Urine Sample

Urine has a long, rich history as a source for measuring health and well-being and remains an important tool for clinical diagnosis. The clinical information obtained from a urine specimen is influenced by the collection method, timing and handling. . Determining which urine collection method and container should be used depends on the type of laboratory test ordered.

PRINCIPLE

Urine samples can be submitted for the detection of Schistosoma hematobium or for the presence of Trichomonas vaginalis.Adult Schistosomes take up residence near the bladder and start to release eggs. The eggs migrate to bladder and are released when the hos turinates

T. vaginallis lives in the vagina and can produce a frothy secretion which can be released during urination.

SPECIMEN

•For Schistosoma hematobium, obtain the last few drops of urine passed about or shortly after noon.

• For T. vaginalis, obtain the first portion of voided urine.

PROCEDURE

1) Spin urine sample at 500 g for 5 minutes.

2) Examine sediment under low powe r

parasites species in urine sample: helminths

- **Sch. haematobium.egg**
- **E. vermicularis. egg**
- **Sch. mansoni egg**
- **Micrfilaria (Ov, Wb)**
- **Hydatid sand**

protozoa:

- ***Tricomonas. Vaginalis*** **troph**

Types of Collection

Laboratory urine specimens are classified by the type of collection conducted or by the collection procedure used to obtain the specimen

Random Specimen This is the specimen most commonly sent to the laboratory for analysis, primarily because it is the easiest to obtain and is readily available. This specimen is usually submitted for urinalysis and microscopic analysis, although it is not the specimen of choice for either of these tests.

First Morning Specimen This is the specimen of choice for urinalysis and microscopic analysis, since the urine is generally more concentrated (due to the length of time the urine is allowed to remain in the bladder) and, therefore, contains relatively higher levels of cellular elements and analytes such as protein, if present.
Midstream Clean Catch Specimen This is the preferred type of specimen for culture and sensitivity testing because of the reduced incidence of cellular and microbial contamination.
Catheter Collection Specimen This assisted procedure is conducted when a patient is bedridden or cannot urinate independently
Pediatric Specimen For infants and small children, a special urine collection bag is adhered to the skin surrounding the urethral area

Urine Collection Containers (cups for collection and transport) Urine collection container cups come in a variety of shapes and sizes with lids that are either snap on or screw on. To protect healthcare personnel from exposure to the specimen and protect the specimen from exposure to contaminants, leak-resistant cups should be utilized
Urinalysis Tubes Urine specimens are poured directly into urinalysis tubes with screw- or snap-on caps. Additionally, there are evacuated tubes similar to those used in blood collection that are filled by using a straw device, from cups with integrated transfer devices built into their lid, or from direct sampling devices that are used to access catheter sampling ports. Conical bottom tubes provide the best sediment collection for microscopic analysis
Preservatives for Urinalysis most Guidelines recommend testing urine within two hours of its collection. However, refrigeration or chemical preservation of urine specimens may be utilized if testing or refrigeration within a two-hour window is not possible. A variety of urine preservatives (tartaric and boric acids being the most common) are available that allow urine to be kept at room temperature while still providing results comparable to those of refrigerated urine.
A terminal urine specimen (the last 10–20 ml passed) or alternatively a 24 hour collection of specimens of terminal urine is required for the diagnosis of Schistosoma haematobium infection.

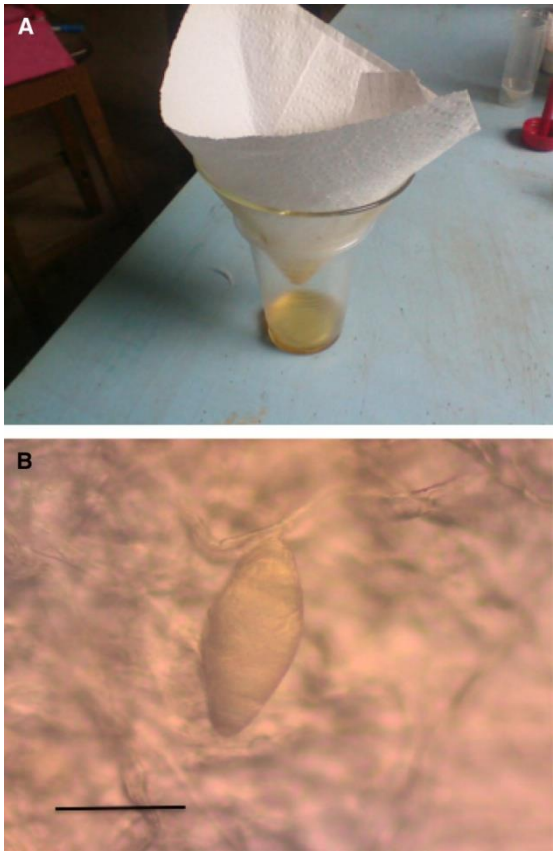
- Egg excretion is highest around midday. Exercise before specimen collection is not considered necessary if the urine is passed around that time, but is helpful, along with fluid intake before micturition, in increasing egg excretion at other times of day.
- If there is a delay in the examination of urine, it is recommended that 0.5 ml of 10% formalin is added to prevent the eggs of S haematobium from hatching.
- A minimum of 10 ml (10–20 ml) of terminal urine should be examined or Schistosomes (30 mL refrigerated urine)
- collect a midday random urine specimen or a 24 hour collection in a container without preservatives.

- Peak egg excretion occurs between noon and 3pm.
- For filariasis (30 mL refrigerated, random urine) collect avoided specimen in a clean container.
- For T. vaginalis in males, (30 mL room temperature, random urine), collect a first-voided urine (particularly after prostatic massage) in a clean container when urine is collected, the urine must be processed for transfer immediately following collection (centrifuge the urine for 10 minutes at 250x g; aspirate the supernatant and inoculate the sediment into a Trichosel broth using a sterile disposable pipette prior to transfer to the lab). If this process cannot be accommodated, the alternative method is a urethral swab or swab of prostatic secretions placed in Trichosel broth for Trichomonas culture.
  techniques used for concentration of eggs from urine involve either centrifugation or filtration by using specialized microporous membranes, which are relatively expensive for use in low-income countries.

FILTRATION

- **Gravity filtration device with single-ply paper towel as filter for urine**

Paper was rolled into a cone, fitted into a funnel, and placed over a glass. Ten milliliters of urine containing *S. haematobium* eggs was poured into the bottom of the rolled filter paper cone, which enabled gravity filtration of urine through the paper into a cup below. After filtration, we used scissors to cut off the bottom of the paper cone into which urine was poured. then placed this paper on a glass slide and examined it for ova by using conventional light microscopy under 10× and 40× objective lenses for identification of *S. haematobium* eggs. The presence or absence of eggs was recorded.



- SYRINGE FILTRATION

Blunt ended forceps were utilized to place a polycarbonate filter paper on the filter support of a filter holder. The filter holder was reassembled and attached to the end of a 10ml syringe, from which the plunger was removed. The syringe was then filled to the 10ml mark with a well mixed urine sample and the plunger replaced. By holding the syringe over a beaker, the urine was slowly passed through the filter. Then, the filter holder was removed and unscrewed. Again, using blunt ended forceps, the filter was carefully removed and transferred with the face upwards to a clean glass slide. A single drop of normal saline was added, and the mixture was then covered with a coverslip. Three slides from each urine sample were prepared by repeating the same procedure. Using a binocular microscope, the entire filter was examined systematically for the presence of S. haematobium eggs. The number of the eggs counted per 10ml of urine was recorded and the average of the three slides was calculated.

- VACUUM FLASK FILTRATION

A membranous filter paper was located in the appropriate place in the filtration apparatus and a vacuum pump was used to evacuate the air from the vacuum filtration flask. The urine sample was poured through the membranous filter paper and the

vacuum pump was switched on, when filtration had finished. Using metal forceps, the filter paper was removed from the holder and placed on a glass plate with the filtered eggs on the filter paper, facing away from the glass plate. Ninhydrine saturated solution was gradually poured drop wise to cover the whole filter paper and allowed to dry gradually under fluorescent light for three hours. The processed filter paper was then completely soaked with normal saline before being located on a glass slide and examined under a microscope, to visualize the darkened bilharzia eggs.

SEDIMENTATION

For each child investigated,10milliliters of urine was placed into a centrifugation tube and centrifuged at 2000 rpm for 3 minutes. Thereafter the supernatant was discarded and the deposit from each tube was placed as drops on three prepared slides and covered with coverslips, then examined using a binocular microscope.The eggs seen on each of the three slides prepared from a tube were counted and recorded as egg count /10ml of urine.

Eggs count

Urine samples were processed on the day of collection. A measured volume (maximum 10 ml) was centrifuged at 300 rpm for five minutes. The sediment was then examined under a light microscope. The eggs seen were counted and the intensity of infection per 10 ml of urine accordingly determined.

Egg hatching methods may occasionally be requested to determine viability or less commonly, to detect a limited infection

- Urine mixed in distilled water is placed in a flask, covered with foil (to keep out light), with neck or a sidearm exposed to bright light
- Miracidia, if present, actively swim to the light and can be detected using a hand lens

Antigen Detection :

for example: Schistosome antigens are present in serum and urine of infected subjects. According to their migratory behaviour in immunoelectrophoresis they are commonly referred to as circulating anodic antigens (CAA) and circulating cathodic antigens (CCA). These two circulating adult worm antigens are the basis of antigen

capture immunoassays. Measurement of CAA in the urine by ELISA-based assays is sensitive, specific and much less variable than egg counts.

Vaginal and Urethral Swabs

PRINCIPLE

Trichomonoas vaginalis can be detected in wet preparations of vaginal and urethral discharges. T. vaginalis has an undulating membrane and a characteristic jerky motility.

SPECIMENS

•swab from cervix,
•vaginal prostatic
•urethral secretions.

If examination is to be delayed, smears should be submitted to the laboratory for staining.

Specimen collection

Three vaginal fluid specimens were collected using sterile cotton swab from the posterior fornix. The first vaginal swab was placed in 10 ml screw-cap plastic tubes containing 0.5 ml of 0.9% saline to carry out the wet mount microscopy. Swab was vigorously rotated in the saline and pressed against the side of the tube to express as much fluid as possible. One drop of the expressed fluid was placed on glass slide with a cover slip and examined at magnification of 200× within 1 hour of collection of the sample. The positive result is defined as the presence of one or more trichomonads with characteristic morphology and jerky motility.

PROCEDURE

This is considered to be a non-routine procedure therefore it should only be performed by experienced personnel.

1) Dilute the sample with a drop of saline and place on a microscope slide.
2) Examine under low power and low intensity light for the presence of jerky motility.

QUALITY CONTROL

•Ensure that the microscope has been calibrated in the last year and that the results of the calibration are displayed on the microscope base.
•Haematoxylin staining can be used to confirm the diagnosis

Culture:

Trypticase-yeast extract-maltose (TYM) medium without agar (pH:6.0) supplemented with 10% heat inactivated bovine serum, penicillin (1000 IU/ml) and streptomycin sulphate (1 mg/ml) was used for the cultivation of the organism. Before vaginal swabs were placed into the medium, culture tubes were warmed to 37 oC or 15 min. Inoculated tubes were incubated for seven days at 37 oC, and examined daily under microscope. Presence of motile organisms was accepted as positive.