



Clinical Flow Cytometry FAQs

What is flow cytometry?

Flow cytometry is a test that determines the immunophenotype of cells by recognizing normal and abnormal expression of cell surface markers on T cells (helper T cells, cytotoxic T cells), B cells, neutrophils, and monocytes.

Clinical flow cytometry is most often used for cats and dogs:

1. To help distinguish between reactive and neoplastic lymphocytes
2. To determine if lymphoma/leukemia is of B or T cell origin and in some cases, identify specific subtypes of B or T cell lymphoma/leukemia which will aid in prognosis
3. To differentiate between lymphoid and non-lymphoid (myeloid/monocytic) neoplasia

Clinical flow cytometry is used most often in combination with clinical history/presentation and cytology/blood smear examination for diagnosis of disease. In some cases, additional diagnostics (PARR-PCR for antigen receptor rearrangement, histopathology, immunohistochemistry, immunocytochemistry) are also required.

When might clinical flow cytometry be most helpful?

1. **Lymphadenopathy, organomegaly, or mediastinal mass with the following cytology results:**
 - a. Confirmed or probable lymphoma composed of intermediate to large lymphocytes
 - b. Homogeneous expansion of small to intermediate lymphocytes
 - c. Suspected lymphoma or thymoma (for mediastinal mass)

Sample to submit: Fine needle aspirate of lymph node/enlarged organ/mass (see sample submission guidelines)

Note: Flow cytometry is not indicated on these samples if cytology results report rare to low numbers of atypical/suspicious cells

2. **Abnormal CBC findings consisting of:**
 - a. Peripheral lymphocytosis with increased numbers of small mature, intermediate, or immature lymphocytes
 - b. Presence of any immature cells/'blasts' with a normal or elevated white blood cell count

Sample to submit: Peripheral blood (EDTA tube)

NOTE: If the lymphocytosis is composed of small to intermediate lymphocytes test for Ehrlichia infection in dogs with lymphocyte counts up to ~20k (rarely up to 30k) and ruleout 'stress' in cats with lymphocyte counts up to ~20k (rarely up to 30k) prior to running flow cytometry

3. **Abnormal bone marrow findings with the following cytology/CBC results:**
 - a. Increased numbers of blasts or small/intermediate lymphocytes with leukopenia on CBC

Sample to submit: Bone marrow (EDTA tube) for samples with leukopenia on CBC. Peripheral blood (EDTA tube) is preferred if lymphocytosis is present.

4. **Body cavity effusion with the following cytology results:**
 - a. High numbers of small/intermediate/OR large lymphocytes consistent with or concerning for lymphoma

Sample to submit: Cavity fluid (EDTA tube and red top tube when possible- 0.5ml in each tube, see sample submission guidelines)

Uncertain if flow cytometry will be helpful for your patient? Email vetclinflow@uga.edu or contact (706)542-9430 alt phone: (706)542-5161

Sample shipping guidelines

- **Collect samples and ship on the same day overnight for Monday through Friday delivery**
- **Keep samples refrigerated (DO NOT FREEZE) until shipped**
- **Ship overnight with an ice pack**
- Samples may be submitted via:
 - *The Athens Veterinary Diagnostic Laboratory*
 - Courier services may be available in your area
 - Or ship overnight via UPS- \$10 UPS overnight labels available by email
 - For courier service information or for shipping labels by email contact AVDL (706) 542-5568 or visit <http://www.vet.uga.edu/dlab/> for information

Sample collection and submission guidelines

Lymph node, organ or mediastinal mass aspirates

- Put 1 ml of 0.9% saline in a red top tube (*no additives, no serum separator*)
- Add 0.1ml of serum from the same patient or another patient of the same species
 - *Note: the volume of serum should be ~10% of the total volume (this helps preserve cells)*
- Using a 22ga needle and 6 or 12cc syringe, **aspirate with suction**
- Eject the contents into the saline, gently aspirate the saline into the syringe and eject back into the tube to recover all cells
- Aspirate several times (2-3X if possible) repeating the above process **until the saline is slightly cloudy**

Blood and bone marrow

- Provide a minimum of 0.5ml in an EDTA tube
- A current CBC (within 2days) of the flow sample collection is required
 - Include a copy of your patient CBC
 - OR
 - Check the CBC test request box and include: 1) *a minimum of 2.0 ml* of blood in an EDTA tube (use the appropriate fill volume for your tube) for the CBC and flow cytometry and 2) one unstained blood smear
 - See pricing schedule for CBC

Body cavity fluid

- Provide a minimum of 0.5ml in an EDTA tube
- If enough fluid is available please provide an additional red top tube (no additives, no serum separator) with a minimum of 0.5ml
- If the total protein is < 5mg/dl please add two drops of serum from the same patient or another patient of the same species (if protein measurement is unavailable, add 2 drops of serum regardless)

Reporting and pricing schedule

- Samples will be processed on the day they are received or the next day to determine the cell count and cell viability and perform flow cytometry
- *Note: If a sample has too few cells or the cell viability is too low, flow cytometry results will not be accurate and flow cytometry will not be performed.*
 - *A fee of \$15 may be charged for the initial sample assessment in these cases*
- **Pricing Schedule**
 - **Canine & Feline Flow Cytometry \$115.00**
- **Reporting**
 - Reports will be generated within approximately 3 business days of sample processing

Questions?

Email: vetclinfo@uga.edu

Phone: (706)542-9430

Alternate phone: (706)542-5161