INTRODUCTORY RESIDENT ROTATION IN HEMATOLOGY
HEMATOPATHOLOGY

Introduction

The basic residency rotation in Hematology-Hematopathology is designed to expose the resident to the
diverse multidisciplinary diagnostic approach utilized in this specialty. The objectives for this rotation are:

1. To permit the resident to attain expertise in the methodological, diagnostic and scientific aspects
   of Hematology-Hematopathology.
2. To permit the resident to integrate the diagnostic test results, and arrive at the proper diagnostic
   conclusions necessary for diagnosis of hematologic disorders.
3. To permit the resident to develop the professional and managerial skills necessary to function as
   a Hematopathologist, including investigative and administrative roles. The resident should
   become familiar with methods of quality control and quality assurance relevant to Hematology.

Guidelines for the Hematology-Hematopathology Residency Rotation

I. Education Goals

The resident will be expected to demonstrate acceptable progress in the following areas by the end of the
initial 3 month rotation:

1. Obtain a clinical history sufficient for hematopathologic diagnosis, including prior relevant diagnostic
   evaluations.
2. Determine a provisional hematologic diagnosis with the appropriate choice of one or more of the
diagnostic methods summarized in section “c” below. The resident will be introduced to the
management skills necessary to ensure the proper performance of special diagnostic tests by the
Hematology staff.
3. Understand the characteristic diagnostic features of the peripheral smear, bone marrow biopsy and
   aspirate. The hematology resident will be expected to understand the general diagnostic features of
cytochemical stains, immunohistochemical stains, flow cytometry, cytogenetics, and coagulation-
related tests. S/he will become familiar with the molecular biologic diagnostic approach to
hematologic disorders including the benign hematologic disorders, such as the hemoglobinopathies
and anemias, as well as the hematologic malignancies.
4. Integrate the results of the multiple diagnostic tests into an appropriate hemato(path)logic diagnosis.
5. Inform the requesting clinician of the diagnosis in an appropriate concise manner; and suggest follow-
up diagnostic tests if necessary.

II. Responsibilities

The Hematopathology resident is expected to organize and coordinate the interpretations of all hematologic
diagnostic tests performed on the Hematopathology service in a timely fashion.

Daily Duties (Monday-Friday)

8-8:30 AM
a. Organize review peripheral smears submitted for pathologist review by the Hematology
   technologists.
b. Organize patient bone marrow aspirate specimens/peripheral smears/BM biopsies/special
   stains/flow cytometric data for pathologist review. The folder with all prior HemePath reports on that
   patient should be pulled for review.
c. The resident should quickly compare prior diagnoses with the current diagnostic material, to ensure
   consistency.
d. The resident will page the HemePath attending when the marrows are ready for review.

Other duties:
On Tuesdays and Wednesdays the histology technologist is expected to beep the resident when the biopsy specimen slides are ready. On these days the resident should contact Histology if the biopsy specimens do not arrive by 9:00 am.

1-2 PM
Obtain from the Medical Transcriptionist the bone marrow priority list, which should include all patients who do not have adequate aspirate specimens.

3-4 PM
a. The resident will contact the Histology supervisor no later than 3:30 PM, and give her a priority list for bone marrow biopsy processing.
b. The resident will quickly review all new bone marrow aspirates for the day, and present for review any new acute leukemias, peripheralizing myelomas (i.e. any cases that need special evaluation before the bone marrow biopsy is ready for review. S/he will be responsible for contacting the Hematopathologist attending so that appropriate flow cytometric, immunohistochemical, cytochemical, etc. procedures can be initiated on the same day the aspirate is obtained.
c. Lymphoma cases of any kind should be briefly reviewed. The resident should ask the Medical Transcriptionist to obtain the original diagnostic histologic sections from Pathology the day before review. The resident is expected to identify the specific specimens needed.
d. My unusual cases, e.g. erythroleukemias, immunoblastic sarcomas, etc, should be briefly examined, and previous biopsy/aspirate specimens should be obtained for review the next day for review by the attending Hematopathologist.

Important point: The teaching process and the diagnostic process will be greatly facilitated by a conscientious resident who obtains previous material for review, and organizes each case appropriately. All attendings will be expected to use interesting cases as a basis for resident teaching when they become available. Efficacious signout will encourage this activity.

3. Training Schedule/Training Objectives:
The resident will rotate through the following areas of the Hematology laboratory and learn the Principles, operation, and applications of the equipment used. This will include "hands on" experience with each major hematology instrument listed below.

This experience is necessary so that the resident will know how much time is required for these tests, and what potential problems might arise. Mutually convenient times for the training will be arranged with technologist personnel. Total time spent in each area will depend on the resident's interest and rate of progress. Residents will review on a daily basis all material referred to the professional staff at mutually convenient times.

a. Coulter Stk-S
   Know how the Coulter STK-S works, and how to determine the clinical significance of the histograms.
b. Manual Routine Hematology:
   Know how to perform and interpret: Manual differential smear count
c. Biochemical Hematology:
   Perform and interpret:
      Special cytochemical stains, myeloperoxidase, TRAP, LAP score
      Hemoglobin electrophoresis
      Screen for sickling hemoglobins
      Flow Cytometry
d. Urinalysis
   Urine sediment analysis
   Dipstick screening
e. **Coagulation**
   The Blood Bank resident is expected to be primarily responsible for a coagulation, however, the Hematology resident is expected to act as a backup. 
   **Screening tests:** PT, PTT, Fibrinogen, mixing studies for inhibitors versus deficiencies, D-Dimer for DIC. 
   **Specialty tests** (*A Hospital): Factor assays, platelet aggregation studies 
   VWD work-up 
   Lupus, other inhibitor work-ups 

Coagulation studies will be performed in collaboration with the special coagulation lab at the VA hospital, supervised by Dr. Fink

f. **Bone Marrow Interpretation**
   Become familiar with the techniques used for bone marrow aspirates and biopsies, aspirate and biopsy specimen preparation.

   Actual performance of BM biopsies will generally be during the flow cytometric rotation.

5. **Available Instrumentation in Hematology:**

   Coulter STK-S whole blood analyzers (2)  
   Platelet aggregometer (VA Hospital)  
   Hemoglobin electrophoresis cells (molecular diagnostics lab)  
   Hemocytometer  
   Cytocentrifuge  
   Ames Clinitek Urinalysis machine  
   Refractometer  
   Fibrometer (VA)  
   Viscosimeter  
   Automatic peripheral smear stainer  
   MLA 700 Coagulometer (2)  
   Microhematocrit centrifuge  
   Microscopes: Routine  
   Polarizing  
   dual-headed  
   6-headed  
   Photomicroscope

**SPECIAL LEARNING OBJECTIVES**

I. **Routine Hematology**

A Coulter

The resident will be given an introductory hands-on demonstration of the principles of the Coulter STK-S. Didactic instruction will includes practical sessions with the instrument, as well as a lecture.

The residents) will be able to:

1. State the Coulter principles.; DC; RF impedance, Laser scattering; and what features of the cells are detected with each method.
2. Process an actual patient sample, and detect problems with processing of patient blood samples; suggest solutions to correct the problem (i.e special procedures for clumped platelets, clumped RBC’s etc.).
3. Explain the diagnostic features of the Coulter histograms.
4. Deduce the clinical diagnosis based on the Coulter 5 part white cell histogram, red cell histograms, and know limitations of the Coulter technique.
5. State the directly IV measured and indirectly measured parameters, and explain how each parameter is derived; compare these results to manual methods of performing a CBC.
6. State the final dilutions for the WBC and RBC counts, platelets, Hbg, Hct.
7. Discuss platelet counting with the STK-S
8. Make acceptable blood smears for differentials.
10. Describe the flow of the specimen from aspiration to printout of results.

B. Microscopic Examination of Peripheral Blood Smears

1. State the normal differential values (age-adjusted), and describe what cell types are commonly seen, and/or the diagnostic significance of cell types not commonly seen.
2. Properly prepare peripheral smears, Understand the principle of Wright-Giemsa staining and the principle of the automatic stainer.
3. Perform leukocyte differentials on normal and abnormal smears.
4. Know the terminology of red blood cell morphology and clinical application and its relation to the reticulocyte count, MCV, etc.
5. Be able to correlate platelet counts, WBC counts, and RBC indices with a manual smear evaluation.
6.

C. Reticulocyte Count

1. Perform retic count from receipt of specimen to calculation of absolute retic Count
2. State proper methods of specimen collection, anticoagulant of choice, and time limitations.
3. Perform proper calculations in reticulocyte percentage and absolute and absolute values.
4. State normal values for adults, children and the reason for a decrease in retics.
5. List three reasons for an increase or decrease in retics.
6. Perform retic count from receipt of specimen to calculation of absolute retic count.

D. Urinalysis

1. Be familiar with the principle and operation of the Clinitek, including daily calibration and maintenance.
2. Review carefully:
   (1) A Handbook of Routine Urinalysis [Sister Laurine Graft]
   (2) Modern Urine Chemistry [Ames]
   (3) Urinalysis Section, Todd & Sanford
   (4) Lab Manual
3. Be thoroughly familiar with the routine urinalysis procedure. This includes color and clarity, specific gravity, all chemistries on the dipstix, any back-up tests, and a microscopic examination.
4. Know the reagents, procedure, type of specimen, normal values, and clinical significance of each urinalysis procedure (see, Urinalysis Manual).
5. Be able to correlate chemical and microscopic urinary findings with certain of the more common kidney and urinary tract disease states.
6. Know the effects of aging of specimens on the different parameters of the routine urinalysis.
7. Be familiar with quality control and instrument maintenance procedures. Know the internal and external quality controls that are utilized by this lab.
8. Be familiar with the general physiology of the kidney and the rest of the urinary tract.

E. Hemostasis (Routine-UAMS, Special-VA)

1. State the basic theory of the coagulation process and fibrinolysis.
2. List the coagulation factors best tested by: PT, PTT, TT, fibrinogen, etc. used for each one.
3. State the proper procedure for quality assurance of instruments and test procedures. Be able to tell what quality control is testing.
4. State the type of specimen required for the various tests, time limitations, methods of collection,
aging effects, types of anticoagulants and their proper anticoagulant plasma ratio.

5. State the theory, perform, and interpret results off all routine tests used in the lab (PT, APU, U, Fibrinogen). Be familiar with the principles of these tests done on the MLA7OO and ACA.

6. State which tests are commonly used to monitor heparin therapy.

7. State the purpose of mixing studies, reagents used and what each reagent contains. Be able to choose appropriate (specialty) diagnostic tests based on the mixing studies.

8. State the theory of the factor assays utilizing deficient plasmas of synthetic substances. (Special-VA)

9. Name, state principle of, and interpret the D-Dimer test used to diagnostic DIC.

10. State the theory behind euglobin lysis test.

11. State the principle and interpret results of the platelet aggregation tests, including ATP release, B-Thromboglobulin and PFA assays. (Special-VA).

12. State the indications for measurement of Anti-thrombin III, factors S and C, antithrombin-, and tissue plasminogen activator.

13. Relate coagulation laboratory data with disease states (such as liver disease, Vitamin K deficiency, hereditary factor deficiencies, thrombocytopenic purpura, etc.)


II. Special, Biochemical Hematology

A. Special stains: Cytochemical and immunohistochemical

The resident should be able to:
1. Explain the purpose of Cytochemical and immunohistochemical stains, and how to interpret appropriate controls.
2. State types of suitable specimens received
3. State the controls used for each tests(s) and why
4. State reason for doing an iron stain.
5. Perform a myeloperoxidase, non specific esterase, and manual Wright's stains.
6. Interpret T and B cell immuno stains.
7. List the types of leukemias and lymphomas and how to distinguish between them using these stains.
8. Perform a LAP Score.

B. Bone Marrow

1. Know how to perform a posterior iliac crest bone marrow biopsy/aspirate. The resident will be expected to perform at least 20 bone marrow procedures, usually during the flow cytometry rotation.
2. Understand various preparations used in bone marrow examination.
3. Review bone marrow aspirates, clot sections, biopsies, and peripheral blood smears.
4. Identify, classify, and diagnose malignant and benign hematologic disorders. Numerous textbook articles are available for ready review. The resident is expected to review this material, and discuss questions with the attending.

Additional Comments with regard to interpretation of bone marrow specimens and ability to diagnose hematologic disorders will be included in the didactic material presented by the professional staff of the division.

C. Hematology Development Area

Immunohistochemical stains on aspirate material.

D. Computer

All residents will be instructed in the use of the Soft computer system, to:
1. Query patient results
2. Request diagnostic tests
   The hematology, urinalysis, special stains, and coagulation manuals are available for individual tests done in the section.
III. Flow Cytometry

The Hematology resident in training will be expected to become familiar with the diagnostic tests available in flow cytometry, specifically:

1. Immunophenotypic analysis of hematopoietic cells: namely markers of myeloid, monocytoid, B lymphoid, T lymphoid cell differentiation.
2. How to choose appropriate samples for flow cytometry, ie criteria for mutability of peripheral blood, bone marrow, lymph node material, etc.
3. Criteria for separating benign hematopoietic proliferations from malignant ones.

A formal rotation in flow cytometry is available.

IV. Cytogenetics

The resident in training is expected to familiarize himself/herself with the diagnostic value of cytogenetics in hematologic diagnosis. Specific topics include knowledge of the characteristic chromosomal arrangements known to be associated with:

   i) Acute nonlymphoid and lymphoid
   ii) Myeloproliferative disorders- the Philadelphia chromosome
   iii) Myelodysplasias
   iv) The lymphomas
   v) The chronic leukemias

A formal rotation in cytogenetics is available

Summary - the resident as a Hematology Consultant

The Hematology pathologist in training should develop the skills necessary to act as a clinical consultant in every aspect of Hematopathology. The pathologist may wish to interview the patient, study available clinical notes, suggest additional specimens and germane laboratory investigation. The subsequent consultative report should include a positive interpretive opinion with continual review during the patient's clinical course. Follow up in morning report is essential for achieving a maximum learning benefit from the rotation, and is of especial value to the other residents.

Reading list

Bone Marrow Procedure / Interpretation


Introduction to Flow Cytometric analysis


Diagnosis of Acute Leukemias, Myelodysplasia, Myeloproliferative disorders.
Tumors of the Bone Marrow. Brunning and McKenna. AFIP. 1994.

Diagnosis of the Lymphomas

Automated Cell Analyzers, (Coulter Stack-S)


"Spurious Results from Automated Hematology Cell Counters" Joanne Cornbleet, Laboratory Medicine, 14:509514(1983).


Coagulation

Multiple Myeloma


OTHER SUGGESTED REFERENCES

GENERAL:

URINALYSIS:

BODY FLUIDS:

COAGULATION:

NAMES AND NUMBERS:
Laboratory Director: James T. Flick, M.D., Ph.D.
Ext. 67015
Pager 688-6778

Hematology Section supervisor: Sue Scott, MT (ASCP)
Ext. 66354
ADVANCED RESIDENT ROTATION IN HEMATOLOGY

Introduction

This advanced residency rotation in Hematopathology is designed to expand the knowledge base beyond that offered in the primary Hematology rotation. The objectives are essentially the same as with the introductory rotation, except that the focus will be on an in depth review of the diagnostic concepts specific to Hematopathology. Senior residents who show a good working knowledge of Hematopathology will be given considerable freedom to interpret cases, suggest appropriate laboratory diagnostic tests: and interact with the clinicians. The Hematopathology attending may act as a consultant to suitably experienced residents.

The objectives for the advanced residency rotation are:

1. To permit the further development of expertise in the methodological, diagnostic and scientific aspects of Hematopathology. At the completion of the rotation: the resident will be expected to have a practical working knowledge of all the appropriate diagnostic tests: and their limitations.

2. Further development of the professional and managerial skills necessary to function as a Hematopathologist, including investigative and administrative roles. The senior resident will be expected to show initiative with the Hematology and Flow cytometry staff in requesting appropriate diagnostic test, and will be expected to understand methods of quality control and quality assurance relevant to Hematology.

3. At the end of the rotation, the resident will be expected to be at a knowledge level sufficient for independent Hematologic pathologic diagnosis. The senior Hematology resident should be able to function independently, know his/her limitations, and know when he/she should request specialty consultation in Hematopathology.

I. Education Goals

The resident will be expected to demonstrate proficiency in the following areas:

a. Independently obtain a clinical history sufficient for provisional hematopathologic diagnosis from the consulting clinician, including prior relevant diagnostic evaluations.

b. Independently determine a provisional hematologic diagnosis.

c. Choose the appropriate choice of Hematologic diagnostic test or test battery

d. Integrate the results of multiple diagnostic tests into an appropriate hemato(path)logic diagnosis.

e. Inform the requesting clinician of the diagnosis in an appropriate concise manner; suggest follow-up diagnostic tests if necessary.

II. Responsibilities

The senior Hematopathology resident is expected to organize and coordinate the interpretations of all hematologic diagnostic tests performed on the Hematopathology service in a timely fashion. He/she will be expected to instruct any junior residents on rotation, and transfer some responsibilities in an appropriate fashion.

Daily Duties (Monday-Friday)

Early AM

i. organize review peripheral smears and body fluid specimens submitted for pathologist review by the Hematology technologists.

ii. organize patient bone marrow aspirate specimens/peripheral smears/BM biopsies/special stains/flow cytometric data for pathologist review. The folder with all prior Hemepath reports on
that patient should be pulled for review.

iii. The resident should quickly compare prior diagnoses with the current diagnostic material, to ensure consistency.

iv. The resident will page the Hemepath attending when the marrows are ready for review.

On Tuesdays and Wednesdays the histology technologist is expected to beep the resident when the biopsy specimen slides are ready. On these days the resident should contact Histology if the biopsy specimens do not arrive by 9:00 am.

1-2 PM

Obtain from the Medical Transcriptionist the bone marrow priority list, is to include all patients who do not have adequate aspirate specimens.

3-4 PM

a. The resident will contact the Histology supervisor no later than 3:30 PM, and give her a priority list for bone marrow biopsy processing.

b. The resident will quickly review all new bone marrow aspirates for the day, and present for review any new acute leukemias, peripheralizing myelomas (i.e. any cases that need special evaluation before the bone marrow biopsy is ready for review. S/he will be responsible for contacting the Hematopathologist attending so that appropriate flow cytometric, immunohistochemical, cytochemical, etc. procedures can be initiated on the same day the aspirate is obtained.

c. Lymphoma cases of any kind should be briefly reviewed. The resident should ask the Medical Transcriptionist to obtain the original diagnostic histologic sections from Pathology the day before review. The resident is expected to identify the specific specimens needed.

d. My unusual cases, e.g. erythroleukemias, immunoblastic sarcomas, etc. should be briefly examined, and previous biopsy/aspirate specimens should be obtained for review the next day for review by the attending Hematopathologist

Important point:

The teaching process and the diagnostic process will be greatly facilitated by a conscientious resident who obtains previous material for review, and organizes each case appropriately. All attendings will be expected to use interesting cases as a basis for resident teaching when they become available and efficacious signout will encourage this activity.

Training Schedule/Training Objectives:

The senior Hematology resident will not have a formal rotation through the Hematology laboratory, however, he/she will be expected to review a good working knowledge of the principles, methods of operation, and applications of the Hematology and flow cytometry equipment. The resident should review the training objectives of the introductory hematology and flow cytometry rotation "Hands on" experience will be offered if necessary, or if requested.

NAMES AND NUMBERS:

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Hematology Section supervisor: Sue Scott, MT (ASCP)
Ext. 66354
Daily Hematology Resident Responsibilities

7:30AM - 8:30AM - Review Critical value log, abnormal log, invalids/delta checks, -body-fluid specimens, coagulation book. Ask about any new Hematologic cases.

8:30AM Begin marrow readout [biopsy slides arrive]. Call Many Finger (66536) if BM slides do not arrive by 8:30.

8:30AM Attend morning report (second Heme resident). On busy days, a single Hematology resident will be expected to sign out marrows, but must check in with on-call resident

10:30AM Begin path signout with the Hematology/pathology attending when several marrow reports are ready for pathologist review.

You Should.

i.) Readout Priority marrows first (as determined by yesterdays marrow schedule). This minimizes interruptions by the Oncology staff. You can expect this to begin by 10-11AM.

ii.) Obtain diagnostic material - ie., LN specimens, etc., on all lymphomas, and all relapsed/new leukemias. [the medical transcriptionist will assist if requested the day before signout].

iii.) Make sure BM report folder has:
SP report summary [double check dx].
CBC/lab summary [manual differential?]

iv.) Pull patient data from folder/Hemepath database if dx complex or unresolved-

3:00PM Review newly stained marrow aspirate smears with the bone marrow Coordinator. Make sure that he/she has called back the "low spic" list.

3:00PM - 5:00PM Other schedules hematology activities (teaching sessions, demonstrations, etc.), depending on workload, staffing, and special studies. These activities win be coordinated by Dr. Flick

4:00PM - Final Checkout on Friday - Unresolved cases, new acute leukemias, new body fluids, etc. should be reviewed with the attending. The attending pathologist is responsible for all new cases that arrive before 5:00 pm, and must triage. After 5:00pm, it is the responsibility of the on call resident and attending.

5:00PM- Signout with on call resident - Hot or pending cases It is your responsibility to contact the on call resident if he/she does not show up.

[you should resolve all major cases before leaving, such as ordering flow cytometry, special stains, etc., and accelerated review with the Hematology-pathology attending. These are usually the most educational cases, since you will have an opportunity to decide how to perform the diagnosis.]

I. Feed Hematology/Pathology study set. All interesting cases including lymphomas should be saved for later slide review by pathology residents. [if you don't save the diagnostic slides, you won't have a good study set].

II. Unusual Coagulation Requests After identifying problem, relay all coagulation screening requests to the Blood Bank resident. However, you must be familiar with the outcome of the test results.

Example: 50/50 mix request/results on a patient with elevated PT or PTT. [if you neglect to inform the Blood Bank resident, and the problem is unresolved, it is your problem].
Exception: You will be expected to sign bleeding time request paperwork. Abnormal results should be relayed to the BB resident, as they may be important for decisions on blood component therapy.

As you become more experienced, and complete more of the organizational work - including dictation, the Hematology-Pathology attending will be expected to respond with a greater teaching effort. If you put more effort into this rotation, you will get more out of it!

Hematology Pathology Fellowship

Response to: General Competencies as defined by the ACGME, to be implemented July 2001

1. The trainee will be expected to demonstrate compassionate, appropriate and effective patient care. The hemepath resident/fellow is expected to have a therapeutic apheresis exposure, as well as exposure to clinical coagulation, both of which involve patient-doctor interactions. Both of these services are expected to evaluate the clinical performance of the resident/fellow, as well as report the performance to the Hemepath Program Director.

2. The trainee will be expected to demonstrate medical knowledge, especially with regards to established and evolving biomedical, clinical and cognate sciences and to demonstrate the application of this knowledge to patient care.

   The trainee is expected to show increased responsibility in a graduated fashion over the training period, and a progressive ability to handle complex clinical cases, which include interaction with CP clinical staff at morning report, during bone marrow signout, and during conversations with the clinical oncologists. The resident/fellow is also expected to present cases at the joint Hemepath/Hemeonc conferences, namely at the Tuesday morning and Thursday morning conferences. This performance is evaluated at three times during the training period. First, during the 3 month "initiation" where the fellow is evaluated by the Hemepath Bone Marrow attendings, specifically for the ability of the fellow to "take charge of the Hemepath Service" and instruct the residents, and in the mid-year and summary composite evaluations.

3. Trainees will be expected to demonstrate practice-based learning and improvement involving investigation and evaluation of their own patient care, the assimilation of scientific evidence and demonstrated evidence in improvement in patient care.

   The Hemepath service is not primarily a practice-based service, so this area is not a major focus of the program. However, the resident/fellow is expected to present his/her interesting patient cases of the day in a progressively more sophisticated manner during CP morning report, as well as show progressively more sophisticated expertise in evaluation and diagnosing patient problems in the clinical rotations: clinical coagulation and therapeutic apheresis.

4. Interpersonal and communication skills will be assessed in order to assure effective information exchange with patients, their families and other health professionals.

   The resident/fellow is expected to contact and otherwise interact with physicians regarding Hemepath diagnoses while on the Hemepath service. This performance is monitored by the hemepath attendings, who evaluate the resident/fellow biannually. Evaluation of the development of these skills is also made on the clinical coagulation and therapeutic apheresis services, where direct interaction with patient and family members is routine.

5. Trainees will be expected to manifest a commitment to carrying out professional responsibilities, adherence to ethical principles, and sensitivity to a diverse patient population in order to assure professionalism. This commitment is monitored by the BM Hemepath attendings, as well as by the clinical attendings responsible for the associate on the therapeutic apheresis and clinical coagulation services. Problems in this area are reported by the relevant attending staff at the time of the biannual
6. Trainees will be expected to demonstrate actions that demonstrate an awareness of and responsiveness to the larger context and system of health care and the ability to effectively call on system resources to provide care that is of optimal value in order to assure an understanding of the systems-based practice of medicine.

The resident/fellow is expected to demonstrate a progressive, graduated level of expertise in fully integrating the resources available to him/her. This includes experience in direct morphologic diagnosis as well as independently initiated literature searches in relevant cases, with the specific goal being the development of a broad-based clinical hematopathologic background that draws on a self-learning/self-starting process, as well as knowing which attending specialist to ask when new, complex problems arise. A minimum requirement in this goal is a demonstrated knowledge in how to access and search computer-based databases, both on the internet and in the lab. Evaluation of the progress of the resident/fellow is integrated into their biannual reports.

**Flow Cytometry Rotation**

**Introduction**

This advanced residency rotation is designed as an intensive exposure to the diagnostic applications of flow cytometry. The Flow resident is expected to act as a consultant, and interact closely with the Hematology resident and attending. This rotation may be offered in coordination with immunology or molecular diagnostics rotations.

**Education Goals**

The resident will be expected to demonstrate proficiency in the following areas:

1. Obtain a clinical history, preliminary morphologic evaluation (if possible) before processing of flow cytometric material, and make a provisional hematopathologic diagnosis.
2. Interpret partially processed cytospin cell samples for specimen adequacy.
3. Interpret small angle and large angle laser scatter histograms of cells to determine appropriate location of gating windows to ensure proper cell choice.
4. Interpret single and dual immunofluorescence histograms, and know how the thresholds for positive and negative immunofluorescence are determined.
5. Interpret three color immunofluorescence to determine appropriate location of gating
6. Know how to interpret classic immunofluorescent patterns for:
   a. AML - myeloid, monocytic, megakaryocytic, erythroid differentiation - morphologic correlation.
   b. ALL - patterns for pre-B vs mature B, etc. and the morphologic correlation.
   c. Non-ALL lymphoid malignancies - diagnostic criteria for diagnosis of B-cell and T-cell lymphomas, hairy cell leukemia, myeloma, etc.
7. Interpret immune status panels
8. Interpret high resolution DNA histograms for solid tissue tumors
9. Interpret CD3 results for OKT3 therapy patients.
10. Interpret CD34 results for bone marrow transplant patients.
11. Interpret DNA/cytoplasmic immunoglobulin histograms for myeloma patients.

**Responsibilities:**

The flow cytometry resident is expected to review clinical information and cytospin slides of analyzed material. Data is to be interpreted and presented in a timely fashion to the Flow Cytometry or Hematology attending. He/she is expected to develop skills as a consultant in flow cytometry, and ultimately interact independently with the requesting physicians, offering preliminary flow cytometric interpretations.

The flow cytometry resident is expected to perform 25 bone marrow aspirate and biopsy procedures while on rotation in flow cytometry, as clinical duties permit a Diagnostic problems with flow cytometric evaluations take priority. However, time for the procedures 15 usually available. The iliac crest marrow procedures are to be performed under the supervision of the senior bone marrow technologist, until he/she deems the resident
ready for independent activity. However, all bone marrows must be performed with the full knowledge of the senior bone marrow technologist, and with an experience marrow assistant. Male residents must always be accompanied by a female assistant if the marrow procedure is to be done on a female. If the resident documents completion of the 25 bone marrow procedures, they are invited to return as a senior resident to perform additional bone marrows before completion of residency.

Sternal marrow aspirates are not permitted, either with or without bone marrow technologist supervision, unless an experienced pathologist or Hematologist/Oncologist is present

**Daily Duties**

1. The flow cytometry resident should check in daily at approximately 9:00AM with the flow cytometry technologists.

2. The flow resident should be available if clinical or morphologic information is necessary before flow cytometric Processing of clinical material. He/she is expected to expedite the handing of cases submitted to the Flow lab that are "pending" evaluation of morphology. He/she is also expected to review all diagnostic material on bone marrow specimens with the Hematology attending and non-bone marrow specimens with the Flow cytometry attending.

3. The flow resident will also be expected to review surgical material when high resolution DNA analysis is requested. Review and analysis will be discussed with the flow cytometry attending.

4. Review cases at Hemepath signout to ensure Consistency of diagnosis with flow cytometric material, and to determine if farther diagnostic material should be obtained:
   a. Review clg/DNA data on myeloma patients at marrow signout.
   b. Confirm Consistency of flow cytometric report with clinical/morphologic data.
   c. Determine whether flow cytometric request was appropriate.
   d. Review cases that require specialized "Work-ups" or panels with the appropriate attending as they are received.

Important point: The teaching process and the diagnostic process in Hematopathology will be greatly facilitated by an organized and conscientious resident, who presents the flow cytometric data appropriately. All attendings will be expected to use interesting cases as a basis for resident teaching when they become available, and efficacious signout will encourage this activity.

**Training Schedule/Training Objectives**
The resident will rotate through the different areas of flow cytometry, so that he/she will become familiar with the principle, operation, and applications of the equipment used. This will include hands-on experience. This experience is necessary for two reasons: to make sure the resident knows 1) the strengths and weaknesses of the different methods, and 2) the time necessary for the test to be performed. Mutually convenient times for the framing will be arranged with the technologist personnel. Total time spent in each area will depend on the resident's interest and rate of progress.

**B-D Flow Cytometer**

a. General principles of operation
   - The sheathed flow chamber
   - Laser beam for cell type identification
   - Laser beam for immunofluorescence
   - Cell sorting principles

b. Immunofluorescent techniques
   - Methods of cell preparation and immunofluorescence staining
   - Use of antibody panels to determine cell differentiation
   - Setting gates to determine specific cell population results
   - Adjustment of fluorescent intensity thresholds
   - Interpretation of single and dual fluorescence histograms

c. DNA analysis
Specific Learning Objectives

The resident will be given an introductory hands-on demonstration of the principles of the Flow cytometer. Instruction will include practical sessions with the instrument, as well as didactic sessions.

Immunofluorescent techniques

The resident will be expected to:
1. State the general flow cytometric principles as they relate to immunophenotyping and DNA analysis.
2. State the different cellular features detected by forward and side light scatter, and predict where different cell types are most likely to be found in the laser scatter histogram.
3. Describe the method for determining the difference between positive and negative immunofluorescence.
4. Describe the diagnostic advantage of multiparameter immunofluorescence over single parameter immunofluorescence.
5. Know the diagnostic criteria for identifying the different types of acute and chronic leukemias.
6. Know the difference between biclonal and biphenotypic
7. Know the diagnostic pitfalls of the laser Scatter histogram, and the value of morphology obtained from a cytospin of partially purified cells.
8. Know how to diagnose B and T-cell lymphomas, and multiple myeloma.
9. Know the basics of instrument and specimen quality Control, i.e. how many cells are required; presence of nonspecific staining; poor instrument compensation, etc.

DNA Ploidy techniques

1. Know what is measured and how to determine GO/G1, S and G2M peaks.
2. Appreciate the significance of an increased S phase in solid tumor malignancies
3. Know how to identify an aneuploid peak, its significance, and how this relates to cytogenetic findings.
4. Know how the baseline diploid DNA peak is identified for fresh tissue versus formalin fixed paraffin tissue.

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Reading list for flow cytometry rotation


Borowitz MJ Guenther KL, Shults KE, Stelzer GT. Immunophenotyping of acute leukemia by flow cytometric analysis. Use of CD45 and right-angle light scatter to gate on leukemic blasts in three-color analysis. Am J


