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Section: Parasitology Manual	Subject Title: Collection and Laboratory Procedures for Specimens Other Than Stool or Blood	
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Duodenal Aspirates

PRINCIPLE

In infections with *Giardia*, *Strongyloides*, *Clonorchis*, *Fasciola* and hookworm species routine stool examinations may be negative when the organisms are present. It may therefore be necessary to obtain a sample of the contents of the duodenum, either by using an Entero-Test capsule, or by more invasive means.

SPECIMENS

Either an Entero-Test capsule or duodenal drainage should be submitted. If the sample will not be examined within 2hrs add 10% formalin. The sample may vary in volume from <0.5ml to several mls. Often the duodenal fluid may contain mucus; this is where the organisms will tend to be found. Centrifugation of the specimen is important, and the sedimented mucus should be examined. If the amount of duodenal material submitted is very small, then permanent stains can be prepared rather than using any of the specimen for a wet smear examination.

SAFETY

Consider if *Strongyloides* larvae might be present.

REAGENTS

Saline (commercial product, PML)
 If needed 0.85% NaCl (9g/L NaCl)
 phosphate buffer
 Na₂HPO₄ 10.7 g
 NaH₂PO₄ 0.23 g
 Water 11,200.00 ml
 Giemsa stain (commercial product, VWR)
 methanol

PROCEDURE

1. If the sample is an aspirate then centrifuge the specimen at 500g for 5 minutes.
2. If the sample is supplied as an Entero-Test capsule then scrape or “milk “ the string to release material into a tube.

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3. Place a drop of sediment on a microscope slide.
4. A coverslip is added and the preparation is examined under low power. Examination of the slide under 400X may be useful to detect the flutter of a *Giardia* flagella.
5. For speciation the sample can be fixed in methanol and stained with 1:50 dilution of Giemsa stain for 30 minutes.

QUALITY CONTROL

- It may take organisms several minutes to acclimate to their new environment and to start moving again.
- If limited sample is available do the permanent stained smears first.
- Examine 5 or 6 drops of material before declaring the sample negative
- Test pH of the string for alkaline pH (ie. to ensure it was in the duodenum) and look for a yellow-green color on the section that should have reached the duodenum.
- As it may not be possible to have a positive control specimen to use with this procedure, the technologist should review the appearance and size of the organisms present to ensure that they match reference material (i.e. *Bench Aids for the Diagnosis of Intestinal Parasites* (WHO)).
- Ensure that the microscope has been calibrated in the last year and that the results of the calibration are displayed on the microscope base.
- The presence of the “falling leaf” motility of *Giardia* in fresh samples is an excellent indication of its presence.
- Monoclonal FA or EIA detection kits are available for *C. parvum* and *G. lamblia*.
- All QC results should be recorded.

REPORT

Report the organism seen and the stage (trophozoite, cyst, oocyst).

LIMITATIONS OF PROCEDURE

- Many of the parasites will get caught up in mucus, therefore centrifugation may be required.

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- Some organisms are more easily seen in wet preps, others are only visible with staining.

AUTHOR

Ian Crandall

REFERENCES

Beal, C.B., P. Viens, R.G.L. Grant, and J.M. Hughs 1970 A new technique for sampling duodenal contents: Demonstration of upper small-bowel pathogens. *American Journal of Tropical Medicine and Hygeine* 19:349-352

NCCLS Guidlines, M28-A, Vol. 17 No.23 Dec. 1997 Procedures for the recovery and identification of parasites from the intestinal tract; approved guide. pp45-47