8.8.2023 Best Practices in Parasitology, Addressing Common Diagnostic Challenges Q&A Responses

Question: Can you provide an example of a permanent stain for fecal parasites?
Answer: Some examples of permanent stains suitable for fecal smears include Trichrome, Iron Hematoxylin (Modified Spencer-Monroe method), and the VIDRL modification of the Trichrome procedure. The Clinical Microbiology Procedures Handbook, 5th edition contains detailed information on how to perform these three staining techniques.

Question: Does CAP offer any guidance for the implementation of stool autofluorescence microscopy for coccidian protozoa (e.g., Cyclospora, Cystoisospora)?
Answer: Autofluorescence of the oocysts of these coccidia can be used for diagnosis in laboratories equipped with an appropriate fluorescent microscope.
The following reference provides some brief procedural details how to process the stool specimen and examine using a fluorescent microscope-
Berlin O, Peter J, Gagne C, et al. Autofluorescence and the Detection of Cyclospora Oocysts. Emerging Infectious Diseases. 1998;4(1):127-128. doi:10.3201/eid0401.980121.
Regulatory guidance for the validation and verification of test methods, including stool autofluorescence microscopy for coccidian protozoa (e.g., Cyclospora, Cystoisospora), can be found within the All Common Checklist.

Question: Where can I find more information about *Angiostrongylus cantonensis*?
Answer: Information about *Angiostrongylus cantonensis* can be accessed through the Expanded Parasitology Survey (PEX) Master List.
A good reference for *Angiostrongylus cantonensis* parasite biology, images, laboratory diagnosis, and additional resources can be found on the CDC's DPDx Laboratory Identification of Parasites of Public Health Concern website- https://www.cdc.gov/dpdx/angiostrongyliasis_can/index.html

Question: What level of quantitation of stool ova and parasites is considered significant?
Answer: It is recommended to report the organism and stage (trophozoite, cyst, egg, larvae, etc.) identified in the O&P exam. A suggested quantification scheme for parasites is:
Few = ≤2 per 10 oil immersion fields (x1,000)
Moderate = 3 to 9 per 10 oil immersion fields (x1,000)
Many = ≥10 per 10 oil immersion fields (x1,000)
Rare = can be used for scarcely quantified organisms that are present at very low levels.

Question: Where can I access the M 28 and M15 material?
Answer: Access to the M28-A2 (Recovery & ID of Parasites from Intestinal Tract) and

M15-A (Lab Diagnosis of Blood-borne Parasitic Diseases) materials can be obtained through the CLSI website using the following links:

- M28: Recovery & ID of Parasites from Intestinal Tract (clsi.org)
- M15AE: Lab Diagnosis of Blood-borne Parasitic Diseases (clsi.org)

Question: Which survey should I order if my laboratory conducts counts on thick blood films?

Answer: Both the Blood Parasite (BP) and Parasitology (P) Surveys offer percent parasitemia educational challenges that are suitable for laboratories calculating percent parasitemia from thick blood films.

Question: If both concentration and permanent stain are required, why does the CAP still offer a PT option (Parasitology Assessment) with materials for wet mounts only, without fixed smears?

Answer: In clinical practice the direct wet preparation/direct smear is helpful in assessing parasite burden and identifying motile flagellates and other helminths. The P3 Parasitology PT/EQA product contains 5 challenges if a laboratory would like more wet mount challenges than is included in the P or P4 programs.

Question: How should quality control (QC) be conducted for concentration and permanent staining methods?

Answer: Quality control consists of documenting the proper functioning of reagents and equipment on the days patient testing is performed. Known positive specimens can be used; but if not available, smears of stool specimens containing epithelial cells or white cells can be considered. If desired, an individualized quality control plan (IQCP) can be developed to decrease the frequency QC is required. QC should always be performed when lots of new reagent are implemented.
The CAP Microbiology Checklist requirement MIC.51160 offers guidance for conducting QC on permanent parasitology stains.

Question: Is there a specific CAP External Quality Assessment (EQA) program for tissue parasites/parasites in H&E tissue sections?

Answer: The CAP Parasitology (P) Survey Master List includes tissue parasites and may be presented on a rotational basis.

Question: Fasciola and Diphyllobothrium eggs are quite similar. How can they be differentiated?

Answer: *Fasciola* eggs (130-150 micrometers x 60-90 micrometers) can primarily be distinguished from *Diphyllobothrium* eggs (58-75 micrometers x 40-50 micrometers) based on their size. *Dilphyllobothrium latum* also has a small,

inconspicuous knob at the abopercular end of the egg that is not present on *Fasciola* spp. eggs.

Question: How can parasitemia be estimated?
Answer: Parasitemia is calculated from the thin smear using the following calculation- percent parasitemia (%) = (# of infected RBCs/total # of RBCs counted) x 100. Additional information on calculating parasitemia from thick and thin smears is found in this resource- Parasitemia_and_LifeCycle.pub (cdc.gov)

Question: Is it important to report commensal parasites like *Endolimax nana*?
Answer: While *Endolimax nana* is considered a non-pathogenic intestinal amoeba and is found worldwide, it should still be reported if identified in all O&P exams. Its presence indicates fecal-oral contamination and should prompt the laboratory employee to continue to evaluate the slides for pathogenic organisms as well. The detection of *Endolimax nana* and other non-pathogenic amebae does not exclude the presence of a true pathogen.