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Applicability Dearborn, Trenton, Wayne

Point of Care Wet Mount

Document Type: Procedure

I. PURPOSE AND OBJECTIVE:

- A. To describe how to perform microscopic wet mount examinations at the point of care (POC) for non-laboratory personnel designated as a provider (Doctor, Mid-Level Provider/Advanced Practice Provider, Resident).
- B. This document is only applicable to areas that are approved for testing under one of the laboratory's Clinical Laboratory Improvement Amendments (CLIA) certificates.

II. PRINCIPLE AND CLINICAL SIGNIFICANCE:

- A. A wet mount is the direct microscopic examination of vaginal discharge. Wet mount microscopic examinations are performed to evaluate patients for the presence of bacterial vaginosis, *Trichomonas vaginalis*, and yeast (*Candida* species).
- B. Non-specific bacterial vaginosis involves the overgrowth of bacteria that may include *Gardenerella vaginalis*, *Mycoplasma hominis*, *Mobiluncus* species, *Prevotella bivia*, *Prevotella disiens*, other *Prevotella* species, *Peptostreptococcus anaerobius*, *Peptostreptococcus asaccharolyticus*, *Peptostreptococcus magnus*, other anaerobic cocci, and anaerobic gram positive rods. The diagnosis of bacterial vaginosis is based on demonstrating 3 of the 4 following criteria:
 - 1. Thin, but profuse, vaginal discharge
 - 2. A pH greater than 4.5 (refer to the [pHizatest](#) procedure)
 - 3. A "fishy" odor after the addition of 10% potassium hydroxide (KOH) to a vaginal specimen
 - 4. Presence of clue cells on microscopic examination (wet preparation or Gram stain). Clue cells are vaginal epithelial cells with adherent coccobacilli.
- C. *Trichomonas vaginalis* causes trichomoniasis in both males and females, although the infection is more serious in female patients. Trophozoites are found in vaginal and prostatic secretions and urine. Females infected with *T. vaginalis* may produce foul-smelling, frothy yellow-green vaginal discharge. *T. vaginalis* infections are diagnosed by detecting live, motile flagellates (trophozoites) from direct saline

wet mounts. Microscope slides are examined under low (10X) and high power (40X) for the presence of actively moving organisms.

- D. Vaginal candidiasis in females is characterized by the production of a thick, yellow, milky vaginal discharge. *Candida* may be confirmed by microscopic examination of clinical specimens and the observation of budding yeast and/or pseudohyphae. The addition of KOH will aid in cell lysis and makes yeast easier to view.

III. SPECIMEN COLLECTION AND HANDLING:

Always follow established procedures for [Standard Precautions/Hand Hygiene](#) when collecting and handling a specimen. Hands must be washed or disinfected with antiseptic soap or an alcohol-based hand rub as outlined in the [Laboratory Infection Control](#) policy before and after gloves are used. Gloves must be worn when performing patient testing and changed between patients.

A. Patient Preparation

1. Verify the collection area is clean of fecal material prior to collection. Specimens contaminated with fecal material may cause false positive results for *T. vaginalis*. See the Limitations section for more information.

B. Patient Identification

1. Patients must be identified at the bedside using two identifiers (Joint Commission).

C. Specimen/Slide Labeling

1. If the specimen is taken to another location for patient testing, the swab may be placed in a tube with 0.85% sterile saline for transit to the microscope and must be labeled with patient name and identification (ID) number (Joint Commission). If the slide is prepared at the bedside, it must be labeled with the patient name and ID number.

D. Acceptable Specimen

1. Vaginal discharge

E. Specimen Collection, Handling, and Disposal

1. Verify that the specimen collection supplies are not expired.
2. Collect the specimen with a cotton or Dacron swab. Place the swab immediately in a small tube with <1.0 mL of 0.85% sterile saline.
 - a. Note: The tube must be labeled with the patient's name and ID prior to transport to the microscope.
3. Prepare to examine the specimen immediately after collection.
 - a. Note: If the specimen cannot be examined immediately, the swab may be placed in Amies transport media. This will allow the *T. vaginalis* organisms to remain viable for 24 hours. Do not refrigerate the specimen.
4. Upon testing completion, dispose of the collection supplies in biohazard waste and the slide in a sharps biohazard container.

F. Specimen Acceptability Criteria

1. Specimens (vaginal discharge) tested immediately that are stored in <1 mL 0.85% sterile saline
2. Specimens (vaginal discharge) tested within 24 hours of collection that are stored in Amies

media

G. Specimen Rejection Criteria

1. Specimens contaminated with fecal material
2. Specimens more than 1 hour prior to collection that have not been placed in Amies media
3. Specimens in Amies media collected more than 24 hours prior to examination
4. Refrigerated specimens
5. Specimens collected using Calgiswabs

IV. EQUIPMENT AND SUPPLIES:

- A. Bright Field Microscope
- B. Sterile Vaginal Speculum (Optional)
- C. Sterile Cotton Swab or Dacron Swab
- D. Specimen Tube
- E. Transfer Pipette
- F. Glass Microscope Slide
- G. Glass Coverslip
- H. Gloves

V. REAGENTS:

A. 0.85% Sterile Saline

1. Ingredients

- a. 0.85% Sodium Chloride

2. Handling

- a. Store at room temperature (15-30°C).

3. Expiration

- a. Do not use beyond the manufacturer's expiration date printed on the packaging.

4. Warnings/Precautions

- a. For *in vitro* diagnostic use only.

B. 10% Potassium Hydroxide Reagent Droppers

1. Ingredients

- a. 0.5 mL 10% KOH with 1% Dimethyl Sulfoxide

2. Handling

- a. Store at room temperature (15-30°C).

3. Expiration

- a. Do not use beyond the manufacturer's expiration date printed on the packaging.

4. Warnings/Precautions

- a. For *in vitro* diagnostic use only.

VI. MAINTENANCE:

- A. Microscope preventative maintenance is performed by an outside vendor on an annual basis. A copy of the record of maintenance is stored in the site-specific laboratory.
- B. POC personnel will round on a quarterly basis to confirm that the microscope is clean.
 1. The microscope should be kept clean and covered when not in use.
 2. The 10X and 40X objectives must be free of oil.
 3. If the microscope housing, objectives, lenses, condenser, stage, etc. are dirty, staff should clean with a lint-free cloth dampened with water.
- C. The annual professional preventative maintenance, inspection by POC staff, and cleaning, if needed, will be documented on the Fern Test and Wet Mount Microscope Cleaning and Maintenance Log attached to the [Fern Test](#) procedure.

VII. QUALITY CONTROL (QC):

- A. No QC is required for this procedure.

VIII. PROCEDURE:

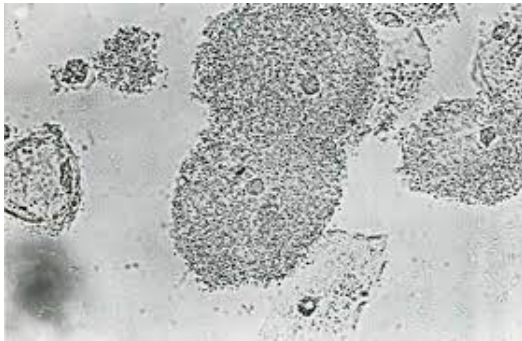
- A. Label a glass microscope slide with the patient's name and ID.
- B. Using a disposable pipette or the swab soaked in saline/Amies, apply the specimen to a small area on a clean microscope slide by rolling the swab back and forth.
- C. Place a coverslip over the sample.
 1. Alternatively, prior to the application of a coverslip, the specimen may be mixed with 1 or 2 drops of KOH if a clearing effect is needed for the visualization of yeast.
- D. Observe the slide using bright field microscopy using the 10X and 40X objectives. Scan the entire coverslipped area of the slide.
 1. Confirm that cellular material is present to verify that the slide has been adequately prepared.
- E. Upon completion of analysis, discard the swab, and specimen tube in a biohazard container and discard the slide and coverslip in a puncture-resistant biohazard container.

IX. INTERPRETATION OF RESULTS:

- A. Clue Cells: Examine the entire coverslip for clue cells (squamous epithelial cells covered with rods, the cells may appear fuzzy). Suspicious cells may be examined using the high power (40X) objective.



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- B. *Trichomonas vaginalis*: Examine the slide using the low power (10X) objective under low light. Observe for motile flagellates. Suspicious objects may be viewed using the high power (40X) objective. The organism is slightly larger than a white blood cell and oval-shaped. The flagella movement and undulating membrane should be visible under 40X.

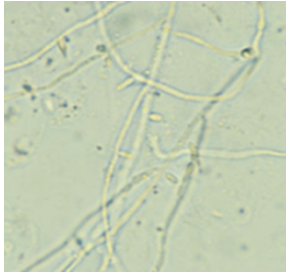


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- C. Yeast: Examine the wet mount with the low power (10X) objective under low light. Slowly adjust the illumination by moving the condenser so that the hyaline fungal elements can be seen. Examine the entire coverslip for budding yeast (round or oval) and/or pseudohyphae (chains of elongated yeast).



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X. RESULT REPORTING:

- A. Document the results in the Hospital Information System. The results must include the specimen collection/analysis date and time and the name of the person performing the collection/analysis.
- B. Quantitation is not necessary. Report either "seen" / "present" or "not seen" / "absent" for each of the following:
 1. Clue Cells
 2. *Trichomonas vaginalis*
 3. Yeast
 - a. Example of a report: "Clue cells seen. *Trichomonas vaginalis* not seen. Yeast absent. Collected and performed by John Doe 01/01/2022 0850"

XI. EXPECTED RESULTS:

- A. Clue cells: Absent/Not Seen
- B. *Trichomonas*: Absent/Not Seen
- C. Yeast: Absent/Not Seen

XII. UNEXPECTED RESULTS:

- A. Microscopic examination for clue cells, yeast, and *Trichomonas* is considered a definitive test and may be used for treatment when correlated with clinical findings. Any results exhibiting inconsistency with the patient's clinical status should be repeated or supplemented with additional testing and diagnostic procedures such as vaginal pH testing.

XIII. CRITICAL VALUES:

- A. None

XIV. SYSTEM DOWNTIME:

- A. Document results on the Point of Care Downtime Result Log found in the [Point of Care Downtime Result Recording, Reporting and Recovery](#) procedure.
- B. When Epic becomes available, enter the results in the Epic chart notes.

XV. LIMITATIONS AND INTERFERING SUBSTANCES:

- A. *Trichomonas* organisms will lose motility after one hour. Examine saline specimens immediately. Examine specimens preserved in Amies within 24 hours.
- B. *Trichomonas hominis* (intestinal parasite) is similar in shape to *Trichomonas vaginalis*. Specimens contaminated with fecal material containing *T. hominis* may be falsely reported as positive for *T. vaginalis*.
- C. Delay in reading the slide may yield false negative results.
- D. Specimens should not be refrigerated.
- E. Thickly prepared slides may obscure elements and lead to a false negative reading.

XVI. REFERENCES:

- A. [Point of Care Testing Approval Process](#)
- B. The Joint Commission. (2022) Standard NPSG.01.01.01 EP 1 in The Joint Commission. Comprehensive accreditation manual. Hospital edition. Oak Brook, IL: The Joint Commission.
- C. Barbara Haller, MD, PhD, Director, University of California, San Francisco – Department of Laboratory Medicine Zuckerberg San Francisco General Hospital and Trauma Center – Clinical Laboratory 1001 Potrero Avenue, San Francisco, CA 94110 <https://www.testmenu.com/zsfglab/TestDirectory/SiteFile?fileName=sidebar%5CSaline%20and%20KOH%20Vaginal%20Wet%20Mounts%202021.pdf>

Attachments

[POC Wet Mount Quality Monitor Checklist.pdf](#)

[POC Wet Mount Training and Competency Assessment.pdf](#)

[POC Wet Mount Training Guide.pdf](#)

Approval Signatures

Step Description	Approver	Date
CLIA Medical Directors	Muhammad Arshad: Chief, Pathology	2/28/2024

CLIA Medical Directors	Jeremy Powers: Chief, Pathology	2/21/2024
Policy and Forms Steering Committee Approval (if needed)	Jessica Czinder: Mgr, Division Laboratory	2/21/2024
CP System Medical Director	Ann Marie Blenc: System Med Dir, Hematopath	2/15/2024
	Caitlin Schein: Staff Physician	1/31/2024
Technical Director	Nga Yeung Tang: Tech Dir, Clin Chemistry, Path	1/29/2024
POC Best Practices	Jessica Czinder: Mgr, Division Laboratory	1/26/2024
	Jessica Czinder: Mgr, Division Laboratory	1/26/2024

Applicability

Dearborn, Trenton, Wayne

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