Flow Cytometry Service Submission Guidelines

Quick Guide

• Fill out submission form ENTIRELY; submit via lab portal: https://ohio.labs.tracefirst.com

• Submit peripheral blood (≥0.5mL) in EDTA (LTT)
  - Include either a recent CBC report OR additional blood for the Clinical Pathology Service to perform a CBC

• Submit tissue aspirates (e.g. lymph node) into 1 mL of flow media in a no-additive tube
  - Flow media = 90% normal saline + 10% serum (from the same species)

• Ship OVERNIGHT with an ice pack for delivery Monday-Friday business days only to:
  Ohio State Veterinary Medical Center
  ATTN: Flow Cytometry for Clin Path Room #0044
  601 Vernon L. Tharp Street
  Columbus, OH 43210

• Turn-around time is 1-3 business days; results available through the lab portal

• Questions about what test to run, what sample to submit or what the results mean?
  - Call or email Dr. Samantha Evans at 614-292-9706 or evans.2608@osu.edu

Detailed Guide and FAQs

When do you accept samples?

• Samples are accepted during business days (Monday through Friday, except for university holidays) and must arrive at the Veterinary Medical Center before noon on Friday.

• Samples are NOT accepted on the weekends or after noon on Fridays because the samples will not be sufficiently viable to run flow cytometry on the next business day. These samples will be rejected (without charging the clinic).

What species’ samples do you accept?

• Canine and feline

What samples do you accept? continued on page 2

Peripheral blood

• Submit ≥0.5mL in EDTA (LTT)

• Submit a recent (<48h) CBC report OR 0.5mL additional blood (1mL total blood volume) for the Flow Cytometry Service to perform a CBC

• A pathology review of the blood smear is not required but is highly recommended for peripheral blood samples. At a minimum, please ensure that a DVM has reviewed a blood smear prior to sending in blood for flow cytometry. Automated analyzers are sometimes known to provide false results which mislead veterinarians into thinking that flow cytometry is indicated when it is not. Moreover, the morphology of the cells can be helpful in interpreting flow cytometry results.

• If no CBC report is included in your submission, you will automatically be charged for a CBC (cost = $64.25).

NOTE: Flow cytometry is most diagnostic when there is a lymphocytosis (or “other” leukocytosis in the case of myeloid neoplasia) present in peripheral blood. With a normal or low lymphocyte count, there would usually need to be a very markedly abnormal phenotype in order to obtain a diagnostic result.
What samples do you accept? continued from page 1

Tissue aspirates (lymph node, mediastinal mass, abdominal organs, etc.)
- Prepare flow media in a no-additive tube (RTT/WTT/microcentrifuge tube): 90% saline (normal saline or LRS) + 10% serum from the same species. For most samples, 1mL volume for submission is ideal, so that would be 0.9mL of saline + 0.1mL serum.
- Aspirate the lesion several times using suction
- Squirt the contents into the tube with flow media and re-aspirate the media into the syringe a couple of times to rinse out the needle and syringe.
- Repeat the aspiration and rinsing procedure until the solution is turbid. (Turbidity or cloudiness indicates that the sample is sufficiently cellular).

NOTE: It is best to aspirate the most affected (i.e. most cellular and most effaced by neoplastic cells) organ. If multiple organs are equally affected, choose lymph nodes over other organs. Spleen can be difficult to interpret, so it is best not to choose spleen if there are other highly-affected organs (ex: liver) which can be aspirated instead. Feel free to call Dr. Evans if there is a question about what to submit.

Cavity fluid (abdominal, thoracic, pericardial, synovial)
- Submit 0.5mL in EDTA (LTT)
- If the protein content of the fluid is <4.0 g/dL, add ~10% serum from the same species (ex: add 0.1mL of serum to a tube containing 1mL of cavity fluid). If the protein content is ≥4.0 g/dL, no addition of serum is necessary.

Cerebrospinal fluid (CSF)
- CSF is very rarely cellular enough for flow cytometry. A minimum estimated 200,000 cells are needed for a diagnostic flow cytometry result. So if you have 0.5mL of CSF, the cell count would need to be at least 400 cells/µL (normal is <5 cells/µL).

How should samples be shipped?
- Please fill out the submission form ENTIRELY and include this with your shipment.
- Keep sample refrigerated until shipment.
- Send samples OVERNIGHT with an ice pack, but do not freeze. Do not use dry ice as this may freeze the sample.
- Send samples to the following address (UPS shipments preferred):
  Ohio State Veterinary Medical Center
  ATTN: Flow Cytometry for Clin Path Room #0044
  601 Vernon L. Tharp Street
  Columbus, OH 43210
- Highlight the “Attn: Flow Cytometry” section if you can; this will alert our receiving department to deliver the package to clinical pathology promptly.

What is the cost of flow cytometry & how do we pay?
- Cost = $120 per sample
- Your clinic’s account will be billed by the VMC.

When and how will I receive results?
- Turn-around time is expected within 1-3 business days from receipt of the sample (ex: if you ship the sample on Monday and it arrives on Tuesday, you can expect results between Wednesday and Friday).
- Reports will be available through the VMC CoreOne portal (https://ohio.labs.tracefirst.com). It can be set to send an email notification when results are ready.

Do you run PARR (PCR for antigen receptor rearrangement), XCIP (X-chromosome inactivation pattern), histiocytic malignancy (HM), BRAF mutation, or other clonality and genetic-based assays?
- We do not run genetic testing in-house, but we will save the flow pellet and left-over sample for up to 2 weeks. If additional testing is warranted, we can ship this material to another reference lab for the appropriate testing.

Who can I contact with additional questions?
- For logistical questions (shipping, handling, portal, etc), please contact the VMC Hematology Laboratory at 614-292-7955.
- For questions regarding when to order flow cytometry, what sample to submit, what the results mean, diagnostic next steps, etc., please contact Dr. Samantha Evans at 614-292-9706 or evans.2608@osu.edu.