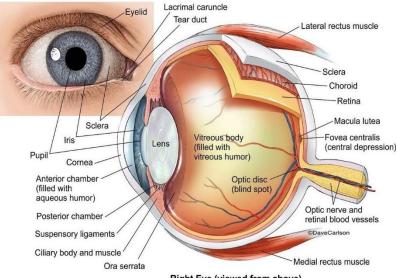
UPMC Vision Institute, Mercy Pavilion

Ophthalmic Microbiology Specimen Collection Manual

1.0 INTRODUCTION

Infections of the eye can be fulminant, leading to permanent loss of sight or the eye itself. Timely and properly collected specimens for culture of ocular samples can be helpful for patient treatment and management, especially from critical sites such as the cornea, anterior or posterior chambers, and vitreous or aqueous fluid. Infections and eye sources can be classified into three primary groups based on various anatomical sites, as the external eye, the cornea, and the internal eye. Collection methods of eye specimens for infectious microorganism detection is dependent upon the specific source and suspected infection.



Right Eye (viewed from above)

Eye specimens for microorganism detection may be submitted from any UPMC owned and affiliated eye care provider to the Campbell Ophthalmic Laboratory. The Campbell Ophthalmic Laboratory along with UPMC Clinical Laboratories can generally isolate, detect, and identify any microorganism that may infect the eye. In some cases, depending on the suspected microorganism and source, additional samples, different media types and collection methods may be required, or other appropriate specimens submitted to a reference laboratory for processing. Please consult with the listed clinical and laboratory consultants listed on the laboratory testing requisition form if you require additional assistance with specimen collection.

Specimens must always be handled and transported in a manner that ensures the safety of staff, patients, and visitors. Care must be taken to ensure that the integrity of each specimen has not been compromised. All specimens to be transported should be tightly capped in leak-proof containers when applicable, be sealed in plastic bags, and carried in an appropriately labeled biohazard transport bag. Biohazard labels must be affixed on the outside of the plastic bag that contain specimens if leaving the facility. Never carry specimens into any eating area.

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3.0 Specimen Collection for EXTERNAL Eye Culture

Primary Use Case: Suspected Conjunctivitis, Blepharitis.

These instructions are specifically tailored for proper specimen collection of EXTERNAL eye samplings via the ESwab® for aerobic, facultative & obligate anaerobic bacterial organisms and fungi. The specimen collection instructions herein are NOT appropriate for Chlamydia spp., Acanthamoeba spp., acid fast bacilli, virus detection/isolation.

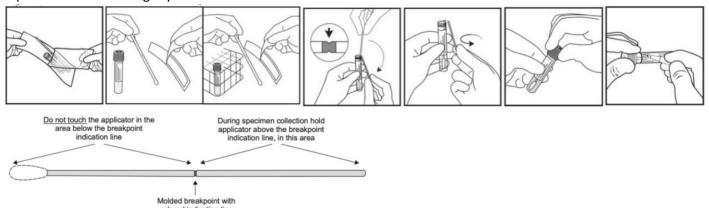
- **3.1** Acceptable external eye specimens a. Eyelid b. Lid margin c. Conjunctiva d. Lacrimal duct or gland/sac e. Canaliculus f. Pus or fluid when the diagnosis is pre-septal or septal cellulitis
- **3.2 SAMPLE SUBMISSION** Please register all patients and submit a properly filled out paper requisition with each specimen. Immediately after patient sampling, label all ESwab® collection tubes with the patient's name, date of birth, medical record number, specimen source (with indication of left or right eye), and collection date & time. Samples cannot be processed without these pertinent pieces of information. After collection, please submit your labeled ESwab® specimen with a paper requisition in a biohazard specimen collection bag to the Campbell Ophthalmic Laboratory (1622 Locust Street Room 2.431, Pittsburgh, Pa 15219).
- **3.3 SUPPLIES AND REAGENTS** 3.3.1 ESwab® collection kit (pictured to the right), 3.3.2 Sterile Physiological Saline

3.4 PROCEDURE

right eye in the source description.

NOTE: For the apparent externally infected eye, use one ESwab® kit per each external eye source. (i.e., if the patient requires a sampling of both the conjunctiva (suspected conjunctivitis) and eyelid (suspected blepharitis), please use one E-swab kit for the conjunctiva, and a separate unused Eswab kit for the eyelid), and submit separate requisitions for each. The uninfected eye may also be sampled with another unused E-swab kit and submitted for culture to help the provider rule in/out infection. Please indicate left or

3.4.1 Use of the ESwab® (pictured below) The operator must only handle the part of the swab applicator shaft above the breakpoint indication line as shown in Fig 3. After the swab sample is taken from the patient, the swab applicator shaft is broken off at the colored breakpoint indication line into the ESwab® tube of transport medium. The operator then discards the handle part of the swab into an approved medical waste disposal container. The tube's screw cap is then replaced and secured tightly.



- 3.4.2 Open the ESwab® sample collection pouch and remove the tube and swab
- 3.4.3 Conservatively premoisten the ESwab® with sterile physiological saline. Do NOT use the ESwab® liquid or broth (TSB) for pre-moistening or pre-wetting the applicator swab prior to collecting the sample or for rinsing or irrigating the sampling sites.

- 3.4.4 Aseptically collect the appropriate sample from the patient by rotating the entire surface of the sterile swab upon the suspected infected source area.
- 3.4.5 Unscrew and remove the cap from ESwab tube making sure not to spill the liquid.
- 3.4.6 Break the swab off into the tube as follows
 - 3.4.6.1 With the other hand grasp the swab shaft at the very end with the thumb and first finger and lift it up until the breakpoint mark is at the level of the rim of the tube.
 - 3.4.6.2 Lean the part of the shaft with the breaking point against the rim of the tube.
 - 3.4.6.3 Bend the swab shaft at a 180 degrees angle to break it off at the colored ink breakpoint mark. If needed, gently rotate the swab shaft to.
 - 3.4.6.4 Complete the breakage and take away the upper part of the swab shaft.
 - 3.4.6.5 Discard the broken handle part of the swab shaft into an approved medical waste disposal container.
- 3.4.7 Replace cap on the tube and secure tightly.
- 3.4.8 Confirm patient information on the tube label is correct.
- 3.4.9 Send your properly labeled ESwab® specimen with a properly filled out paper requisition form in a biohazard specimen collection bag to the Campbell Ophthalmic Laboratory (1622 Locust Street Room 2.431, Pittsburgh, Pa 15219). NOTE: Do not use excessive force, pressure or bending when collecting swab samples from patients as this may result in accidental breakage of the swab shaft. Swab shafts often exhibit diameter changes to facilitate different sampling requirements. Swab shafts may also have a molded breakpoint designed for intentional breakage of the swab into the transport tube. In all circumstances when collecting swabs samples from patients, do not use excessive force, pressure or bending of the swab as this may result in accidental breakage of the swab shaft.

3.5 Gram Staining/Slide Preparation

3.5.1 For external eye cultures, slide planting and gram stain procedures will be performed at the Campbell Ophthalmic Laboratory using the ESwab® Please submit your properly labeled ESwab® specimen with a paper requisition in a biohazard specimen collection bag to the Campbell Ophthalmic Laboratory (1622 Locust Street Room 2.431, Pittsburgh, Pa 15219).

3.6 Culture Plating

3.6.1 For external eye cultures, specimen plating will be performed at the Campbell Ophthalmic Laboratory using the ESwab®. Please submit your properly labeled ESwab® specimen a paper requisition in a biohazard specimen collection bag to the Campbell Ophthalmic Laboratory (1622 Locust Street Room 2.431, Pittsburgh, Pa 15219).



Bacterial/Fungal Culture

ESwab



VTM Swab

Viral/amoebic PCR



Aptima Multitest Swab

Chlamydia Testing

4.0 Specimen Collection for CORNEA Eye Culture

Primary Use Case: Bacterial & Fungal Keratitis

These instructions are specifically tailored for proper specimen collection of CORNEA eye samplings via spatula/blade or invasive surgical removal for aerobic, facultative anaerobic, and obligate anaerobic bacterial organisms. Swab only collection, using Eswab, will be accepted. The specimen collection instructions herein are NEITHER appropriate for Chlamydia spp., Acanthamoeba spp., NOR virus detection/isolation.

- 4.1 Acceptable corneal eye specimens a. Corneal Scrapings b. Corneal Rims c. Cornea
- **4.2 SAMPLE SUBMISSION** Please register all patients and submit a properly filled out paper requisition with each specimen. Prior to patient sampling, label all plates, slides, and tube specimens with the patient's name, date of birth, medical record number, specimen source, and collection date & time. Samples cannot be processed without these pertinent pieces of information. After collection, please submit your labeled specimens with a paper requisition in a biohazard specimen collection bag to the Campbell Ophthalmic Laboratory (1622 Locust Street Room 2.431, Pittsburgh, Pa 15219).
- **4.3 SUPPLIES AND REAGENTS** 4.3.1 Sterile Spatula/Blade 4.3.2 Sterile Forceps 4.3.3 Sterile Physiological Saline 4.3.4 Glass Microscope Slides w/frosted edge 4.3.5 TSA Blood Agar Plate (red plate) 4.3.6 Chocolate Agar Plate (brown plate) 4.3.7 Fungal Plate (Inhibitory Mold Agar) (white plate) 4.3.8 Thioglycollate Broth w/ Vitamin K and Hemin 4.3.9 Eswab (swab only collection; no plates or other media required)

4.4 PROCEDURE

NOTE: Inoculate each agar plate and thioglycollate broth present in the Corneal Specimen Collection Kit with corneal scrapings obtained with the spatula or blade. Inoculate donor cornea/corneal rims into thioglycollate broth only. Alternatively, you may submit donor cornea/corneal rim specimens that are in pink/red preservative directly to the Campbell Ophthalmic Laboratory for culture.

4.4.1 Inoculation of Media Corneal Scrapings

- 4.4.1.1 Label all plates and collection tubes with the patient's name, date of birth, medical record number, specimen source, and collection date & time
- 4.4.1.2 Inoculate agar plates with corneal scrapings retrieved via spatula/blade by streaking the spatula or blade flat edge containing the scrapings against the surface of the agar in a large "S" shape. **BE CAREFUL AS TO NOT CUT THE AGAR WITH THE SPATULA/BLADE.**
- 4.4.1.3 Each plate should include scrapings from at least 2 independently derived samplings of the cornea in a large streaked "S" shape inoculation.
- 4.4.1.4 Inoculate the thioglycollate broth with a fresh independent scraping of the cornea with a sterile spatula or blade by inserting it into the liquid medium. Make sure to touch the bottom of the tube with the spatula/blade, and subsequently stir the liquid. Alternatively, gently agitate the spatula/blade against the interior tube wall while completely submerged to the bottom of the tube to dislodge the scrapings from the spatula/blade.
- 4.4.1.5 Put the cap on the tube tightly and mix by inverting the tube at least once and returning it to upright position.
- 4.4.1.6 Please submit your properly labeled agar plates and tubes with a paper requisition in a biohazard specimen collection bag to the Campbell Ophthalmic Laboratory (1622 Locust Street Room 2.431, Pittsburgh, Pa 15219). NOTE: After collection, bacterial plates (Chocolate and TSA Blood Agar) should be placed in a 35°C

incubator, and Fungal plates (Inhibitory Mold Agar) should be placed in a 30°C incubator ASAP at the Campbell Ophthalmic Laboratory for lab processing. Thioglycollate tubes are placed in a 35°C incubator.

4.4.2 Gram Stain Slide Preparation

- 4.4.2.1 Slides should only be prepared after all culture media have been inoculated.
- 4.4.2.2 Label all slides with the patient's name, date of birth, and medical record number.
- 4.4.2.3 Using a sterile saline ampule, add a drop of saline to the slide.
- 4.4.2.4 Inoculate glass slides with corneal scrapings present on the spatula or blade into the drop of saline in a circular motion.
- 4.4.2.5 Allow the slide to completely dry, and then insert it into the protective slide holder.
- 4.4.2.6 Please submit your properly labeled slides with requisition (if submitting a paper requisition) in a biohazard specimen collection bag to the Campbell Ophthalmic Laboratory (1622 Locust Street Room 2.431, Pittsburgh, Pa 15219).

4.4.3 Swab Only Collection

Only Eswabs will be accepted for cornea swab sample collection.

- 4.4.3.1 Using only an Eswab, swab only the cornea or to retrieve scrapings. Do not inoculate any plates or broth with Eswab.
- 4.4.3.2 Refer to Section 6.0 of this manual for detailed directions on how to use Eswabs.



TSA Blood Agar Aerobic Bacterial Culture



Chocolate Agar
Aerobic Fastidious Bacterial Culture



Inhibitory Mold Agar Fungal Culture



Thioglycollate Broth w/Vitamin K and Hemin Anaerobic Bacterial Culture



Frosted Glass Slide w/holder for Microscopy

5.0 Inoculation of Media Donor Cornea Rim

- 5.1 NOTE: DO NOT INCOCULATE/PLANT ANY MEDIA PLATES WITH CORNEAL RIMS/DONOR CORNEAS. You may directly submit donor cornea/corneal rim specimens that are in pink/red preservative (Optisol or equivalent).
 - 5.1.1 Label the thioglycollate tube with the patient's name, date of birth, medical record number, specimen source, and collection date & time.
 - 5.1.2 Loosen the cap of the broth tube.
 - 5.1.3 Using sterile forceps, aseptically grasp the corneal rim/donor cornea at the outer most edge.
 - 5.1.4 Subsequently, aseptically remove the cap of the broth tube.
 - 5.1.5 Submerge the donor cornea into the broth.
 - 5.1.6 Recap the thioglycollate tube tightly.
 - 5.1.7 Please submit your properly labeled tube specimen with a properly filled out paper requisition in a biohazard specimen collection bag to the Campbell Ophthalmic Laboratory. After collection, thioglycollate broths should be placed in a 35° C incubator when specimen storage is required before submission to the Campbell Ophthalmic Laboratory (1622 Locust Street Room 2.431, Pittsburgh, Pa 15219).

6.0 Specimen Collection for FOREIGN BODY Eye Culture

Primary Use Case: Screening for presence of pathogenic microbes

These instructions are specifically tailored for proper specimen collection of FOREIGN BODY eye samplings for aerobic, facultative anaerobic, and obligate anaerobic bacterial organisms. The specimen collection instructions herein are **NEITHER** appropriate for Chlamydia spp., Acanthamoeba spp., NOR virus detection/isolation. Section 7.0 lists the acceptable types of internal eye specimens. Standard contact lenses and contact lens & eye-glass cases are NOT acceptable.

- **6.1** Acceptable foreign body specimens a. Prosthetic lens (Kontur Kontact Lens) b. Other Eye Prosthetics and Lenses
- **6.2 SAMPLE SUBMISSION** Please register all patients and submit a paper requisition with each specimen. Prior to patient sampling, label all plates, slides, and tube specimens with the patient's name, date of birth, medical record number, specimen source, and collection date & time. Samples cannot be processed without these pertinent pieces of information. After collection, please submit your labeled specimens with a properly filled out paper requisition in a biohazard specimen collection bag to the Campbell Ophthalmic Laboratory (1622 Locust Street Room 2.431, Pittsburgh, Pa 15219).
- 6.3 SUPPLIES AND REAGENTS 6.3.1 Sterile Forceps 6.3.2 Thioglycollate Broth w/ Vitamin K and Hemin

6.4 PROCEDURE

NOTE: DO NOT INOCULATE/PLANT ANY MEDIA PLATES WITH FOREIGN BODY SPECIMEN

6.4.1 Inoculation of Foreign Body

- 6.4.1.1 Label the thioglycollate tube with the patient's name, date of birth, medical record number, specimen source, and collection date & time.
- 6.4.1.2 Loosen the cap of the broth tube.
- 6.4.1.3 Using sterile forceps, aseptically grasp the foreign body at the outer most edge.

- 6.4.1.4 Subsequently, aseptically remove the cap of the broth tube.
- 6.4.1.5 Submerge the foreign body into the broth.
- 6.4.1.6 Recap the thioglycollate tube tightly.
- 6.4.1.7 Please submit your properly labeled tube specimen with a properly filled out paper requisition in a biohazard specimen collection bag to the Campbell Ophthalmic Laboratory (1622 Locust Street Room 2.431, Pittsburgh, Pa 15219).



Thioglycollate Broth w/Vitamin K and Hemin Anaerobic & Aerobic Bacterial Culture

7.0 Specimen Collection for Internal Eye Culture

Primary Use Case: Endophthalmitis

These instructions are specifically tailored for proper specimen collection of INTERNAL eye samplings via needle aspiration or invasive surgical removal for aerobic, facultative anaerobic, and obligate anaerobic bacterial and fungal organisms. The specimen collection instructions herein are NOT appropriate for Chlamydia spp., or Acanthamoeba.

- **7.1** Acceptable sterile inner eye specimens a. Anterior chamber (Tissue) b. Posterior chamber/Other Internal Eye (Tissue) e. Iris (Tissue) f. Uvea (Tissue) g. Lens (Tissue) h. Choroid (Tissue) i. Retina (Tissue) j. Orbit (Tissue/Swab of) k. Aqueous humor l. Vitreous humor m. Pus or fluid when the diagnosis is orbital cellulitis
- **7.2 SAMPLE SUBMISSION** Please register all patients and submit a paper requisition with each specimen. Prior to patient sampling, label all plates, slides, and tube specimens with the patient's name, date of birth, medical record number, specimen source, and collection date & time. Samples cannot be processed without these pertinent pieces of information. After collection, please submit your labeled specimens with a paper requisition in a biohazard specimen collection bag to the Campbell Ophthalmic Laboratory (1622 Locust Street Room 2.431, Pittsburgh, Pa 15219).

7.3 SUPPLIES AND REAGENTS 7.3.1 Syringe with Luer-Lock (Needle removed and capped before sample submission) 7.3.2 Sterile Forceps 7.3.3 Sterile Physiological Saline 7.3.4 Sterile Container 7.3.5 Glass Microscope Slides w/frosted edge 7.3.6 TSA Blood Agar Plate (red plate) 7.3.7 Chocolate Agar Plate (brown plate) 7.3.8 Anaerobic BBA Plate w/gas pack (red plate) 7.3.9 Fungal Plate (Inhibitory Mold Agar) (white plate).

7.4 PROCEDURE

7.4.1 Inoculation of Eye tissue

- 7.4.1.1 For tissue specimen eye culture, specimen plating will be performed at the Campbell Ophthalmic Laboratory. Using sterile forceps, insert tissue into a sterile container and moisten with 2-3 drops of sterile saline.
- 7.4.1.2 Please submit your properly labeled specimen in a sterile container with a paper requisition in a biohazard specimen collection bag to the Campbell Ophthalmic Laboratory (1622 Locust Street Room 2.431, Pittsburgh, Pa 15219).
- 7.4.1.3 For direct swabs of the orbit, refer to the external eye collection instructions for using Eswabs. Aspirate of the orbit is the preferred specimen type; please collect via needle aspirate whenever possible.
- 7.4.1.4 For swabs of fluid or pus, refer to the external eye collection instructions using Eswabs.

7.4.2 Inoculation of Aqueous & Vitreous humor

- 7.4.2.1 Label all plates with the patient's name, date of birth, and medical record number
- 7.4.2.2 For syringe or surgically obtained aqueous and vitreous humor, inoculate the center of all plates with 1 drop of fluid while keeping plate horizontally leveled. If virological studies are required on the remaining syringe sample, engage the Luer lock, remove needle, cap the syringe.
- 7.4.2.3 Cover agar plate with lid, keep plate upright, horizontally leveled, and do disturb plate. Allow to slightly dry before transport.
- 7.4.2.4 Anaerobic plates should be placed in the provided anaerobe bag and sealed after slight drying, before transport.
- 7.4.2.5 Alternatively, you may submit aqueous or vitreous humor in a Luer-locked syringe, capped without the needle, directly to the Campbell Ophthalmic Laboratory (1622 Locust Street Room 2.431, Pittsburgh, Pa 15219). All plating and gram staining can be performed in the laboratory. INDICATE PRIORITY OF REQUESTED TESTING TYPE BASED UPON YOUR DIAGNOSTIC DIFFERENTIAL ON THE REQUISTION FOR QNS SPECIMENS WITH MULTIPLE ORDERS.
- 7.4.2.6 Please submit your properly labeled specimen in a sterile container and/or syringe with a properly filled out paper requisition in a biohazard specimen collection bag to the Campbell Ophthalmic Laboratory (1622 Locust Street Room 2.431, Pittsburgh, Pa 15219). After collection, bacterial plates (TSA Blood Agar, Chocolate Agar, and Anaerobic Agar media) should be placed in a 35°C incubator, and Fungal plates (Inhibitory Mold Agar) should be placed in a 30°C incubator when specimen storage is required before submission to the Campbell Ophthalmic Laboratory.

7.4.3 Gram Stain Slide Preparation

7.4.3.1 For tissues and ESwabs, slide planting and gram stain procedures will be performed at the Campbell Ophthalmic Laboratory. Please submit your properly labeled ESwab and/or tissue in sterile container with a paper requisition in a biohazard specimen collection bag to the Campbell Ophthalmic Laboratory (1622 Locust Street Room 2.431, Pittsburgh, Pa 15219).

- 7.4.3.2 Due to the low sensitivity of Gram stain, if volume is limited, inoculation of culture media should be prioritized over slides. Only inoculate slides after all culture media have been inoculated.
- 7.4.3.3 For aqueous and vitreous humor, label all slides with the patient's name, date of birth, and medical record number.
- 7.4.3.4 Inoculate the center of the glass slide with 1 drop of humor, while keeping plate horizontally leveled. Do not disturb the slide after inoculation.
- 7.4.3.5 Allow the slide to completely dry, and then insert it into the protective slide holder.
- 7.4.3.6 Alternatively, you may submit aqueous or vitreous humor in a Luer-locked syringe, without the needle, directly to the Campbell Ophthalmic Laboratory. All plating and gram staining can be performed in the laboratory.
- 7.4.3.7 Please submit your properly labeled slides with a paper requisition in a biohazard specimen collection bag to the Campbell Ophthalmic Laboratory (1622 Locust Street Room 2.431, Pittsburgh, Pa 15219).



TSA Blood Agar Aerobic Bacterial Culture



Chocolate Agar
Aerobic Fastidious Bacterial Culture



Inhibitory Mold Agar Fungal Culture



Anaerobic EZ GasPak



Frosted Glass Slide w/holder for Microscopy

8.0 Specimen Collection for Acanthamoeba spp. & Virus Detection by PCR

Primary Use Case: Ocular Viral Infections, Viral Keratitis, Viral Conjunctivitis, Amoebic Keratitis

These instructions are specifically tailored for proper specimen collection of eye samplings for Acanthamoeba spp. and Virus detection by PCR. The specimen collection instructions herein are NOT appropriate for culture of microorganisms. DO NOT use ESwabs, Aptima Multitest Swabs, or Bartels for Acanthamoeba spp. and Virus Detection by PCR.

- **8.1** Acceptable external & internal eye, and corneal specimens a. Cornea (swabs only) b. Eyelid & lid margin c. Conjunctiva d. Canaliculus e. Vitreous and anterior chamber fluid (syringes only) f. Lacrimal duct or gland/sac g. Pus or fluid
- **8.2 SAMPLE SUBMISSION** Please register all patients and submit a properly filled out paper requisition with each specimen. Prior to patient sampling, label all collection tube specimens with the patient's name, date of birth, medical record number, specimen source, and collection date & time. Samples cannot be readily processed and results released without these pertinent pieces of information. After collection, please submit your labeled specimens with requisition (if submitting a paper requisition) in a biohazard specimen collection bag to the Campbell Ophthalmic Laboratory (1622 Locust Street Room 2.431, Pittsburgh, Pa 15219).
- **8.3 SUPPLIES AND REAGENTS** 8.3.1 Universal or Viral Transport Media (UVT/VTM) w/swab (pictured to the right) 8.3.2 Sterile Physiological Saline 8.3.3 Syringe with Luer Lock (Needle removed and capped before sample submission)

8.4 PROCEDURE

- 8.5 Detection of Viruses and Acanthamoeba spp. in eye specimens NOTE: For swabs, use only UVT/VTM collection kit as pictured to the right.
 - 8.5.1 Open the UVT/VTM collection pouch and aseptically remove the tube and swab.
 - 8.5.2 Conservatively premoisten the UVT/VTM swab with sterile physiological saline. Do NOT use the UVT/VTM medium or Trypticase Soy Broth (TSB) for premoistening or pre-wetting the applicator swab prior to collecting the sample or for rinsing or irrigating the sampling sites.
 - 8.5.3 Sterilely collect the appropriate sample from the patient by rotating the entire surface of the swab upon the suspected infected source area.
 - 8.5.4 Unscrew and remove the cap from the collection tube making sure not to spill the medium
 - 8.5.5 Break the swab off into the collection tube at the marked/scored region of the swab shaft.
 - 8.5.6 Replace cap on the tube and secure tightly.
 - 8.5.7 For vitreous and anterior/aqueous chamber fluid collected via syringe: after collection, engage the Luer lock, remove needle, and cap the syringe.
 - 8.5.8 Write patient information on the tube label or apply patient identification label to the collection tube or syringe.
 - 8.5.9 Send your properly labeled collection specimen with a paper requisition in a biohazard specimen collection bag to the Campbell Ophthalmic Laboratory (1622 Locust Street Room 2.431, Pittsburgh, Pa 15219).

9.0 Specimen Collection for Chlamydia spp. Detection by NAAT

Primary Use Case: Chlamydial Conjunctivitis, Trachoma

These instructions are specifically tailored for proper specimen collection of eye samplings for Chlamydia spp. by NAAT. The specimen collection instructions herein are NOT appropriate for culture of microorganisms. DO NOT use ESwabs, UVT/VTM, or Bartels for Chlamydia spp. testing.

- **9.1 Acceptable external eye specimens** a. Cornea (swabs only) b. Conjunctiva c. Canaliculus d. Lacrimal duct or gland/sac e. Pus or fluid
- **9.2 SAMPLE SUBMISSION** Please register all patients and submit a paper requisition with each specimen. Prior to patient sampling, label all collection tube specimens with the patient's name, date of birth, medical record number, specimen source, and collection date & time. Samples cannot be readily processed and results released without these pertinent pieces of information. After collection, please submit your labeled specimens with a properly filled out paper requisition in a biohazard specimen collection bag to the Campbell Ophthalmic Laboratory (1622 Locust Street Room 2.431, Pittsburgh, Pa 15219).
- **9.3 SUPPLIES AND REAGENTS** 9.3.1 Aptima Collection Tube w/swab (Figure 6) 9.3.2 Sterile Physiological Saline

9.4 PROCEDURE

9.4.1 Detection of Chlamydia spp. in eye specimens NOTE: Use only the APTIMA Multitest collection kit as pictured to the right.



- 9.4.1.1 Open the APTIMA collection pouch and aseptically remove the tube and swab. Do not puncture, poke, or touch the aluminum foil on the cap.
- 9.4.1.2 Conservatively premoisten the APTIMA swab with sterile physiological saline. Do NOT use the APTIMA liquid or broth (TSB) for pre-moistening or prewetting the applicator swab prior to collecting the sample or for rinsing or irrigating the sampling sites.
- 9.4.1.3 Sterilely collect the appropriate sample from the patient by rotating the entire surface of the swab upon the suspected infected source area.
- 9.4.1.4 Unscrew and remove the cap from the collection tube making sure not to spill the medium. Do not puncture, poke, or touch the aluminum foil on the cap.
- 9.4.1.5 Break the swab off into the collection tube at the marked/scored region of the swab shaft.
- 9.4.1.6 Replace cap on the tube and secure tightly.
- 9.4.1.7 Write patient information on the tube label or apply patient identification label.
- 9.4.1.8 Send your properly labeled collection specimen with a paper requisition in a biohazard specimen collection bag to the Campbell Ophthalmic Laboratory (1622 Locust Street Room 2.431, Pittsburgh, Pa 15219).

10. References

- 10.1 Practical Guidance for Clinical Microbiology Laboratories: Diagnosis of Ocular Infections. Clinical microbiology reviews July 2021 volume 34 issue 3.
- 10.2 Clinical Microbiology Procedures Handbook 4th edition 3.10 Ocular cultures
- 10.3 COPAN ESwab® package insert and how to use guide; HPC108 Rev.002017.07