APPLICABILITY: Name of Policy

**PROVIDENCE CLINIC**

**POLICY AND PROCEDURE MANUAL**

**EFFECTIVE DATE:**

**POLICY #: 215 Attachment H**

**TYPE**:

**SUBJECT**: SALINE WET PREP

**APPROVAL**

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**Laboratory Director**

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**VP Physician Operations**

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**Executive Director**

**POLICY:**

*Trichomonas vaginalis* is a flagellated protozoan parasite causing infections primarily acquired by sexual intercourse. Vaginal secretions with an acute infection may be liquid, greenish or yellowish, sometimes frothy and foul smelling. As the infection becomes more chronic, the purulent discharge diminishes, with a decrease in the number of organisms. Infection in the male may be latent, with no symptoms, or may be present as self-limited, persistent, or recurring urethritis.

Bacterial Vaginosis (BV) is characterized by a foul smelling discharge consisting primarily of “sloughed” epithelial cells, usually with an absence of inflammatory WBCs. Besides the disease BV causes in the patient, this syndrome has been implicated in the etiology of premature labor and low birth weight adding to the importance of its diagnosis. Many of the epithelial cells in the discharge of Bacterial Vaginosis may be completely covered by dense aggregates of gram variable rods and coccobacilli. These epithelial cells are called “Clue Cells”. The presence of these Clue Cells is an indicator of BV.

Leukocytes (WBCs) may also be visualized on the saline wet prep. Their presence or absence may be useful to the clinician in the diagnosis and care of the patient and should also be reported.

**PRINCIPLE:**

*T. vaginalis* infections are diagnosed primarily by detecting live motile organisms from direct saline “wet” mounts of vaginal or urethral discharges or prostatic secretions. Microscope slides made from specimens can be examined under low and high power for the presence of actively moving organisms.

The microscope slide is also used to examine epithelial cells for to aid in the diagnosis of Bacterial Vaginosis.

**SPECIMEN:**

Vaginal, Urethral, Penile discharges; Urethral-mucosal scrapings; Urine

**REAGENTS:**

1. 0.85% NaCl (Normal Saline) – stored at room temperature
2. Pipettes, Glass slides, Coverslips – room temperature
3. Microscope slides – room temperature
4. Microscope (with 10x & 40x objectives or equivalent)

**QUALITY CONTROL:**

1. Check the normal saline with each use for clarity and no visible signs of contamination.

2. The microscope should be clean and the micrometer must be appropriately calibrated.

**PROCEDURE:**

1. Collect specimens with a cotton or Dacron swab, or speculum. Place specimen in a small amount of

0.85% NaCl (normal saline), approximately **.**5 - 1 mL.

2. Urine used for *Trichomonas* should be collected in a clean-catch urine container (preferably first

morning specimen) AND should be centrifuged prior to examination.

3. Apply the patient’s specimen to a small area on a clean microscope slide.

4. Immediately place coverslip over specimen to prevent drying.

5. Examine the wet prep with the low power (10x) objective and low light for motile flagellates, WBCs

and epithelial and Clue Cells. Suspicious objects can then be examined with the high power (40x)

objective.

**EXPECTED RESULTS:**

If motile flagellates are seen, then the trophozoites of *T. vaginalis* are present and the specimen should be reported as POSITIVE for Trichomonas.

If no motile organisms are seen then the specimen is NEGATIVE for Trichomonas.

Report the presence of Clue Cells and the presence and relative quantity of WBCs.

***Trichomonas vaginalis***:

Look for typical rapid, jerking movement of the trophozoite. T. vaginalis is a pear-shaped flagellate usually slightly larger than a WBC (PMN). Flagellar movement should be seen and sometimes movement of an undulating membrane along the length of the organism can be seen.

(\*NOTE\*) Motility can be enhanced by warming the specimen to 36 – 37oC (slide or tube).

**Clue Cells**:

Normal Vaginal Epithelial Cells will have only a few organisms associated with them and will have clear distinct edges. Clue Cells are epithelial cells covered with numerous and varied bacterial flora to a point where the outline or edge of the cell is masked and cannot be seen.

**WBCs**:

White Blood Cells are usually seen in cases of bacterial urethritis or gonorrhoeae. If WBCs are present they will usually appear as Polymorphonuclear Neutrophils (PMNs) ranging in size from 10 – 14 μm in diameter.

**LIMITATIONS:**

1. It is very important that specimens are examined within one (1) hours after collection. After 1 hour, organisms will lose their motility, particularly when they begin to dry out.
2. When the specimen is examined microscopically, always confirm that no fecal contamination is present. If the patient has a *Trichomonas hominis* intestinal infestation and the urogenital specimen becomes contaminated with fecal material, a false positive Trichomonas result may be reported because the two organisms are similar and difficult to distinguish on wet prep.
3. Calcium Alginate swabs (“Calgiswabs”) are not recommended due to tight adherence of the specimen to the swab.
4. Clue Cells may be difficult to identify on wet prep in some specimens and a gram stain may be required for further identification.

**REFERENCE:**

Baron, E.J., L.R. Peterson and S.M. Finegold (eds.). 1994. Bailey & Scott’s Diagnostic Microbiology, 9th ed. The C.V. Mosby Co., St. Louis.

DPDx Parasite Image Library. Centers for Disease Control and Prevention. Atlanta. GA.

Isenberg, H.D. (editor in chief ). 1998. Essential Procedures for Clinical Microbiology. American Society for Microbiology, Washington, D.C.

Garcia, Lynne S., & David A. Bruckner. 1993. Diagnostic Medical Parasitology. 2nd ed. American Society for Microbiology, Washington, D.C.