

NEW YORK STATE

Parasitology Proficiency Testing Program

News and Notes

Recent reports in the literature have indicated a high rate of *Cryptosporidium* sp. false positive results associated with Rapid Cartridge Assays such as Meridian's Immunocard Stat and Remel's x/pect *Giardia/Cryptosporidium*. New York State was selected by CDC/APHL as one of four study sites to compare results obtained from RCAs with those of gold standard tests. In order to best evaluate the performance of the RCAs we will test all specimens from NY patients that are positive for *Cryptosporidium* by rapid cartridge or lateral flow assays. If your lab currently uses one of these tests you should have received a letter asking you to participate in this important study. If you have any questions concerning the study please contact the Parasitology Laboratory at 518-474-4177 or email us at parasite@wadsworth.org.

Parasitology Comprehensive 2 October 2012

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. This document reports the results for the October 2012 proficiency test in Parasitology-Comprehensive. Most laboratories in this category previously participated in the Parasitology-General category, which was renamed after the June 2011 event.

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 Parasitology Laboratory of the New York State Department of Health, and were assayed for quality and confirmation of organisms. Extensive quality control tests were also conducted by the supplying vendor 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.

12-K (All Parasites)

Correct identification: *Giardia lamblia*.

Results of Participating Laboratories

Organism reported	# of labs reporting	% of labs reporting	Referee results	Status
<i>Giardia lamblia</i>	104/104	100	10/10	Correct
<i>Blastocystis hominis</i>	35	34	2	No Penalty
<i>Endolimax nana</i>	26	25	4	No Penalty
<i>Entamoeba histolytica</i>	2	2	1	No Penalty
<i>Entamoeba coli</i>	3	3	0	No Penalty

Quality Control and Referee Information

Participating and referee laboratories agreed that *Giardia lamblia* was the correct response (100%). Quality control examination of 4% of this sample showed cysts in every 5-6 40 X fields. *Endolimax nana*, *Blastocystis hominis* and *Entamoeba* sp. are also present. Other tests performed included a Direct Immunofluorescent Assay, which was negative for *Cryptosporidium* sp. and positive for *Giardia lamblia* and a modified acid-fast stained slide, which was negative.

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. Trophozoites are pear shaped and measure 10-20 μ m. They have 2 nuclei, 4 pair of flagella, 2 axonemes, and 2 median bodies. The infective cysts are oval and measure 11-15 μ m. They contain 4 nuclei usually located at one end, filaments, and median bodies.

12-L (Helminths Only)

Correct identification: *Ascaris lumbricoides*.

Results of Participating Laboratories

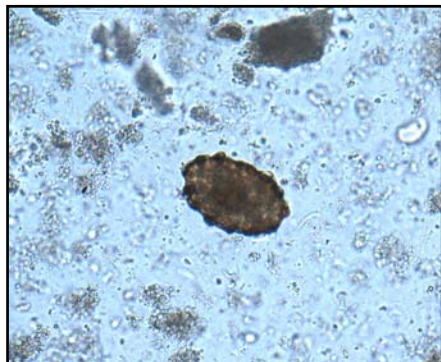
Organism reported	# of labs reporting	% of labs reporting	Referee results	Status
<i>Ascaris lumbricoides</i>	106/107	99	10/10	Correct
<i>Trichuris trichiura</i>	7	7	0	No Penalty
<i>Paragonimus westermani</i>	2	2	1	No Penalty
No Parasites Seen	1	1	0	Incorrect

Quality Control and Referee Information

Participating and referee laboratories agreed that *Ascaris lumbricoides* was the correct response (99 and 100%). Quality control examination of 4% of this sample showed an average of 10 ova per coverslip. Other tests performed included a Direct Immunofluorescent Assay for *Giardia lamblia*

and *Cryptosporidium* sp., which was negative for both organisms. A modified acid-fast stained slide was also negative.

Diagnostic Characteristics



Ascaris lumbricoides is one of the most common intestinal nematode infections of man. It is most prevalent in warm moist climates but can also be found in cooler areas. Infection is acquired when embryonated eggs in contaminated soil are ingested. The fertilized eggs are round to oval, mammillated, and golden brown in color. They measure 45-75 μm by 35-50 μm . Occasionally they may lose their outer mammillated layer. Infertile eggs, like the one shown in the image at left, are larger, less broad, and have thinner shells. They measure 85-90 μm by 43-47 μm .

12-M (Helminths Only)

Correct identification: No Parasites Seen.

Results of Participating Laboratories

Organism reported	# of labs reporting	% of labs reporting	Referee results	Status
No Parasites Seen	103/104	99	10/10	Correct
<i>Ascaris lumbricoides</i>	1	1	0	Incorrect

Quality Control and Referee Information

Participating and referee laboratories agreed that **No Parasites seen** was the correct response (99 and 100%). Quality control examination of 4% of this sample showed normal fecal elements and no organisms present. Other tests performed included a Direct Immunofluorescent Assay for *Giardia lamblia* and *Cryptosporidium* sp., which was negative for both organisms and a modified acid-fast stained slide, which was also negative.

12-N (Protozoa Only)

Correct identification: *Entamoeba histolytica*.

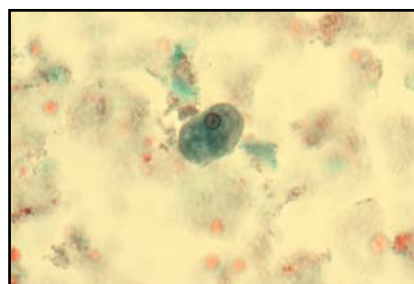
Results of Participating Laboratories

Organism reported	# of labs reporting	% of labs reporting	Referee results	Status
<i>Entamoeba histolytica</i>	92/104	88	10/10	Correct
<i>Entamoeba hartmanni</i>	52	50	7	No Penalty
<i>Entamoeba coli</i>	14	14	0	No Penalty
<i>Blastocystis hominis</i>	12	12	2	No Penalty
<i>Iodamoeba butschlii</i>	1	1	0	No Penalty

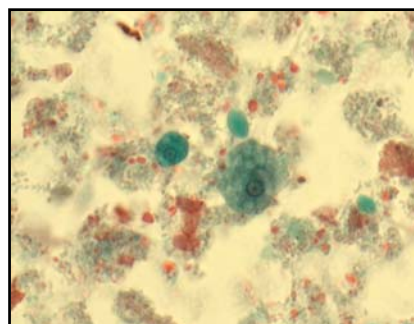
Quality Control and Referee Information

Participating and referee laboratories agreed that *Entamoeba histolytica* was the correct response (88 and 100%). Quality control examination of 4% of this sample showed parasites in almost every 100 X oil immersion field. Also present are *Entamoeba hartmanni*, *Blastocystis hominis*, *Iodamoeba butschlii* and *Entamoeba coli*.

Diagnostic Characteristics



Entamoeba histolytica is distributed worldwide but is more prevalent in the tropics and subtropics. The trophozoites vary in size from 10-60 Φ m with an average size of 15 μ m. They have a single nucleus that generally has a small centrally located karyosome. The peripheral chromatin is generally smooth and evenly distributed. The cysts measure between 8-15 Φ m with an average of 11 Φ m. The mature cyst has 4 nuclei while the immature cyst can have 1 or 2. Chromatin bars are common and have blunt or rounded ends. Infection occurs by ingesting contaminated food or water.



Nonpathogenic *Entamoeba hartmanni*, shown in the image at left with a much larger *E. histolytica* (to the right), has a worldwide distribution and is morphologically similar to *E. histolytica*. Transmission occurs through the fecal oral route and the diagnosis is made by detecting cysts and trophozoites in stool. The cysts are small, measuring 5-8 Φ m and contain 4 nuclei with small, compact, centrally located karyosomes. Rounded chromatoid bodies may or may not be present. The trophozoites measure from 5-12 Φ m and contain 1 nucleus. The karyosome is compact and usually centrally located. The cytoplasm is finely granular and may contain ingested bacteria but not red blood cells.

12-0 (All Parasites)

Correct identification: *Plasmodium falciparum*.

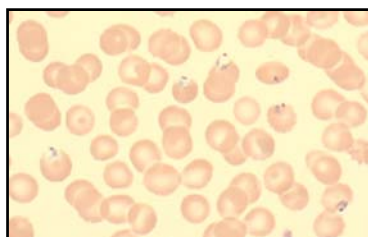
Results of Participating Laboratories

Organism reported	# of labs reporting	% of labs reporting	Referee results	Status
<i>Plasmodium falciparum</i>	89/101	88	10/10	Correct
<i>Babesia</i> sp.	8	8	0	Incorrect
<i>Plasmodium Malariae</i>	4	4	0	Incorrect

Quality Control and Referee Information

Participating and referee laboratories agreed that *Plasmodium falciparum* was the correct response (88 and 100%). Quality control examination of 4% of this sample showed multiple rings in every 100 X oil immersion field. Infected cells are not enlarged, there is no Schüffner's stippling and only rings were observed.

Diagnostic Characteristics



Plasmodium falciparum is one of the four species of Plasmodium known to infect humans. It causes the most dangerous and severe form of malaria and is always considered to be a medical emergency. Death may occur rapidly if proper treatment is not started immediately. *P. falciparum* distribution is limited to the tropics, primarily Africa and Asia. Because this species invades all ages of RBCs the parasitemia can exceed 50%. The usual stages seen in the peripheral blood are rings and gametocytes. Schizogony occurs in the internal organs so it is rare to see other stages although they may be present in cases of severe malaria. The infected RBCs are not enlarged nor do they contain Schüffner's dots. The rings are generally small, and may have one or two chromatin dots. Appliqué forms are also characteristic as shown in the image above. Gametocytes are rounded to banana-shaped and contain a single well-defined chromatin and coarse rice-grain like pigment.

Scoring Information

Immunoassay Results

<i>Cryptosporidium</i>	12-K		12-L		12-M	
METHOD	-	+	-	+	-	+
Alere Giardia/Cryptosporidium Quik Check (TechLab)	3	0	3	0	3	0
MCC Para-Tect	1	0	1	0	1	0
Cryptosporidium/Giardia DFA	28	0	28	0	28	0
Meridian ImmunoCard STAT	17	0	17	0	17	0
Cryptosporidium/Giardia Meridian Merifluor	1	0	1	0	1	0
Cryptosporidium Meridian Premier	15	1	16	0	13	3

EIA						
Remel Xpect	4	0	4	0	4	0
Giardia/Cryptosporidium						
TechLab Cryptosporidium II	2	0	2	0	2	0
ELISA						
TechLab/Wampole Test	4	0	4	0	4	0
EIA						

Giardia	12-K		12-L		12-M	
METHOD	-	+	-	+	-	+
Alere Giardia/Cryptosporidium	0	3	3	0	3	0
Quik Check (TechLab)						
MCC Para-Tect	0	1	1	0	1	0
Cryptosporidium/Giardia DFA						
Meridian ImmunoCard STAT	0	28	28	0	28	0
Crypto/Giardia						
Meridian Merifluor	0	13	13	0	13	0
Crypto/Giardia						
Meridian Premier Giardia	0	1	1	0	1	0
Remel ProspeCT	0	24	24	0	24	0
Giardia EIA						
Remel ProSpecT	0	2	1	1	2	0
Giardia EZ						
Remel Xpect	0	2	2	0	2	0
Giardia						
Remel Xpect	0	4	4	0	6	0
Giardia/Cryptosporidium						
TechLab/Wampole Test	0	7	7	0	7	0
EIA						
TechLab Giardia II ELISA	0	2	2	0	2	0

Distribution of Scores

Score	# of labs	% of labs
100	77	74
90-99	2	2
80-89	24	23
60-69	1	1

Answer Key

Sample	Correct Answer	Points
12-K	<i>Giardia lamblia</i>	20
12-L	<i>Ascaris lumbricoides</i>	20

12-M	No Parasites Seen	20
12-N	<i>Entamoeba histolytica</i>	20
12-O	<i>Plasmodium falciparum</i>	20

TOTAL POSSIBLE POINTS 100

Grading

The answer key was derived from the response of all participating laboratories as per **CLIA Regulations**, Part 493, Subpart I, Section 493.917. These regulations can be viewed at www.cdc.gov/clia/regs/toc.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.

Each sample has a maximum value of 20 points. Credit is given according to the formula:

$$(\# \text{ of Correct Responses} / (\# \text{ of Correct Responses} + \# \text{ of Incorrect Answers})) \times 100$$

Important Reminders

The next Parasitology Proficiency Test is scheduled for **February 5, 2013**. You are responsible for notifying us **before February 12, 2013** if you do not receive your samples. Proficiency test results must be electronically submitted through EPTRS by **February 19, 2013** or the laboratory will receive a score of zero. These requirements are stated in the NYS Proficiency Testing Handbook provided by the NYS Clinical Laboratory Evaluation Program or can be accessed via the Internet at:

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