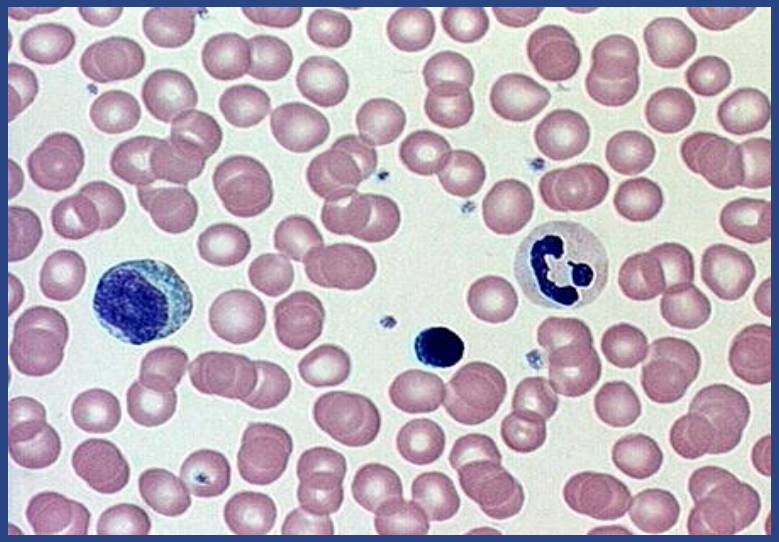
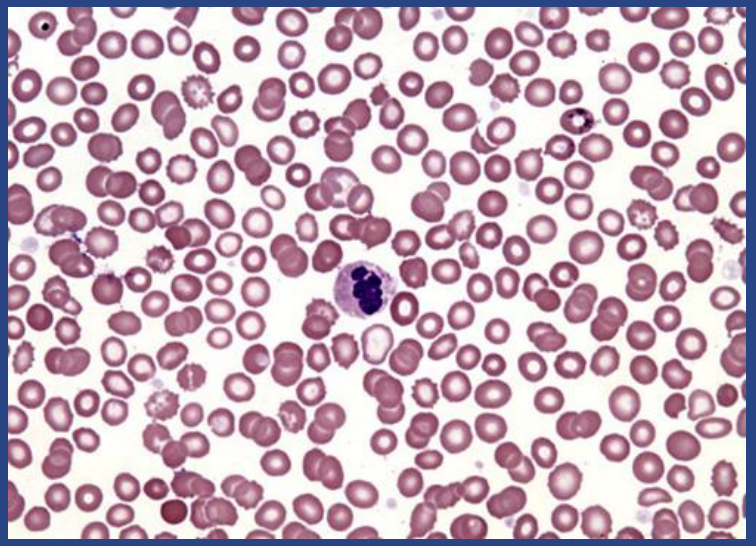
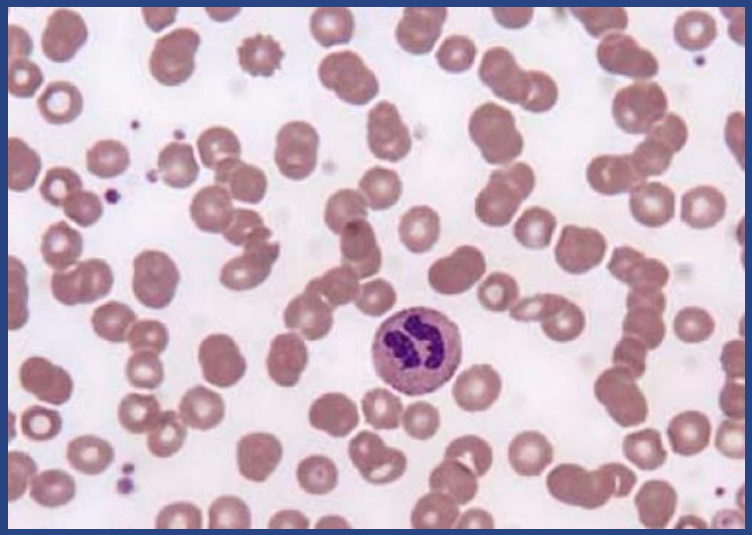




Evaluation of the Anemic Patient



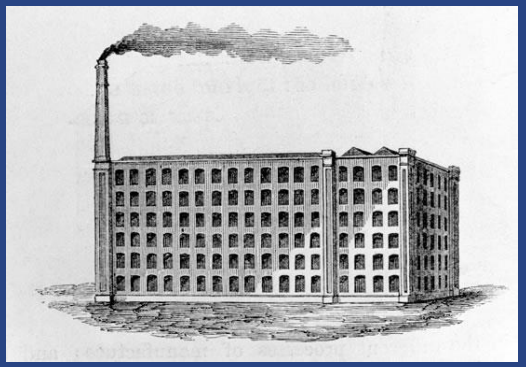
Brenda Shinar, MD, FACP

September 13, 2022

Objectives:

- Understand the basics of hematopoiesis.
- Define anemia and understand how to classify it based on the kinetic model and the morphologic model.
- Understand how to calculate the absolute reticulocyte count and absolute reticulocyte index and what these values mean.
- Understand the differentials of microcytic, normocytic, and macrocytic anemias and how to appropriately evaluate them to make a diagnosis.

Red Blood Cell Factory: What is Required?



- Work space
- Task master
- Building blocks
- Instructions
- Distribution
- Survival

- Bone marrow
- Erythropoietin
- Precursor cell, heme, iron, B12, folate
- Cytokines
- Exit out of bone marrow
- No lysis, sequestration, or loss

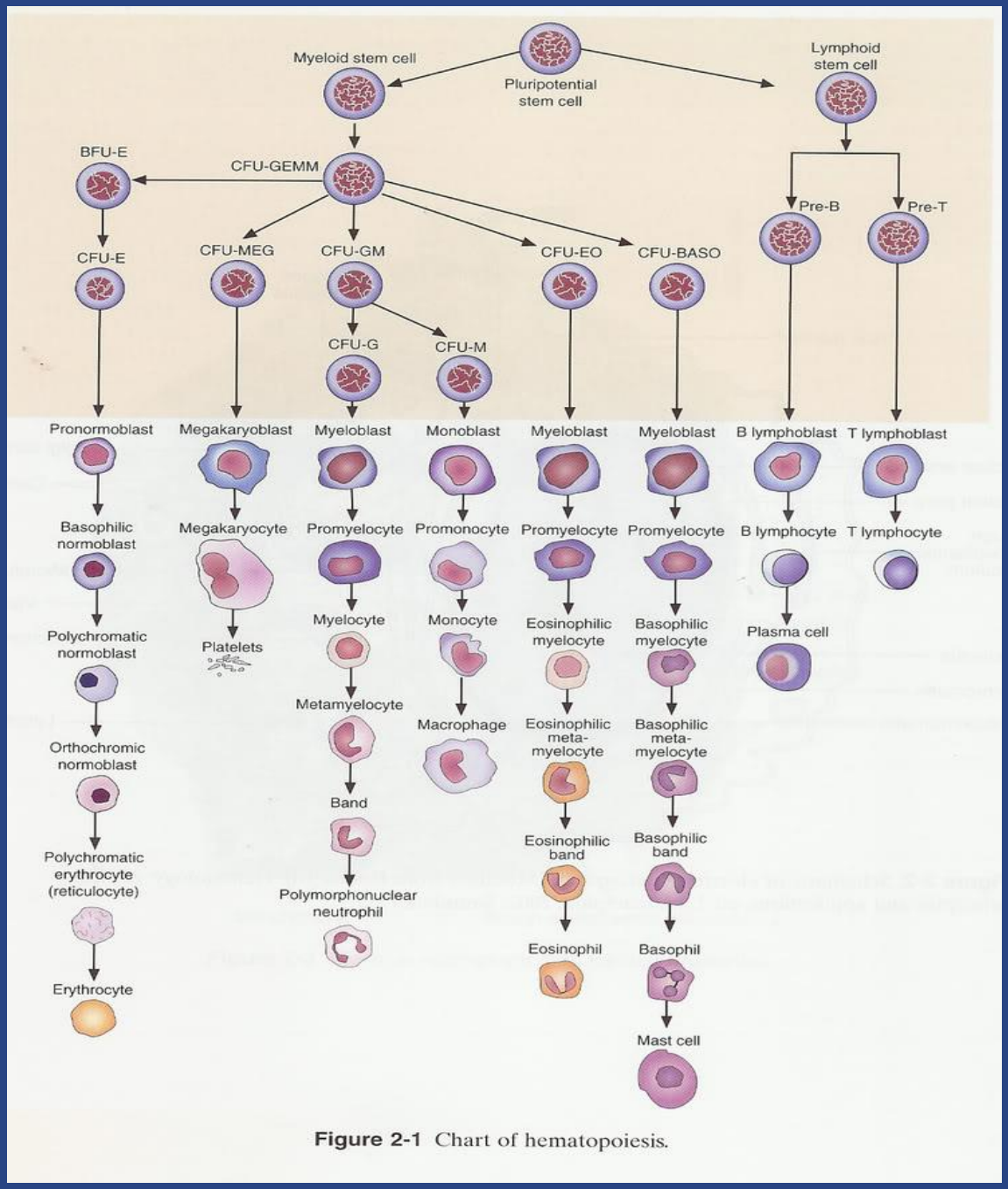
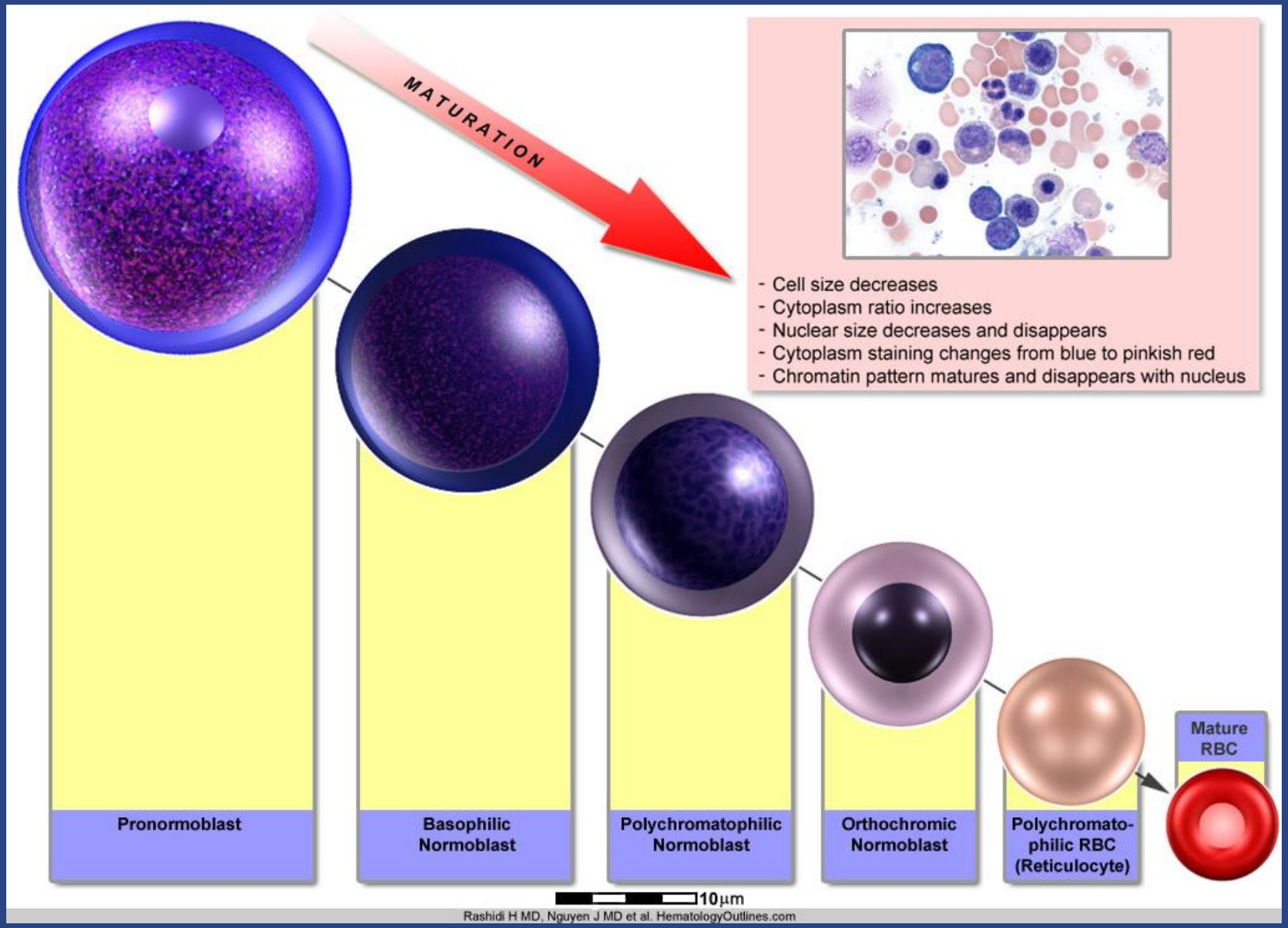


Figure 2-1 Chart of hematopoiesis.

The Red Cell Family Tree

The Red Cell Maturation: Benjamin Button



The Complete Blood Count (CBC)

- Hemoglobin (g/dL)
- Hematocrit (percent)

- Red Blood Cell Count (RBC) (million/mm3)

- Mean Cell Volume (MCV) (fL)
- Red Cell Distribution Width
  - Standard deviation of MCV/Mean MCV x 100
  - CV: coefficient variation
  - SD: standard deviation

- MCH: mean cell hemoglobin (pg)
- MCHC: mean cell hemoglobin concentration (g/dL)

LABORATORY	07/27/2015 09:10 MST	07/27/2015 05:18 MST
CBC		
WBC		10.6
RBC		2.03 (low)
HGB		6.4 (low)
HCT		18.9 (low)
MCV		93
MCH		31.5
MCHC		33.9
RDW-CV		18.6 (high)
RDW-SD		63.4 (high)
Platelet		65 (low)

Normal values for red blood cell parameters in men and women

Red cell parameter	Adult men		Adult women
Hemoglobin, g/dL	15.7 ± 1.7		13.8 ± 1.5
Hematocrit, percent	46.0 ± 4.0		40.0 ± 4.0
RBC count, million/µL	5.2 ± 0.7		4.6 ± 0.5
Reticulocytes, percent	1.6 ± 0.5		1.4 ± 0.5
Mean corpuscular volume, fL		88.0 ± 8.0	
Mean cell hemoglobin, pg/RBC		30.4 ± 2.8	
Mean cell hemoglobin concentration, g/dL of RBC		34.4 ± 1.1	
Red cell volume distribution width, percent (RDW)		13.1 ± 1.4	

Values are mean ± 2 standard deviations.
Adapted from Williams' Hematology, 6th ed, Beutler, E, Lichtman, MA, Coller, BS, et al (Eds), McGraw-Hill, New York, 2001.

Ratio of RBCs to Plasma
Determines Hgb, Hct, and RBC count

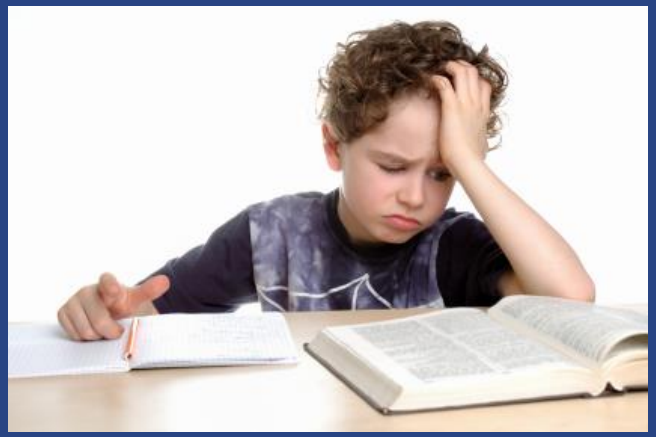
- *Acute severe bleeding* may cause orthostatic changes but gives falsely normal values of Hgb, Hct, and RBC count because the ratio of RBCs and plasma lost is the same
- *Pregnancy* increases plasma volume by 20-25% and therefore lowers the Hgb, Hct, and RBC count
- *Dehydration* decreases the plasma volume and artificially increases the Hgb, Hct, and RBC count

Kinetic Model:

How is the bone marrow responding to the anemia?

Pt Hct: 30% Normal Hct 45%

1) **Reticulocyte Count**= 2.8%
2) **Absolute Retic Count**=
   2.8 x pt's hct/nl hct=
   2.8 x 30/45 = 1.24
3) **Absolute Retic Index**=
   Absolute Retic Ct / Maturation factor
   1.24/1.5= 0.83



Maturation Factor corrects for the length of time a reticulocyte is in circulation so as not to count it twice or more. This factor varies with the degree of anemia.

HCT ≥ 35% = 1.0
25-35% = 1.5
20-25%= 2.0
< 20% =2.5

Evaluating the Anemic Patient:

Kinetic Model:

How the bone marrow is responding to the anemia

Decreased Production

Absolute Reticulocyte Index < 2

- Bone marrow disorders or suppression
- Lack of erythropoietin
- Lack of iron, B12, or folate
- Anemia of chronic disease

Increased Destruction or Blood Loss

Absolute Reticulocyte Index > 2

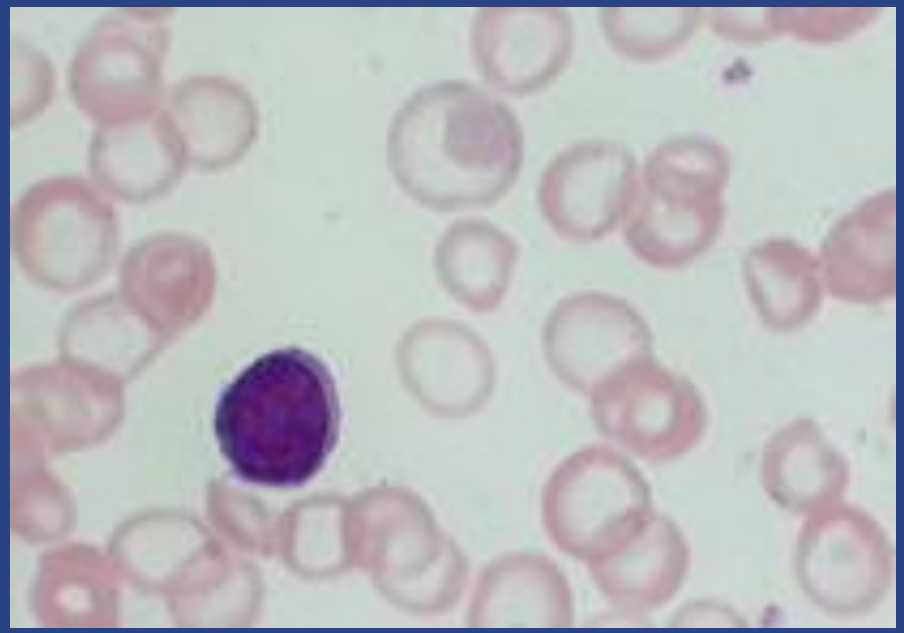
- Blood Loss
- Congenital Destruction
  - Membrane defects
  - Hemoglobinopathies
  - Enzyme deficiencies
- Acquired Destruction
  - Autoimmune HA
  - Microangiopathic HA
  - Infections (Malaria)
  - Paroxysmal Nocturnal Hemoglobinuria

Evaluating the Anemic Patient: The Morphologic Model

Microcytic Anemia :

MCV <80 fL

- Reduced iron availability
  - Iron deficiency
  - Anemia of chronic disease
- Reduced heme synthesis
  - Lead poisoning
  - Congenital or acquired sideroblastic anemia
- Reduced globin production
  - Thalassemias
  - Other hemoglobinopathies



Case 1

ID and CC:

22-year-old previously healthy white female with fatigue and dyspnea with exertion. She is on an OCP and has light, regular periods once a month. She has never been pregnant. In the ED, she has been given 2 units of pRBC and admitted to your service.

Labs:

WBC 5.0 g/dL

Hemoglobin 5.8 g/dL

Platelets 450K

MCV 70 fL

RDW 18 (11-16)

Which of the following is the most appropriate next step to evaluate the cause of her anemia?

A. Order ferritin, TIBC, and % sat
B. Order bone marrow biopsy
C. Order hemoglobin electrophoresis
D. None of the above are correct choices

> Transfusion. 1992 Oct;32(8):764-5. doi: 10.1046/j.1537-2995.1992.32893032107.x.

Transfusion

The effects of blood transfusion on serum ferritin, folic acid, and cobalamin levels

C H Ho [1]

Affiliations + expand

PMID: 1412686 DOI: 10.1046/j.1537-2995.1992.32893032107.x

Abstract

Thirty patients with anemia of various causes received packed red cell transfusions. To evaluate the influence of blood transfusion on the serum levels of different nutrients, serum ferritin, folate, and cobalamin levels were determined before and 2, 2 to 4, 5 to 7, 8 to 10, and 11 to 14 days after blood transfusion. No significant change was found in these levels before or at any time after blood transfusion. Blood transfusion exerted little effect on the serum levels of ferritin, cobalamin, or folate. Moreover, if the blood for testing was drawn 1 to 14 days after transfusion, nearly all cases of nutritional deficiency anemia could have been diagnosed without the influence of blood transfusion.

What are the positive predictors of GI lesions in iron deficient premenopausal women?

1. Symptoms of heartburn, regurgitation, or dyspepsia (OR 3.76 p=.002)
2. MCV < 70 fL (OR 1.88 p=.04
3. Hemoglobin < 10 g/dL (OR 1.7 p=.05)

NEGATIVE risk factor: Heavy menstrual blood loss

(OR 0.46 p=.002)

Nahon S, Lahmek P, Lesgourgues B, Nahon-Uzan K, Tuszynski T, Traissac L, Delas N. Predictive factors of GI lesions in 241 women with iron deficiency anemia. Am J Gastroenterol. 2002 Mar;97(3):590-3. doi: 10.1111/j.1572-0241.2002.05534.x. PMID: 11922551.

Case 2

- ID and CC:

  45-year-old man admitted with SIRS due to a UTI and chronic stage 4 sacral decub

- PMH

  Paraplegic due to GSW

  Noncompliance with urinary self-catheterization

  Sacral decubs

  Bipolar disorder

  Anemia

- Labs:

  Hgb 8.0 g/dL

  MCV 75 fL

  RDW 12.0 (11-16)

  Ferritin 250 ng/mL

  Transferrin 100 mg/dL (188-341 mg/dL)

  Percent saturation 8%

What is the most likely cause of his anemia?

A. High hepcidin, Anemia of Inflammation
B. Low hepcidin, Anemia of Inflammation
C. Iron deficiency anemia
D. Undiagnosed thalassemia

Laboratory Tests in Iron Deficiency of Increasing Severity

	Normal	Fe deficiency without anemia	Fe deficiency with mild anemia	Severe Fe deficiency with severe anemia
Marrow reticulo- endothelial iron	**2+ to 3+**	**None**	None	None
Serum iron, µg/dL	60 to 150	60 to 150	**<60**	<40
Iron binding capacity (transferrin), µg/dL	300 to 360	300 to 390	350 to 400	**>410**
Saturation (SI/TIBC), percent	20 to 50	30	**<15**	<10
Hemoglobin, g/dL	**Normal**	**Normal**	**9 to 12**	**6 to 7**
Red cell morphology	Normal	Normal	Normal or slight hypochromia	Hypochromia and microcytosis
Plasma or serum ferritin, ng/mL	40 to 200	**<40**	<20	<10

Serum ferritin ≤ 30 ng/dL = Iron deficient (PPV 83%, PLR= 11)
Serum ferritin ≥ 100 ng/dL = Iron sufficient (NLR .08)

What about if the ferritin is between 30 and 100?

Other Tests to help distinguish IDA from ACD…

- **Transferrin: (TRUCKS)**
  - High in IDA
  - Low or low normal in ACD

Retic %		1.4
Retic #		51
Immature Retic Fraction (IRF)		21.2 * H
Retic Hgb Equivalent [RET-He]		21.0 * L
Iron		23 L
Transferrin		185 L
Trans % Sat		9.8 L

- **Low reticulocyte-hemoglobin concentration (RET-He) increase with IV iron**
  - Does not distinguish between ACD and IDA but can improve in 2-3 days after IV iron supplementation to prove response to iron therapy

Distinguishing between ACD and IDA in NON-dialysis patients

- A ferritin of > 100 makes iron deficiency statistically unlikely even in patients with chronic inflammation!
- A transferrin level that is low gives further evidence of anemia of chronic disease.
- A low serum iron level and percent saturation *cannot distinguish between ACD and IDA!*
- The reticulocyte-Hgb content may be a way to determine response to IV iron therapy.

Case 3

- ID and CC:
  A 42-year-old black woman with a history of iron deficiency on iron therapy comes to see you as a new patient.

- She was told that she was iron deficient 10 years ago and has been on iron supplementation for as many years.

- Labs:
  Hemoglobin 11.0 g/dL
  MCV 72 fL
  RDW 11 (11-16)
  Ferritin 800 ng/mL

You suspect a thalassemia and order a hemoglobin electrophoresis which comes back normal.

What is the most likely diagnosis?

A. She was iron deficient but is now iron replete
B. She has B-thalassemia trait
C. She has A-thalassemia trait
D. She has a myelodysplastic syndrome

Thalassemias

- Found most frequently in the Mediterranean, Africa, Western and Southeast Asia, India, and Burma

- Distribution parallels that of Plasmodium Falciparum

The thalassemias: Genetic, clinical, and laboratory findings

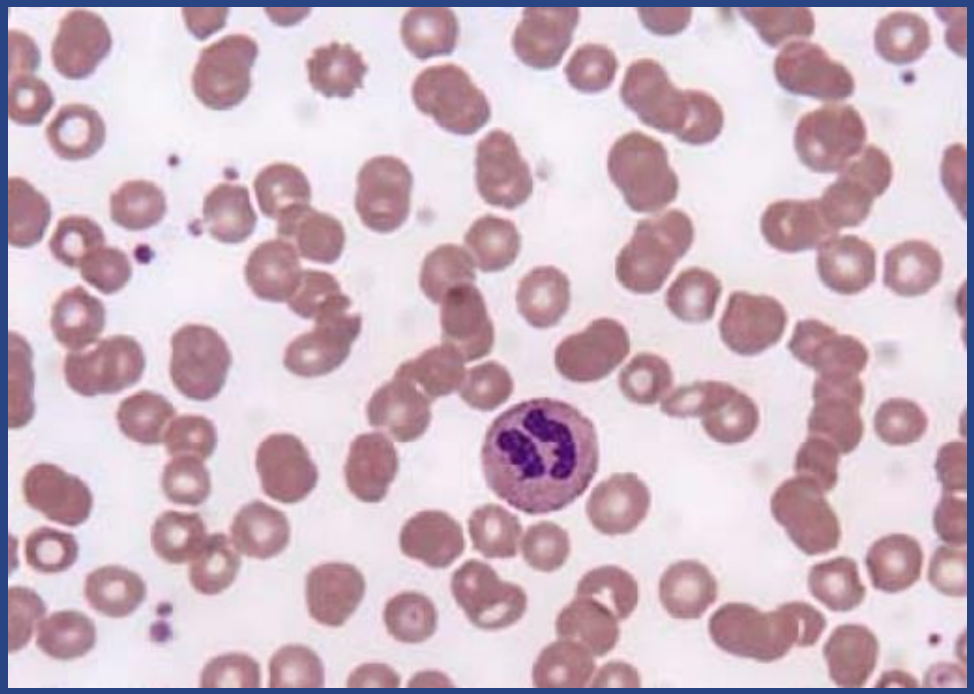
Disorder	Genotype	MCV	Anemia	Hb electrophoresis
Alpha thalassemia				
Silent carrier	α α / α -	NL	None	Normal <3 percent Hb Barts at birth
Trait	α α / - - or α - / α -	Low	Mild	Normal 3-8 percent Hb Barts at birth
Hb H disease (deletional)	α - / - -	Low	Moderate	5-30 percent Hb H present in adults 20-40 percent Hb Barts at birth
Major (fetal hydrops)	- - / - -	Low	Fatal	Hb Barts, Hb Portland, and Hb H present Hb A, Hb F, and Hb A2 are absent
Beta thalassemia				
Trait	β / β°	Low	Mild	Hb A2 increased (3.5-7 percent)
Intermedia	β^{+} / β^{+} and others*	Low	Moderate	Hb F increased in about 50 percent of patients
Major	$\beta^{\circ} / \beta^{\circ}$	Low	Severe	Hb A absent Only Hb A2 and Hb F are present

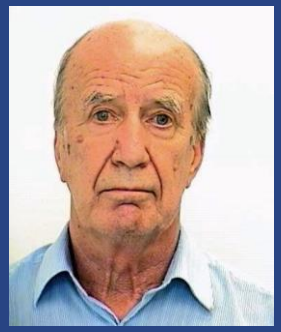
MCV: mean corpuscular volume; Hb: hemoglobin; NL: normal; β^{+}: thalassemic gene producing some β -chain; β°: thalassemic gene producing no β -chain.
* See text for multiple other genotypes.
Courtesy of Stephen A Landaw, MD, PhD.

Evaluating the Anemic Patient: The Morphologic Model

Normocytic Anemia: MCV 80-100

- Early iron deficiency
- Anemia of chronic disease
- Bone marrow disease
  - Invasion, aplasia
  - Myeloma, MGUS
- Chronic renal insufficiency
- Endocrine dysfunction
  - Hypothyroid
  - Hypopituitarism
- *Combined disorders in patients with high RDW*
  - ACD or IDA +B12 or folate deficiency





Case 1

- ID and CC

  75-year-old man with fatigue and palpitations admitted with anemia and oliguric renal failure. He has had some severe back pain for a month and physical therapy visits have not helped.

- Labs:
  Hemoglobin 7.2 g/dL
  MCV 84 fL
  RDW 12
  Creatinine 3.2 mg/dL
  Total Protein 9.1 g/dL
  Albumin 2.5 g/dL
  Urine protein/creatinine ratio: 4.5

Which of the following laboratory tests should be ordered next to help make the diagnosis?

A. Serum protein electrophoresis (SPEP)
B. Serum immunofixation (IFE)
C. Serum free light chains
D. B2-glycoprotein I
E. A,B, and C

Diagnosis of Multiple Myeloma

Table 1. Findings on Presentation for Patients with Multiple Myeloma

Symptom or laboratory finding	*Percentage of patients*
Anemia (hemoglobin < 12 g per dL [120 g per L])	73
Bone pain	58
Elevated creatinine (> 1.3 mg per dL [115 µmol per L])	48
Fatigue or generalized weakness	32
Hypercalcemia (calcium > 10.1 mg per dL [2.52 mmol per L])	28
Weight loss	24

Information from reference 9.

Table 2. Diagnostic Tests for Multiple Myeloma

Initial tests	*Confirmatory tests*
Complete blood count with differential	~~24-hour urine protein~~
Serum albumin, calcium, creatinine, electrolytes, and urea nitrogen	$Beta_2$-microglobulin
	Lactate dehydrogenase
	Serum free light chain assay
	Serum immunofixation electrophoresis
	Serum protein electrophoresis*
	Serum quantitative immunoglobulins
	Skeletal survey†
	~~Urine immunofixation electrophoresis~~
	~~Urine protein electrophoresis~~

https://www.aafp.org/afp/2017/0315/afp20170315p373

Case 2

- ID and CC:
  35-year-old woman with Crohn's disease is admitted with abdominal pain and bloody stools consistent with a flare of her disease for the past week. She has had several abdominal surgeries for her disease.

- Labs:
  Hemoglobin: 7.9 g/dL
  MCV: 85 fL
  RDW: 19
  Ferritin: 8
  B12: 211
  Absolute retic index: 1.2

What is the most likely cause of this patient's anemia?

A. Iron deficiency anemia
B. B12 deficiency
C. A and B
D. Bone marrow suppression

Multifactorial Causes of Anemia Resulting in Normocytic MCV with Elevated RDW

- MCV normal + RDW high
- Check retic count, iron studies, ferritin, B12 and folate
- Peripheral blood smear may give clues as well

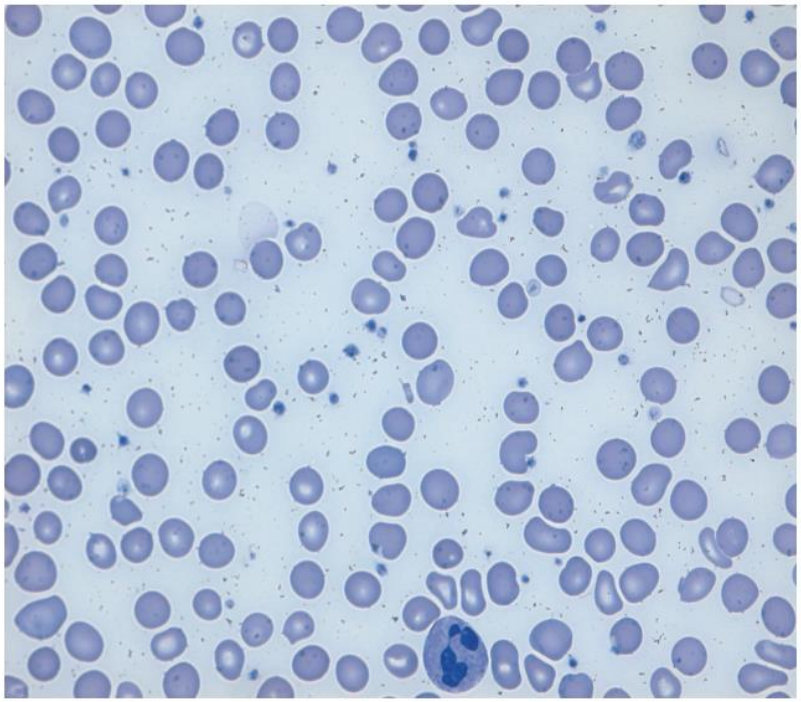


Fig. 1. Peripheral blood smear shows hypersegmented neutrophils and normocytic hypochromic red blood cells (Wright stain, ×1,000 magnification).

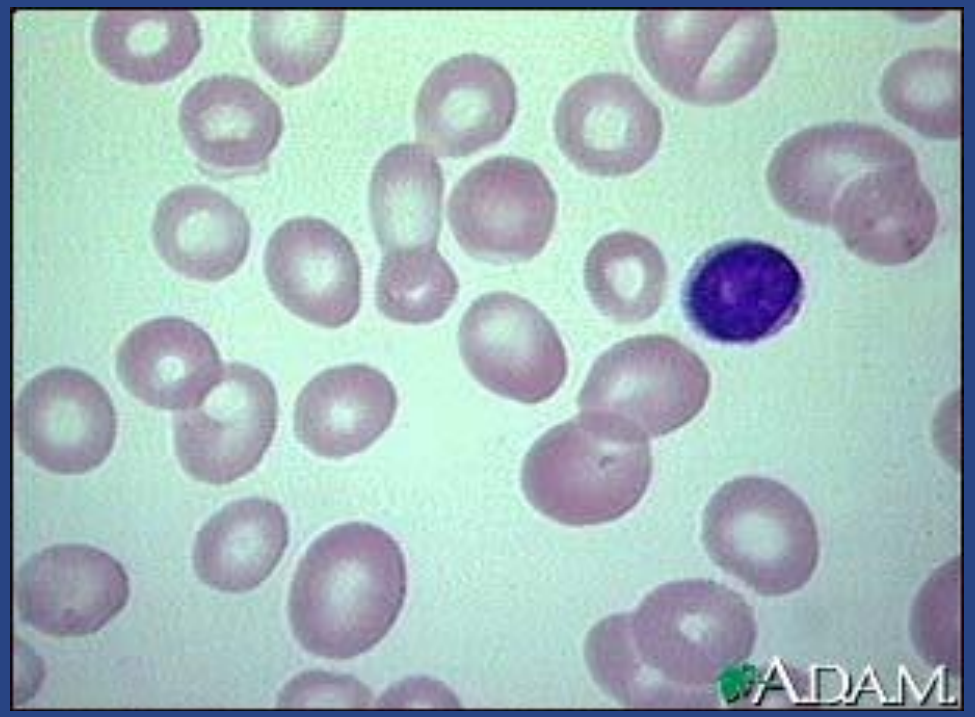
Spivak JL. Masked megaloblastic anemia. Arch Intern Med. 1982 Nov;142(12):2111-4.
PMID: 7138159.

Korean J Pediatr 2010;53(5):661-665

Evaluating the Anemic Patient: The Morphologic Model

Macrocytic Anemia
MCV > 100

- Ethanol abuse
- Drug-induced
  - AZT, hydroxyurea
- Liver disease
- Hypothyroidism
- Reticulocytosis
  - Hemolysis
  - Blood loss
- Myelodysplastic syndromes
- Folate deficiency
- B12 deficiency



Case 1

- ID and CC:

  42-year-old man with Hep C and decompensated liver cirrhosis is admitted for encephalopathy. He has no signs of GI bleeding.

- Labs:

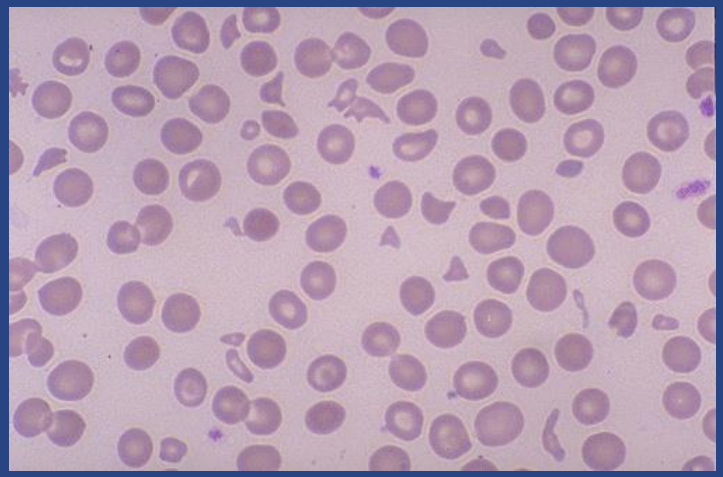
  Hemoglobin 7.0 g/dL
  Platelets 52K
  MCV 107 fL
  RDW 20 (11-16)
  Absolute retic index: 2.5
  Ferritin 1700

What is the most appropriate lab test to order next?

A. Peripheral smear
B. B12 and folate
C. Bone marrow biopsy
D. LDH and haptoglobin
E. Direct Coomb's test

Hemolytic Anemia

- Combination of normal LDH and haptoglobin rules OUT hemolysis with 92% sensitivity.
- Combination of elevated LDH and low haptoglobin rules IN hemolysis with 90% specificity.



- Peripheral smear can help to guide the work up *once hemolysis is confirmed:*

- Schistocyte
  - (mechanical shear)
- Spherocyte
  - (autoimmune)
- Acanthocyte
  - (liver disease)
- Bite cell
  - (oxidant damage)

Case 2

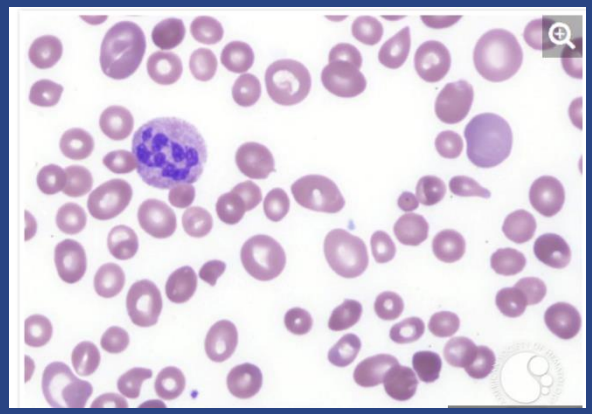
ID and CC:

59-year-old man with progressive dyspnea with exertion over 3 years

Labs:

- Hemoglobin 4.7 g/dL
- MCV 118 fL
- Creatinine 1.6 mg/dL
- Platelets 97 K
- WBC 2.3 K
- Retic count 7.5
- LDH 450
- Haptoglobin <10

A peripheral smear is shown:



What do you want to order next to evaluate the cause of his anemia?

A. B12
B. ADAMSTS13
C. Methylmalonic acid
D. Bone Marrow Biopsy

Causes of vitamin B12 deficiency

Gastric abnormalities
Pernicious anemia
Gastrectomy/Bariatric surgery
Gastritis
Autoimmune metaplastic atrophic gastritis
Small bowel disease
Malabsorption syndrome
Ileal resection or bypass
Crohn's disease
Blind loops
Pancreatitis
Pancreatic insufficiency
Diet
Strict vegans
Vegetarian diet in pregnancy
Agents that block absorption
Neomycin
Biguanides (eg, metformin)
Proton pump inhibitors (eg, omeprazole)
N2O anesthesia inhibits methionine synthase
Inherited transcobalamin II deficiency

UpToDate

Practice Question #1

- 61 year old AA man

WBC		14.8 H
RBC		1.88 L
HGB		5.4 ' C
HCT		15.1 ' C
MCV		80
MCH		28.7
MCHC		35.8
RDW-CV		17.8 H
RDW-SD		51.3 H
Platelet		162
Immature Platelet Fraction (IPF)		
MPV		11.6
Platelet Estimate		
Retic Hgb Equivalent (RET-He)		32.7 *
Diff Type		AUTO
Immature Granulocyte % (IG%)		1.0 *
Immature Granulocyte # (IG#)		0.2 H
Segs %		40
Lymphs %		38
Monos %		11
Eos %		7
Basos %		3 H
Neut#		5.9
Lymph#		5.6 H

- Retic count: 4.7%
- Retic index: 0.7 %
- B12: 910 pg/mL
- RBC Folate: 971 ng/mL
- Ferritin: 7,650 ng/mL

HEMOGLOBINOPATHY EVALUATION			
Helec RBC			2.84 L
Helec HGB			8.3 L
Helec MCV			84
Helec MCH			29.2
Helec RDW-CV			17.1 H
Helec RDW-SD			51.8
Hgb A			92.4 L
Hgb S			5.0 H
Hgb A2			2.6
Helec Interp			Helec Interp
GENERAL COAGULATION			

- Hemoglobin SS disease

Practice Question #1

- Differential diagnosis for a patient with sickle cell disease who presents with Hgb 5 and low retic index?

- Parvo B19
- Aplastic crisis from folate deficiency
- ? Epopoietin deficiency

CHEM GENER		
Specimen Integrity		
Glucose Level		120 * H
BUN		41 H
Creatinine		2.47 * H
Estimated Glomerular Filtration Rate		28 * L
BUN/Creat Ratio		17
Sodium		143
Potassium		4.1
Chloride		111 H
CO2		22
Anion Gap		10
Magnesium		
Calcium		8.2 L
Phosphorus		
Protein, Total		6.5
Albumin		2.3 L
Alb/Glob Ratio		0.5 L
Bilirubin Total		3.4 H
AST		98 H
ALT		49
Alkaline Phos		252 H

- Epo level: 5.0 mIU/mL (2.6-18.5)

Practice Question #2

- 27 yo male
- Abdominal pain and bloody diarrhea x 6 months associated with 40 lb weight loss.

- He was diagnosed with C diff in May and failed flagyl treatment.

- He presents with increasing abdominal pain and diarrhea with new symptoms of vomiting.

WBC	13.3 K/MM3 H
RBC	6.44 M/MM3 H
HGB	13.0 g/dL L
HCT	41.5 %
MCV	64 fL L
MCH	20.2 pg L
MCHC	31.3 g/dL
RDW-CV	17.2 % H
RDW-SD	35.0 fL L
Nucleated RBCs, Automated	0 %
Platelet	303 K/MM3
MPV	10.3 fL

MISC HEMO		
Retic %		1.6 %
Retic #		85 K/ul
Immature Retic Fraction (IRF)		27.9 % * H
Retic Hgb Equivalent [RET-He]		18.7 pg * L
Sed Rate		43 mm/hr H
Iron		53 ug/dL
Transferrin		142 mg/dL L
Trans % Sat		29.4 %
Ferritin		222 ng/mL

Hgb A				95.2 %
Hgb A2				4.8 % H
Helec Interp				Helec Interp

Result type: Helec Interp
Date/Time of Service: July 23, 2017 17:15 MST
Result status: Auth (Verified)
Performed By: ZHOU MD PhD, WENDI on July 25, 2017 14:39 MST
Verified by: ZHOU MD PhD, WENDI on July 25, 2017 14:39 MST
Encounter info: 36902559, BEMC, Observation, 07/23/2017 - 07/25/2017

*** Final Report ***

Elevation of Hemoglobin A2, consistent with beta thalassemia trait.

Reviewed by Dr. Wendi Zhou at Banner -- University Medical Center Phoenix.

Hemoglobinopathy evaluation involves interpretation of high performance liquid chromatography (HPLC) results in the context of red cell indices. Variant hemogolobins such as S, C, E, and others are detected. Some, but not all thalassemic disorders are detected. Consultative assistance is sought when necessary.

Objectives:

- Understand the basics of hematopoiesis.
- Define anemia and understand how to classify it based on the kinetic model and the morphologic model.
- Understand how to calculate the absolute reticulocyte count and absolute reticulocyte index and what these values mean.
- Understand the differentials of microcytic, normocytic, and macrocytic anemia and how to appropriately evaluate them to make a diagnosis.