## TITLE: Semen Analysis: Post Vasectomy

### PRINCIPLE

Post vasectomy semen specimens are examined for the presence or absence of sperm 6-8 weeks following surgery to determine the effectiveness of the vasectomy.

***\*New\****

*The American Urological Association (AUA) Vasectomy Guideline recommends a careful evaluation of an uncentrifuged specimen, and does not recommend centrifugation of the specimen for further assessment. The AUA Guideline also recommends reporting both the presence and absence of sperm and the presence or absence of sperm motility on the patient report.*

# PERSONNEL

Medical Technologists and Technicians

# SPECIMEN

###### Patient Preparation: The semen sample should be collected by masturbation following a

###### two (2) to seven (7) day period of abstinence, 3 days is ideal; or as the doctor has directed, from any form of ejaculation.

Type of Specimen: The entire seminal ejaculate is collected by masturbation into a sterile plastic

container provided by the physician or obtained at the Out Patient Lab, along with instructions

and transported to the laboratory within one hour of collection. **\*See Procedural Notes\***

**Unacceptable specimens: Greater than 48 hours old or specimens transported in a condom. Cancel the test with a comment as to the reason.**

Handling Precautions: Handle all specimens using Standard Precautions.

## REAGENT PREPARATION & EQUIPMENT

10uL pipette and tips

Glass slides, cover slips, disposable transfer pipette, and microscope

## QUALITY CONTROL

1. We examine a Negative control smear and a Positive control smear and record on the QC log sheet. Slides are in plastic slide holders in the drawer between the microscopes. To verify the accuracy of the procedure, the test will be performed in duplicate by reviewing 2 slides made from the well mixed sample.

2. If counting sperm, Hemocytometer QC is performed using the Streck Cell-Chex on the day of a patient sample. It is performed and documented on our QC log sheets. QC is performed in the same manner as we do Body Fluid QC, alternating between Level 1 and Level 2.

We participate in the CAP program: CAP Survey - SEM

## CALIBRATION

N/A

### STEPWISE PROCEDURE

1. Mix the specimen and transfer entire amount with a transfer pipette to a conical centrifuge tube and cap (from Urinalysis).
2. Record the volume on our worksheets; this result is not reported out.
3. Mix the sample well and make 2 wet prep smears with 10uL of the specimen to observe for presence/absence of sperm and motility.
4. Make 1 more smear, pull style to stain with Wright-Giemsa stain. Label with a small label from the M27\_Hemo label printer. The slide will be retained with our body fluid smears.
5. Cover slip the wet prep smears and using the high dry lens, examine at least 25 fields on each smear for presence of sperm and motility.
6. If sperm are absent:

Refer to Reporting Results section for no sperm present.

1. If sperm are present:

**Count Sperm**:

1. Pipette 10 µL of the semen on **each** side of the hemocytometer. No bubbles are allowed on the hemocytometer. Clean and reload if there were bubbles.
2. Let it settle 3-5 minutes in a humidified place.
3. Count all 25 RBC squares on **each** side of the hemocytometer at 40x
4. Whether or not a sperm is counted is determined by the location of the sperm head on the hemocytometer grid. When counting, include sperm whose whole head is touching the top and left grid lines and exclude those that are on the bottom and right grid lines.
5. Count only intact sperm with a head and tail.
6. Begin by focusing on the hemocytometer lines. Then count only sperm that are in focus. **(Take note if you see any motile sperm at all.)**

**Calculating sperm concentration:**

To calculate sperm concentration, divide the **total** number of sperm counted on both sides by 200. This number equals the concentration in millions of sperm per milliliter.

Ex: Total number of sperm seen **on both sides** = 10

10/200 = 0.05 x 10^6/mL = 50,000

# REPORTING RESULTS

Report results through the Laboratory Information System.

1. If sperm are absent, result using:

0: No Sperm Seen (Azoospermia)

2. If sperm are present, result using the appropriate option based on number calculated and motility:

Calculated sperm less than or equal to 100,000 with no motile sperm seen:

1: < 100,000 non-motile sperm present

Calculated sperm less than or equal to 100,000 with **at least** 1 motile sperm seen:

2: < 100,000 motile sperm present

Calculated sperm greater than 100,000 with no motile sperm seen:

3: > 100,000 non-motile sperm present

Calculated sperm greater than 100,000 with **at least** 1 motile sperm seen:

4: > 100,000 motile sperm present

There is an interpretive message built for the test that says: “Testing follows The American Urological Association (AUA) Vasectomy Guidelines.”

**NORMALS**

The Normal Reference Range is:

No Sperm Seen (Azoospermia), below the level of detection.

# PROCEDURAL NOTES

The samples for Post Vasectomy examination do not have the urgency for delivery within

1 hour of collection, **however**; ideally the samples should be received within 4 hours.

Prolonged delays may affect the results. Some labs accept specimens only if stored at room temperature. **Samples that have been delayed in delivery or variant temperature storage prior to delivery cannot have any comment as to the motility of the sperm. State only the quantity present and a disclaimer about the motility.**

Make a comment on the patient's results and our worksheet of any delay in delivery or temperature variations.

# REFERENCES

1. Body Fluids, by Carl P. Kjeldsberg and Joseph A. Knight, Second edition, pp. 118-119.

## WHO. WHO Laboratory Manual for the Examination and processing of human semen, 5th ed., WHO 2010

1. CAP, 04.21.2014; HEM.35661