NEW YORK STATE

Parasitology Proficiency Testing Program

Blood Smears Only 07 February 2012

The purpose of the New York State Proficiency Testing Program in the category of Parasitology - Blood Smears Only is to monitor the performance of applicant laboratories that detect and identify parasites on blood films. This document reports the results for the February 2012 proficiency test in Blood Smears Only. Most laboratories in this category previously participated in the Parasitology-Blood Borne Parasites Only category, which was renamed after the June 2011 event.

This category is divided into two sub-categories. **Parasite Identification** is intended for labs that identify parasites and report them to the species level on patient reports. **Parasite Screen** is intended for labs that report "Parasites Seen" and never report organisms to the species level on patient reports. Participants in both sub-categories examine the same samples, however the scoring criteria for the two sub-categories are different. When reading this critique, ensure that you are comparing your performance to other laboratories in your sub-category.

Sample Preparation and Quality Control

All slides used in this test were prepared and stained by a commercial source. Numerous samples of each test specimen were selected at random by the Parasitology Unit of the David Axelrod Institute for Public Health, and were assayed for quality and confirmation of contents. Extensive quality control tests were also conducted by the supplying vendor and a detailed quality control report was submitted to the New York State Parasitology Laboratory for inspection and verification. Samples were authenticated by 80% of participating laboratories and/or referee laboratories.

12B-A

Correct identification: Leishmania sp.

Results of Participating Laboratories Who Perform Parasite Identification

Organism reported	# of labs reporting	% of labs reporting	Referee results	Status
Leishmania sp.	20/21	95	10/10	Correct
Trypanosoma cruzi	1	5	0	Incorrect

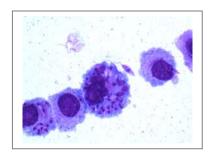
Results of Participating Laboratories Who Perform Parasite Screen

Organism reported	# of labs reporting	% of labs reporting	Referee results	Status
Parasites Seen	2/2	100	10/10	Correct

Quality Control and Referee Information

Participating and referee laboratories agreed that *Leishmania* **sp.** was the correct response (95 and 100%). Quality control examination of 4% of this sample showed parasites in every 100 X oil immersion field. Both promastigotes and amastogotes are present inside and outside macrophages. The nuclei and kinetoplasts are clearly visible. The overall staining quality is good.

Diagnostic Characteristics



Leishmania spp. are intracellular protozoan parasites transmitted to humans through the bite of infected sand flies. Leishmaniasis is a disease found primarily in the tropics and subtropics. Infection causes two main types of disease, cutaneous leishmaniasis and visceral leishmaniasis. The disease type is determined by the infecting species, geographic location and the immune response of the host. Cutaneous leishmaniasis causes skin lesions that can range in severity from relatively mild, self-healing lesions to debilitating mucocutaneous lesions. Visceral leishmaniasis also

has a wide range of disease severity from subclinical to disseminated visceral disease which leads to death in untreated patients.

This parasite is found in two morphological forms, amastigotes and promastigotes. Diagnosis is traditionally made by detecting amastigotes on Giemsa stained slides made from the infected tissue, i.e. skin, bone marrow or spleen. The amastigotes are small, oval intracellular forms that have a nucleus and a kinetoplast. Promastigotes are elongated extracellular forms that have a flagellum and are transmitted from the vector to the host during a bite. This sample was taken from a culture of macrophage cells infected with visceral *Leishmania* and contains both amastigotes and promastigotes.

12B-B

Correct identification: No Parasites Seen.

Results of Participating Laboratories Who Perform Parasite Identification

Organism reported	# of labs reporting	% of labs reporting	Referee results	Status
No Parasites Seen	20/21	95	10/10	Correct
Trypanosoma cruzi	1	5	0	Incorrect

Results of Participating Laboratories Who Perform Parasite Screen

Organism reported	# of labs reporting	% of labs reporting	Referee results	Status
No Parasites Seen	2/2	100	10/10	Correct

Quality Control and Referee Information

Participating and referee laboratories agreed that **No Parasites Seen** was the correct response (95 and 100%). Quality control examination of 4% of this sample showed erythrocytes of normal size and staining characteristics. Normal blood elements are present and exhibit typical staining characteristics. The overall staining quality is good.

12B-C

Correct identification: *Plasmodium falciparum*.

Results of Participating Laboratories Who Perform Parasite Identification

Organism reported	# of labs reporting	% of labs reporting	Referee results	Status
Plasmodium falciparum	15/21	71	10/10	Correct
Plasmodium vivax	3	14	0	Incorrect
Plasmodium malariae	3	14	0	Incorrect

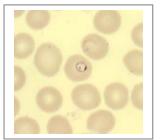
Results of Participating Laboratories Who Perform Parasite Screen

Organism reported	# of labs reporting	% of labs reporting	Referee results	Status
Parasites Seen	2/2	100	10/10	Correct

Quality Control and Referee Information

Referee laboratories agreed that *Plasmodium falciparum* was the correct response (100%). Quality control examination of 4% of this sample showed parasites in every 15-20 100 X oil immersion fields. Infected cells are not enlarged and have no stippling. The only stage seen was the ring stage trophozoite. The overall staining quality is good.

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. Its distribution is limited to the tropics, primarily Africa and Asia. *P. falciparum* invades all ages of RBCs and so 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 seen

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. Gametocytes are rounded to bananashaped and contain a single well defined chromatin and coarse rice-grain like pigment.

12B-D

Correct identification: Loa loa.

Results of Participating Laboratories Who Perform Parasite Identification

Organism reported	# of labs reporting	% of labs reporting	Referee results	Status
Loa loa	11/21	52	7/10	Unauthenticated
Mansonella sp.	5	24	1	No Penalty
No Parasites Seen	5	24	2	No Penalty

Results of Participating Laboratories Who Perform Parasite Screen

Organism reported	# of labs reporting	% of labs reporting	Referee results	Status
Parasites Seen	2/2	100	10/10	Correct

Quality Control and Referee Information

Participating and referee laboratories failed to agree that *Loa loa* was the correct response. Quality control examination of 4% of this sample showed an average of 4 microfilaria per coverslip. The organisms are sheathed and have nuclei extending all the way to the tail tip. The overall staining quality is good.

Diagnostic Characteristics

Loa loa, also called the African eye worm, infects humans when they are bitten by infected deer or



mango flies. The larvae are deposited into the bite wound and develop into adults within 6-12 months. Adults migrate beneath the conjunctiva or the skin, or through subcutaneous tissues. Years after the initial infection the adults give rise to microfilariae which can be detected in the blood.

The microfilariae are sheathed and measure between $250-300 \, \mu m$. They have nuclei that extend all the way to the tip of the tail. Although the sheath does not stain with Giemsa it is still visible as a clear area around the stained body, as shown in the images above.

12B-E

Correct identification: Plasmodium malariae.

Results of Participating Laboratories Who Perform Parasite Identification

Organism reported	# of labs reporting	% of labs reporting	Referee results	Status
Plasmodium malariae	20/21	95	10/10	Correct
Plasmodium vivax	1	1	0	Incorrect

Results of Participating Laboratories Who Perform Parasite Screen

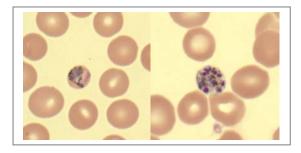
Organism reported	# of labs reporting	% of labs reporting	Referee results	Status
Parasites Seen	2/2	100	10/10	Correct

Quality Control and Referee Information

Participating and Referee laboratories agreed that *Plasmodium malariae* was the correct response (95 and 100%). Quality control examination of 4% of this sample showed parasites in almost every 100 X oil immersion field. Infected cells are not enlarged and most are in fact smaller than uninfected cells. The predominant stages seen were mature trophozoites and schizonts. The staining quality is good.

Diagnostic Characteristics

Plasmodium malariae is the least common species of malaria to infect humans, and is sporadic in



distribution. It tends to infect older red blood cells and so the parasitemia is often low. The ring stage is short lived so it is not usually seen. The most common stages seen are mature trophozoites and schizonts. The infected cells are not enlarged and may actually be smaller than uninfected cells. There is no stippling. The trophozoites (left) are not amoeboid and often appear as compact rounded or band forms. The schizonts (right) contain 6-12

merozoites which are usually arranged in a rosette, although they may be in an irregular cluster.

Scoring Information

Distribution of Scores

Score	# of labs	% of labs
100	15/23	65
80-89	6	26
60-69	1	4
40-49	1	4

Answer Key

Sample	Correct Answer	Points
12B-A	Leishmania sp.	20
12B-B	No Parasites Seen	20
12B-C	Plasmodium falciparum	20
12B-D	Loa loa	20
12B-E	Plasmodium malariae	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.phppo.cdc.gov. These regulations state that 80% or more of participating laboratories **or** referee laboratories must identify the parasite for it to be correct. Similarly, reporting of a parasite identified by less than 10% of the participating laboratories **or** referees finding parasites or ova is an incorrect response. Organisms reported by more than 10% but less than 80% 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:

Number of correct responses by lab X 100 # Correct Parasites Present + # Lab's Incorrect Answers

Important Reminders

The next Parasitology Proficiency Test is scheduled for **May 15, 2012**. You are responsible for notifying us **before May 22, 2012** if you do not receive your samples. Proficiency test results must be electronically submitted through EPTRS by **May 29, 2012** 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/WebGuide.pdf

News and Notes

There will be an intensive one-day hands-on workshop on the identification of blood borne parasites held on May 8, 2012 at the Center for Medical Science in Albany, NY. For registration information please go to http://www.wadsworth.org/parasitology/index.htm or phone us at 518-474-4177 or send an email to parasite@wadsworth.org

Beginning with the February 2009 proficiency exam, the **grading policy changed**. In order to make the score on the NYS Parasitology PT exam more accurately reflect laboratory performance, and be more consistent across categories, a new scoring system was put into effect. Under the new scoring system, grades are based only on the specimen or organism types processed by your laboratory.