# NEW YORK STATE

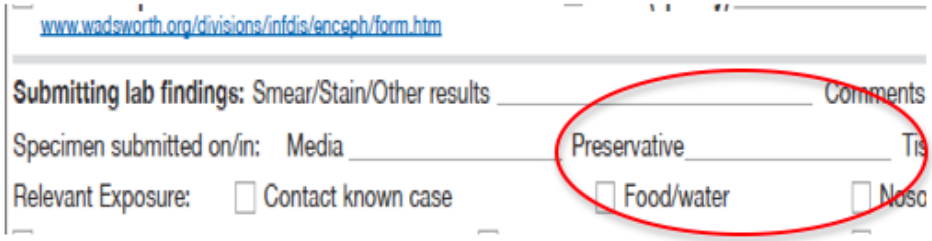
*Parasitology Proficiency Testing Program*

## News and Notes

Beginning with the May 2013 event a separate set of 3 samples were supplied for laboratories performing antigen detection. For this event those were samples 14I-F, 14I-G, and 14I-H. These are distinct from samples 14-F, 14-G, and 14-H and could not be used interchangeably.

As molecular methods become increasingly common in the clinical parasitology lab, so does the necessity of knowing what preservative was used with the specimen. Preservatives commonly used for parasitology are not ideal for DNA extraction, and newly developed tests may only be approved for use with specific preservatives. For example, the assay may be approved for use with specimens preserved in 10% formalin but not SAF. Please remember to include the preservative information by filling out that section of the Infectious Disease Requisition, whenever submitting specimens to Wadsworth.

Please note that an unpreserved specimen should also be submitted, whenever possible, to maximize the likelihood of extracting good quality DNA.




## Parasitology Comprehensive
## 20 May 2014

The purpose of the New York State Proficiency Testing Program in the category of Parasitology - Comprehensive is to monitor the performance of applicant laboratories that detect and identify parasites in fecal emulsions, fecal smears, and blood films. Below please find the results for the May 2014 proficiency test in Parasitology - Comprehensive and Antigen Detection.

## Sample Preparation and Quality Control

All emulsions and slides used in this test were prepared by a commercial source. The emulsions were dispensed into the vials from pools, which were continuously mixed during the loading process. Numerous samples of each test specimen were selected at random by the Wadsworth Center Parasitology Laboratory (NYSDOH), and were assayed for quality and confirmation of organisms. The supplying vendor also conducted extensive quality control tests and a detailed quality control report was submitted for inspection and verification. Samples were authenticated by at least 80% of participating laboratories and/or referee laboratories.

# 14-F (All Parasites)

Correct Identification: *Cryptosporidium* species

## *Results of Participating Laboratories*

| Organism reported | # of labs reporting | % of labs reporting | Referee results | Status |
|---|---|---|---|---|
| *Cryptosporidium* species | 86/93 | 92 | 10/10 | **Correct** |
| *Endolimax nana* | 1 | 1 | 0 | **Incorrect** |
| No Parasites Seen | 6 | 6 | 0 | **Incorrect** |

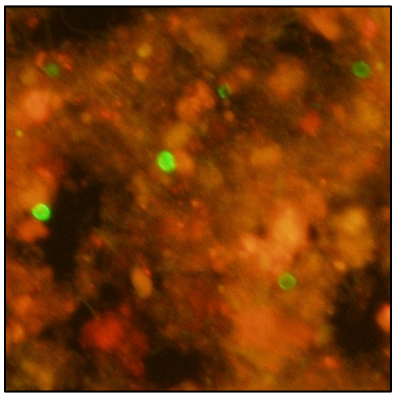
## *Quality Control and Referee Information*

Participating and referee laboratories agreed that ***Cryptosporidium* sp.** was the correct response (92 and 100% respectively). Quality control examination of 4% of the specimens showed oocysts in every 40X field of a modified acid-fast stained slide. Other tests performed included a direct immunofluorescent assay, which was positive for *Cryptosporidium* sp. and negative for *Giardia lamblia*.

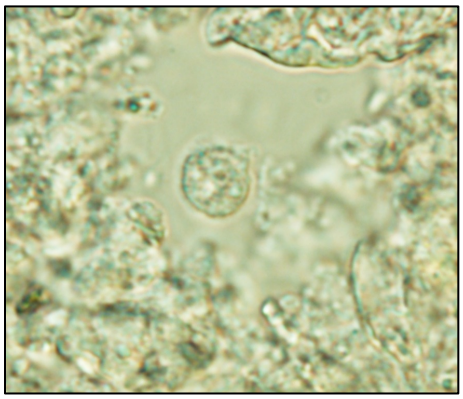
**Please Note:** As described in the instructions available on the website, you should include a test for *Cryptosporidium* sp. whenever required by the type of parasite requested. Thus, when "report all parasites" is indicated for a fecal emulsion, a test for *Cryptosporidium* should be performed, unless testing for this parasite is not offered in your laboratory.

## *Diagnostic Characteristics*

***Cryptosporidium*** is typically spread through contaminated water. The diagnostic stage is the oocyst, which is passed in the stool and is immediately infective. The combination of low infective dose, multiplication in the infected hosts, which leads to high number of shed oocysts, and hardiness of the oocysts, make this a common intestinal parasite. On a modified acid-fast stained smear the oocyst measure 4-5 μm and range from unstained to pale or deep pink. Other methods of diagnosis include immunoassays and immunofluorescent stains. Rapid card tests, or lateral flow devices, are also available and have been used with varying degrees of success.



DFA



Wet Mount

# 14-G (All Parasites)

Correct Identification: *Necator americanus / Ancylostoma duodenale*

## *Results of Participating Laboratories*

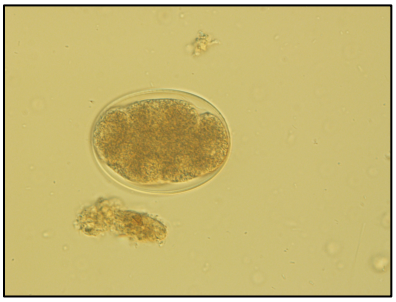
| Organism reported | # of labs reporting | % of labs reporting | Referee results | Status |
|---|---|---|---|---|
| *Necator americanus / Ancylostoma duodenale* (Hookworm) | 92/93 | 99 | 10/10 | **Correct** |
| *Entamoeba coli* | 18 | 19 | 3 | **No Penalty** |
| *Blastocystis hominis* | 7 | 8 | 1 | **No Penalty** |
| *Balantidium coli* | 1 | 1 | 0 | **Incorrect** |
| *Schistosoma mansoni* | 1 | 1 | 0 | **Incorrect** |

## *Quality Control and Referee Information*

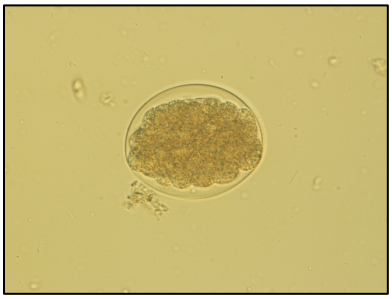
Participating and referee laboratories agreed that **hookworm** was the correct response (99 and 100% respectively. Quality control examination of 4% of the vials for this sample showed at least 20 eggs per coverslip. Eggs were observed at various stages of development.

## *Diagnostic Characteristics*

***Necator americanus/Ancylostoma duodenale* (Hookworm)** infection occurs via skin penetration of filariform larvae found in contaminated soil. The larvae travel via the blood to the heart and lungs. They migrate to the trachea and pharynx and are swallowed. Larvae pass through the stomach and take up residence in the small intestine where they mature into adults. The diagnostic stage is the egg passed in stool. Eggs are oval, measure approximately 60 by 40 µm and have a space between the thin hyaline shell and the developing embryo. Eggs are typically observed at the 8 to 32-cell stage of development. *Necator americanus* and *Ancylostoma duodenale* cannot be distinguished from each other based on the morphology of the eggs.



A



B

# 14-H (All Parasites)

Correct Identification: *Balantidium coli*

## *Results of Participating Laboratories*

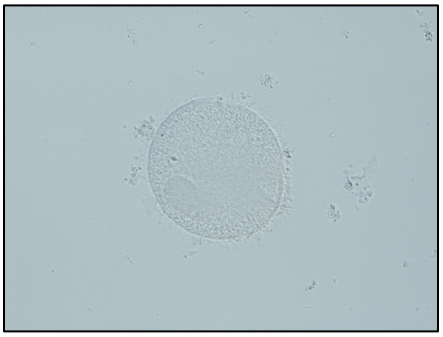
| Organism reported | # of labs reporting | % of labs reporting | Referee results | Status |
|---|---|---|---|---|
| *Balantidium coli* | 89/93 | 96 | 10/10 | **Correct** |
| *Necator americanus / Ancylostoma duodenale* (Hookworm) | 1 | 1 | 0 | **Incorrect** |
| *Schistosoma japonicum* | 1 | 1 | 0 | **Incorrect** |
| No Parasites Seen | 2 | 2 | 0 | **Incorrect** |

## *Quality Control and Referee Information*

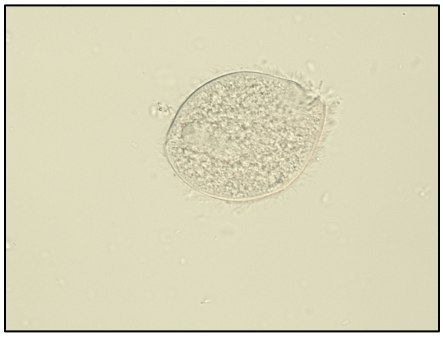
Participating and referee laboratories agreed that ***Balantidium coli*** was the correct response (96% and 100% respectively). Quality control examination of 4% of the vials for this sample showed >20 organisms per coverslip. Both cysts and trophozoites were observed.

## *Diagnostic Characteristics*

***Balantidium coli*** is usually found in warm climates but can also occur in cooler climates. Pig farmers and people working in slaughterhouses are at increased risk for infection with this parasite. It is the only pathogenic ciliate to infect humans and the largest of all protozoa. The trophozoites measures 50-100 µm by 20-50 µm and are covered with cilia. Both trophozoites and cysts have two nuclei of unequal size. More prominent is the large bean-shaped macronucleus. The anterior end of the trophozoite is slightly pointed and in some cases the cytostome is visible. Cysts measure 50-70 µm and also have a macro- and micronucleus.



Cyst



Trophozoite

# 14-I (All Parasites)

Correct Identification: *Giardia lamblia*

## *Results of Participating Laboratories*

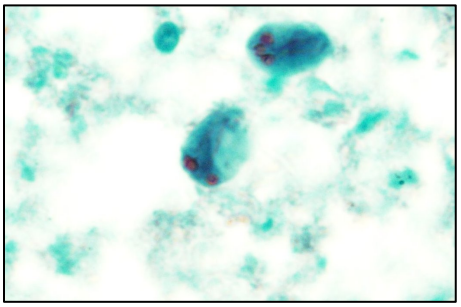
| Organism reported | # of labs reporting | % of labs reporting | Referee results | Status |
|---|---|---|---|---|
| *Giardia lamblia* | 93/93 | 100 | 10/10 | **Correct** |

## *Quality Control and Referee Information*

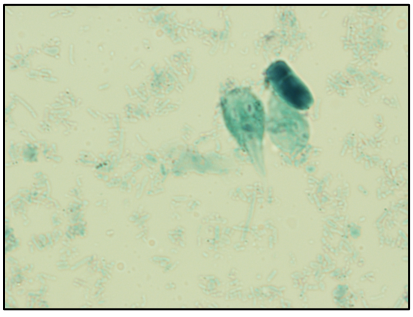
All participating and referee laboratories agreed that ***Giardia lamblia*** was the correct response. Quality control examination of 4% of the slides for this sample showed multiple organisms in every 40X oil immersion field. Both trophozoites and cysts were observed, with cysts being more prevalent.

## *Diagnostic Characteristics*

***Giardia lamblia*** is the most commonly diagnosed flagellate in humans. It has a worldwide distribution and is more prevalent in children than in adults. The infective cysts are oval and measure 11-15 μm. Trophozoites are pear shaped and measure 10-20 μm.



Cysts



Trophozoites

# 14-J (All Parasites)

Correct Identification: *Loa loa*

## *Results of Participating Laboratories*

| Organism reported | # of labs reporting | % of labs reporting | Referee results | Status |
|---|---|---|---|---|
| *Loa loa* | 52/88 | 59 | 6/10 | **Correct*** |
| *Mansonella* sp. | 29 | 33 | 3 | **Correct*** |
| *Babesia* sp. | 1 | 1 | 0 | **Incorrect** |
| No Parasites Seen | 6 | 7 | 1 | **Incorrect** |

* Credit was given for both *Loa loa* and *Mansonella* sp. as this sample was authenticated only as microfilaria.

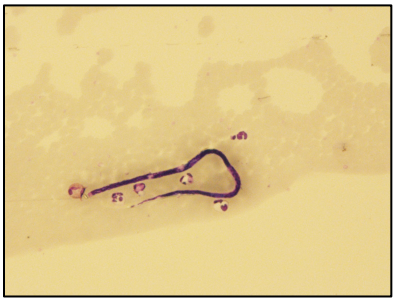
## *Quality Control and Referee Information*

This sample contained microfilariae of *Loa Loa*, the African eye worm. Because participating and referee laboratories were split between identification as *Mansonella* and *Loa loa* the specimen was authenticated as **microfilaria** (92% and 90% respectively). Quality control examination of 4% of the slides for this sample showed an average of 3 organisms per coverslip.

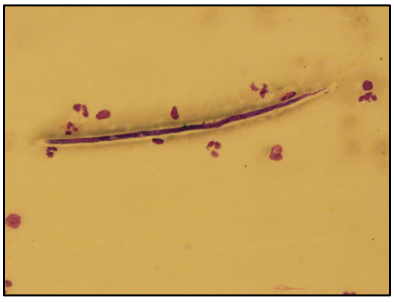
## *Diagnostic Characteristics*

**Microfilariae** of ***Loa loa,*** are sheathed, measure between 250-300 μm, and have nuclei that are irregularly spaced but extend to the tip of the tail. However, the sheath of *Loa loa* does not stain well with Giemsa and the microfilaria can lose their sheath. This situation is likely to have lead to misidentification as *Mansonella*. *Mansonella perstans* is also found in the blood and has nuclei that extend to the tip of the tail, though they have no sheath and the nuclei are more regularly spaced.

Both *Loa loa* and *Mansonella perstans* are found in Africa, though *M. perstans* is also found in South America. *Loa loa* is transmitted by deer or mango flies while *Mansonella* species are transmitted by midges. In both cases the larvae are deposited into the bite wound. *Loa loa* is most likely to be found in peripheral blood between 10 AM and 2 PM, and is the only filarid known to have diurnal periodicity.



**A**



**B**

A separate set of samples (14I-F, 14I-G, and 14I-H) was sent for antigen detection. These results are reported below and show that **100% of labs obtained the correct answers for all three specimens.**

# Scoring Information

## *Immunoassay Results*

| ***Cryptosporidium*** | **14I-F** | | **14I-G** | | **14I-H** | |
|---|---|---|---|---|---|---|
| **METHOD** | **-** | **+** | **-** | **+** | **-** | **+** |
| MCC Para-Tect Cryptosporidium/Giardia DFA | 1 | 0 | 0 | 1 | 1 | 0 |
| Meridian ImmunoCard STAT Cryptosporidium/Giardia | 23 | 0 | 0 | 23 | 23 | 0 |
| Meridian Merifluor Cryptosporidium/Giardia | 16 | 0 | 0 | 16 | 16 | 0 |
| Meridian Premier Cryptosporidium | 2 | 0 | 0 | 2 | 2 | 0 |
| Remel ProSpecT Cryptosporidium EIA | 12 | 0 | 0 | 12 | 12 | 0 |
| Remel Xpect Giardia/Cryptosporidium | 6 | 0 | 0 | 6 | 6 | 0 |
| TechLab Cryptosporidium II ELISA | 2 | 0 | 0 | 2 | 2 | 0 |
| TechLab Giardia/Cryptosporidium Quik Chek (Alere) | 7 | 0 | 0 | 7 | 7 | 0 |
| TechLab/Wampole Test EIA | 4 | 0 | 0 | 4 | 4 | 0 |

| ***Giardia*** | **14I-F** | | **14I-G** | | **14I-H** | |
|---|---|---|---|---|---|---|
| **METHOD** | **-** | **+** | **-** | **+** | **-** | **+** |
| MCC Para-Tect Cryptosporidium/Giardia DFA | 0 | 1 | 0 | 1 | 1 | 0 |
| Meridian ImmunoCard STAT Cryptosporidium/Giardia | 0 | 23 | 0 | 23 | 23 | 0 |
| Meridian Merifluor Cryptosporidium/Giardia | 0 | 13 | 0 | 13 | 13 | 0 |
| Meridian Premier Giardia | 0 | 1 | 0 | 1 | 1 | 0 |
| Remel ProSpecT Giardia EIA | 0 | 19 | 0 | 19 | 19 | 0 |
| Remel ProSpecT Giardia EZ | 0 | 2 | 0 | 2 | 2 | 0 |
| Remel Xpect Giardia | 0 | 2 | 0 | 2 | 2 | 0 |
| Remel Xpect Giardia/Cryptosporidium | 0 | 6 | 0 | 6 | 6 | 0 |
| TechLab Giardia II ELISA | 0 | 2 | 0 | 2 | 2 | 0 |
| TechLab Giardia/Cryptosporidium Quik Chek (Alere) | 0 | 7 | 0 | 7 | 7 | 0 |
| TechLab/Wampole Test EIA | 0 | 6 | 0 | 6 | 6 | 0 |

## *Distribution of Scores  Parasitology - Comprehensive*

| Score | # of labs | % of labs |
|---|---|---|
| 100 | 76 | 80 |
| 90-99 | 2 | 2 |
| 80-89 | 13 | 14 |
| 70-79 | 1 | 1 |
| 60-69 | 2 | 2 |
| 0-59 | 1 | 1 |

## *Distribution of Scores  Parasitology - Antigen Detection*

| Score | # of labs | % of labs |
|---|---|---|
| 100 | 82 | 100 |
| 90-99 | 0 | 0 |
| 80-89 | 0 | 0 |
| 70-79 | 0 | 0 |
| 60-69 | 0 | 0 |
| 0-59 | 0 | 0 |

## *Answer Key  Parasitology - Comprehensive*

| Sample | Correct Answer |
|---|---|
| **14-F** | *Cryptosporidium* sp. |
| **14-G** | *Necator americanus / Ancylostoma duodenale* (Hookworm) |
| **14-H** | *Balantidium coli* |
| **14-I** | *Giardia lamblia* |
| **14-J** | *Loa loa* * |

* Sample contained *Loa loa* but was authenticated only as microfilaria.

## *Answer Key  Parasitology - Antigen Detection*

| Sample | Correct Answer |
|---|---|
| **14I-F** | *Giardia lamblia* |
| **14I-G** | *Cryptosporidium* sp. and *Giardia lamblia* |
| **14I-H** | Negative |

## *Grading*

The answer key was derived from the response of all participating laboratories as per **CLIA Regulations**, CFR Title 42, Part 493, Subpart I, Section 493.917. These regulations can be viewed at www.cdc.gov/clia/Regulatory/default.aspx. These regulations state that 80% or more of participating laboratories **or** referee laboratories must identify the parasite for it to be authenticated as a correct answer. Similarly, reporting of a parasite identified by less than 10% of the participating laboratories **or** referees is an incorrect response. Organisms that are not authenticated, but which were reported by more than 10% of the participating laboratories **or** referees, are "Unauthenticated" and are not considered for grading.

Credit is given according to the formula:

[# of Correct Responses / (# of Correct Responses + # of Incorrect Responses)] X 100

For example, if a sample contained one principal parasite and the laboratory reported it correctly but reported the presence of an additional parasite, which was not present, the sample grade would be:

$1/(1+1)\times 100 = 50$ percent.

# Important Reminders

The next Parasitology Proficiency Test is scheduled for **September 30, 2014.** Participating labs will need to notify us **before October 7, 2014** if the samples are not received. Proficiency test results must be electronically submitted through EPTRS by **October 15, 2014** or the laboratory will receive a score of zero. This and additional information can be found in the NYS Proficiency Testing Program Guide provided by the NYS Clinical Laboratory Evaluation Program, which can be accessed via the Internet at:

http://www.wadsworth.org/labcert/clep/ProgramGuide/pg.htm