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Section: Virology Manual	Subject Title: Genital/Peri-anal/Mouth/Nose	
	Skin Lesions	
Issued by: LABORATORY MANAGER	Original Date: March 14, 2001	
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GENITAL/PERI-ANAL/MOUTH/NOSE/SKIN LESIONS

I. <u>Introduction</u>

Specimens from genital, perianal and oro-labial (mouth/nose) lesions will only be examined for herpes simplex virus (HSV) unless otherwise requested. Specimens from skin lesions will be examined for both herpes simplex virus (HSV) and varicella-zoster virus (VZV). For other viruses requested, refer to Appendix XV (Virus Isolation and Identification) to ensure that the appropriate media is inoculated.

II. Collection and Transport

The roof of the vesicle(s) is disrupted. The fluid and cells released from the base of the lesion are collected using a sterile syringe and needle or a sterile swab. If the specimen is collected with a syringe and needle, aspirate viral transport medium into and out of the syringe several times, then express the contents into the viral transport container. Do not leave the needle and syringe in the transport container. (These should be discarded in an appropriate sharps container). If a swab is used, place the swab immediately into viral transport medium. Transport to the lab as soon as possible. If a delay in transport or processing is anticipated, the specimen should be kept at 4°C until processed. If a delay of more than 72 hours is anticipated, the specimen should be frozen at –70°C. Avoid repeated freeze-thaw cycles.

III. <u>Procedure</u>

A. Processing of Specimens:

Specimens for HSV and other viruses can be kept refrigerated for up to 72 hours. However, if VZV is specifically requested, the specimen should be set up immediately or stored at -70°C. Vortex patient sample in transport medium for 30 seconds. Remove excess fluid from the swab and discard the swab. Specimen in the transport medium can be transferred to a 2 mL cryovial. After preparation of slide (if needed) and inoculation of cultures, store cryovial at -70°C for 6 months. The original specimen container should be kept at 4°C for 1 week.

Refer to Appendix II for Shell Vial inoculation.

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B. Direct Examination:

- i) For genital, perianal and oro-labial (mouth/nose) lesions, prepare one double-well cytospin **only if requested**. Stain one well with HSV 1 monoclonal antibody and the other with HSV 2 monoclonal antibody.
- ii) For all other skin lesions, **always** prepare one double-well cytospin. Stain one well with herpes 1/2 bivalent antibody and the other well with VZV monoclonal antibody.

If a slide comes with the original specimen, it will be fixed in acetone for 10 minutes and stained in addition to the in-house prepared slide. The slide will be stained for VZV primarily and HSV if possible.

Refer to Appendix V for immunofluorescent staining techniques.

C. Isolation and Identification:

Specimen	Method	Cell Lines ^a	Incubation at 36°C	Stain ^b used
Oro-	Shell Vial	MRC-5	1 day	HSV1
facial/genital			1 day	HSV2
Skin	Shell Vial	MRC-5	1 day	HSV1
		MRC-5	1 day	HSV2
		MRC-5	2 days	VZV

MRC-5 = Human fibroblast cells

HSV1= Monoclonal antibody DFA stain for Herpes simplex 1

HSV2= Monoclonal antibody DFA stain for Herpes simplex 2

VZV= Monoclonal antibody DFA stain for Varicella zoster

See Appendix II for detailed shell vial procedure

IV. Reporting Results

- A. For genital, perianal, oro-labial specimens
 - a. Direct Examination:

Negative Report: "Negative for Herpes Simplex virus".

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Positive Report*: Insufficient Cells Report:		vir cellular material	us". to interpret test. Cultur	re to
b. Shell Vial: Negative Repo Positive RepoB. For skin lesion specir	rt*: "POS	ative for Herpes ITIVE for		
a. Direct Examination Negative Repo Positive Repo Insufficient Co	ort: "Nega "Nega rt* "Posi	ative for Herpes tive for	virus". ellular material to interp	pret
c. Shell Vial: Negative Repo	C	ative for Varicel "Negative for	Herpes Simplex virus"	

*Telephone all positive VZV results to ward/ordering physician and Infection Control.

Telephone all positive HSV results from neonates and post-partum women to appropriate ward/ordering physician.

When entering results in LIS use "control G" to send to appropriate infection control queue.

* When entering positive results in the Lab Information System (LIS), enter the virus name in the isolate window (under F7). See LIS Manual for entering results.

V. Reference

1. Gleaves, Curt A. et al. Cumitech 15A "Lab Diagnosis of Viral Infections". American Society for Microbiology, August 1994.