NEW YORK STATE
Parasitology Proficiency Testing Program

Blood Borne Parasites
05 October 2010

The purpose of the New York State Proficiency Testing Program in the category of Blood Borne Parasites is to monitor the performance of applicant laboratories in detecting and identifying parasites on blood films. This document reports the results for the October 2010 proficiency test in Blood Borne Parasites.

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.

10B-K

Correct diagnosis: No Parasites Seen.

Results of Participating Laboratories

Organism reported	# of labs reporting	% of labs reporting	Referee results	Status
No Parasites Seen	19/20	95	10/10	**Correct**
Plasmodium malariae	1	5	0	Incorrect

Quality Control and Referee Information

Participating and referee laboratories agreed that **No Parasites Seen** was the correct response (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.

10B-L

Correct diagnosis: *Trypanosoma brucei.*

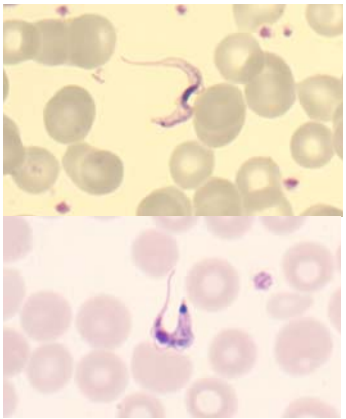
Results of Participating Laboratories

Organism reported	# of labs reporting	% of labs reporting	Referee results	Status
Trypanosoma brucei	17/20	85	8/10	**Correct**
Trypanosoma cruzi	2	10	1	Incorrect
Trypanosoma gambiense	1	5	1	Incorrect

Quality Control and Referee Information

Participating and referee laboratories agreed that ***Trypanosoma brucei*** was the correct response (85 and 80%). Quality control examination of 4% of this sample showed parasites in every 8-10 100 X oil immersion fields. The parasites have a large central nucleus and a small posterior kinetoplast. The staining quality is good.

Diagnostic Characteristics



Top: *T. brucei*. Bottom: *T. cruzi*.

Trypanosoma brucei is the causative agent of African sleeping sickness (human African trypanosomiasis, HAT). Trypanosomiasis is limited to the tse-tse fly endemic area of sub-Saharan Africa where it has and does have a significant economic and health impact. There are 3 subspecies of *T. brucei*. *T. brucei brucei* causes a wasting disease in cattle but is not a human pathogen. *T. brucei gambiense* and *T. brucei rhodesiense* are both human pathogens causing different forms of African trypanosomiasis. The three subspecies are morphologically indistinguishable and thus can only be identified to the species level by microscopy.

Trypomastigotes are detected in the blood on thick and thin Giemsa-stained smears. They measure 14-33 µm long and in some cases the undulating membrane and flagellum may be seen. The nucleus is located in the middle of the organism and the *kinetoplast*, which is the mitochondrial DNA, is located at the posterior end. On a Giemsa-stained smear the cytoplasm will stain blue and the nucleus and kinetoplast will stain red or purple.

Trypomastigotes of *T. cruzi* are similar but are generally shorter, have a 3-5X larger, more prominent kinetoplast at the posterior end, and typically assume a C or U shape.

10B-M

Correct diagnosis: *Plasmodium vivax.*

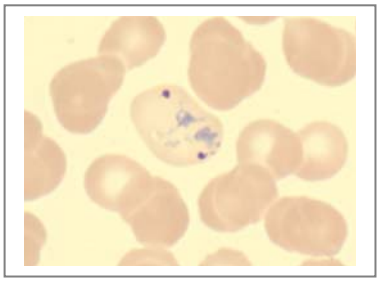
Results of Participating Laboratories

Organism reported	# of labs reporting	% of labs reporting	Referee results	Status
Plasmodium vivax	8/20	40	5/10	**Unauthenticated**
Plasmodium ovale	8	40	3	No Penalty
Plasmodium falciparum	2	10	0	No Penalty
Plasmodium malariae	2	10	2	No Penalty

Quality Control and Referee Information

Participating and referee laboratories failed to agree that ***Plasmodium vivax*** was the correct response (40 and 50%). Quality control examination of 4% of this sample showed parasites in every 15-20 100 X oil immersion fields. Infected cells are enlarged and pale staining. The predominant form seen is the amoeboid trophozoite. Where present the pigment is scattered and fine. The staining quality is good.

Diagnostic Characteristics



Plasmodium vivax is the most common species of malaria to infect humans. It may account for as much as 80% of all malaria cases. It also has the widest distribution. Infected red cells are usually enlarged and stain paler than uninfected ones. They may also contain Schüffner's dots. The trophozoites are generally amoeboid and have a large chromatin. Occasionally cells will contain more than one parasite. Mature schizonts contain 12-24 merozoites and gametocytes are round and fill the entire cell. Pigment is fine and scattered.

10B-N

Correct diagnosis: *Plasmodium malariae*.

Results of Participating Laboratories

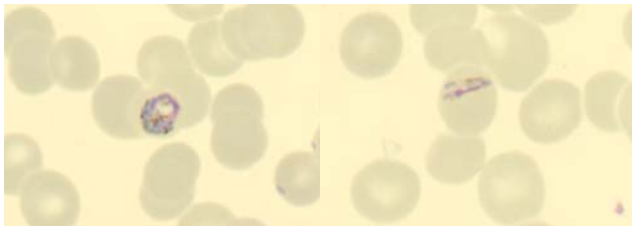
Organism reported	# of labs reporting	% of labs reporting	Referee results	Status
Plasmodium malariae	20/20	100	10/10	**Correct**

Quality Control and Referee Information

Participating and referee laboratories agreed that ***Plasmodium malariae*** was the correct response (100%). Quality control examination of 4% of this sample showed parasites in every 5-8 100 X oil immersion fields. Infected cells are not enlarged and no Schüffner's stippling was observed. The predominant stage seen was the trophozoite with many band and basket forms present. Where present the pigment is coarse. The staining quality is fair.

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 are not amoeboid and often appear as compact rounded or band forms. The schizonts contain 6-12 merozoites usually arranged in a rosette although they may be in an irregular cluster.



Plasmodium malariae basket (left) and band (right) forms.

10B-O

Correct diagnosis: *Plasmodium falciparum.*

Results of Participating Laboratories

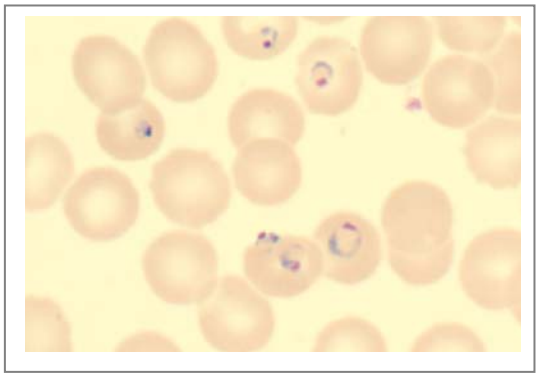
Organism reported	# of labs reporting	% of labs reporting	Referee results	Status
Plasmodium falciparum	20/20	100	10/10	**Correct**

Quality Control and Referee Information

Participating and referee laboratories agreed that ***Plasmodium falciparum*** was the correct response (100%). Quality control examination of 4% of this sample showed parasites in every 100 X oil immersion field. The infected cells are not enlarged. There are many rings, some with double chromatin, and appliqué forms. The staining quality is good.

Diagnostic Characteristics

Plasmodium falciparum is one of the four species of *Plasmodium* know 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 RBC's 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 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. Gametocytes are rounded to banana- shaped and contain a single well defined chromatin and coarse rice-grain like pigment.



Scoring Information

Distribution of Scores

Score	# of labs	% of labs
100	16/20	80
80	4	20

Answer Key

Sample	Correct Answer	Points
10B-K	No Parasites Seen	20
10B-L	*Trypanosoma brucei*	20
10B-M	*Plasmodium vivax**	20
10B-N	*Plasmodium malariae*	20
10B-O	*Plasmodium falciparum*	20

TOTAL POSSIBLE POINTS 100

* Unauthenticated

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:

$$\frac{\text{Number of correct responses by lab}}{\text{\# Correct Parasites Present + \# Lab's Incorrect Answers}} \times 100$$

Important Reminders

The mailout dates for Parasitology have been changed from the first Monday of February, June, and October to the first Tuesday.

The next Parasitology Proficiency Test is scheduled for **February 1, 2011.** You are responsible for notifying the New York State Parasitology Unit **before February 8, 2011** if you do not receive your test. Proficiency test results must be electronically submitted through EPTRS by **February 15, 2011** or you will receive a zero. These requirements are clearly stated in your NYS Proficiency Testing Handbook provided by the NYS Clinical Laboratory Evaluation Program, and can be accessed via the Internet at:
http://www.wadsworth.org/labcert/clep/ProgramGuide/WebGuide.pdf

News and Notes

Beginning with the February 2009 proficiency exam, the **grading policy has 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 is in effect. Under the new scoring system, grades will be based only on the specimen or organism types processed by your laboratory. Laboratories that process all of the types of samples included in the exam will not observe any changes in scoring method.