION FACTOR	DROPS (FLUID)	DROPS (SALINE)	DROPS IN SAMPLE CHAMBER	0-101	1:1	8	0	8	101-300	1:2	4	4	8	301-700	1:4	2	6	8	701-1500	1:8	1	7	8
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6	Press a closure cap onto each sample chamber.																									
7	Lock the lid of the sealed head onto the base and set the combination onto the tapered drive boss.																									
8	Close the Cytocentrifuge cover.																									
9	Program the Cytocentrifuge to accelerate Hi and spin at 600 RPM for 10 minutes and start.																									
10	Open the Cytocentrifuge and remove the head.																									
11	Unlock and remove the sealed lid.																									
12	Remove the sample chambers and place on a paper towel.																									
13	Open the assembler and remove the sample chamber, blotter and slide as a unit. Back the slide away from the blotter to avoid wiping off cells.																									
14	Air dry the slide.																									
15	When dry, stain the slide on the Ames Hematek slide stainer.																									

Reference

The Shandon Cytospin 2 Techniques, Tips, and Troubleshooting for Cell Suspensions in Cytology, Hematology, and all Disciplines, Gary W. Gill, The Johns Hopkins Medical Institutions Qualitative Cytopathology Laboratory, March 1982.

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			