About the speaker

- Dr Vishakha Mittal
- Professor, Department of Medicine, ABVIMS & Dr RML Hospital, New Delhi
- Alumnus of Lady Hardinge Medical College, 1991-96, did MD (Medicine) & Senior Residency from University College of Medical Sciences & GTB Hospital, 1997-2003.
- After a short stint at Maharaja Agrasen Hospital, Delhi, joined as Medical Officer at CGHS Wing, Safdarjung Hospital, New Delhi, in 2005, where also worked in the Hematology Department, thereafter in Department of Medicine, ABVIMS (formerly PGIMER) & Dr RML Hospital, New Delhi in 2014, as Assistant Professor, Medicine.
- PG supervisor with GGSIP University.

POLYCYTHEMIA

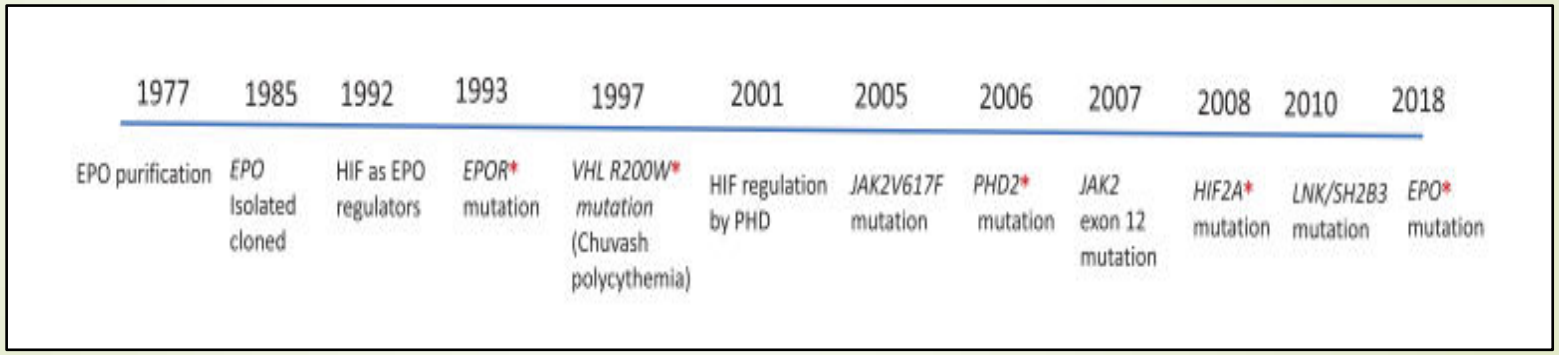
Current concepts and approach

APIDSC XXXII Annual Conference 17th December 2022

- Polycythemia means an increased amount of blood
- Refers to group of disorders characterized by increased red blood cell (RBC) mass, measured for convenience by an increase in hemoglobin level or hematocrit above normal for age, sex, race and altitude
- Erythrocytosis and polycythemia often used interchangeably
- Erythrocytosis appropriate when only the RBC lineage involved
- Polycythemia used for Polycythemia Vera (PV), a clonal stem cell disorder

Historical aspects

- Plethoric patients described by ancient clinicians[1]
- “Maladie de Vaquez,” referring to PV described by Vaquez in 1892
- Reviewed by William Osler in 1903
- William Dameshek categorised PV as Myeloproliferative neoplasm
- Seminal work by Drs. William Kaelin, Peter Ratcliffe and Gregg Semenza in understanding molecular mechanisms behind hypoxia sensing in erythropoiesis



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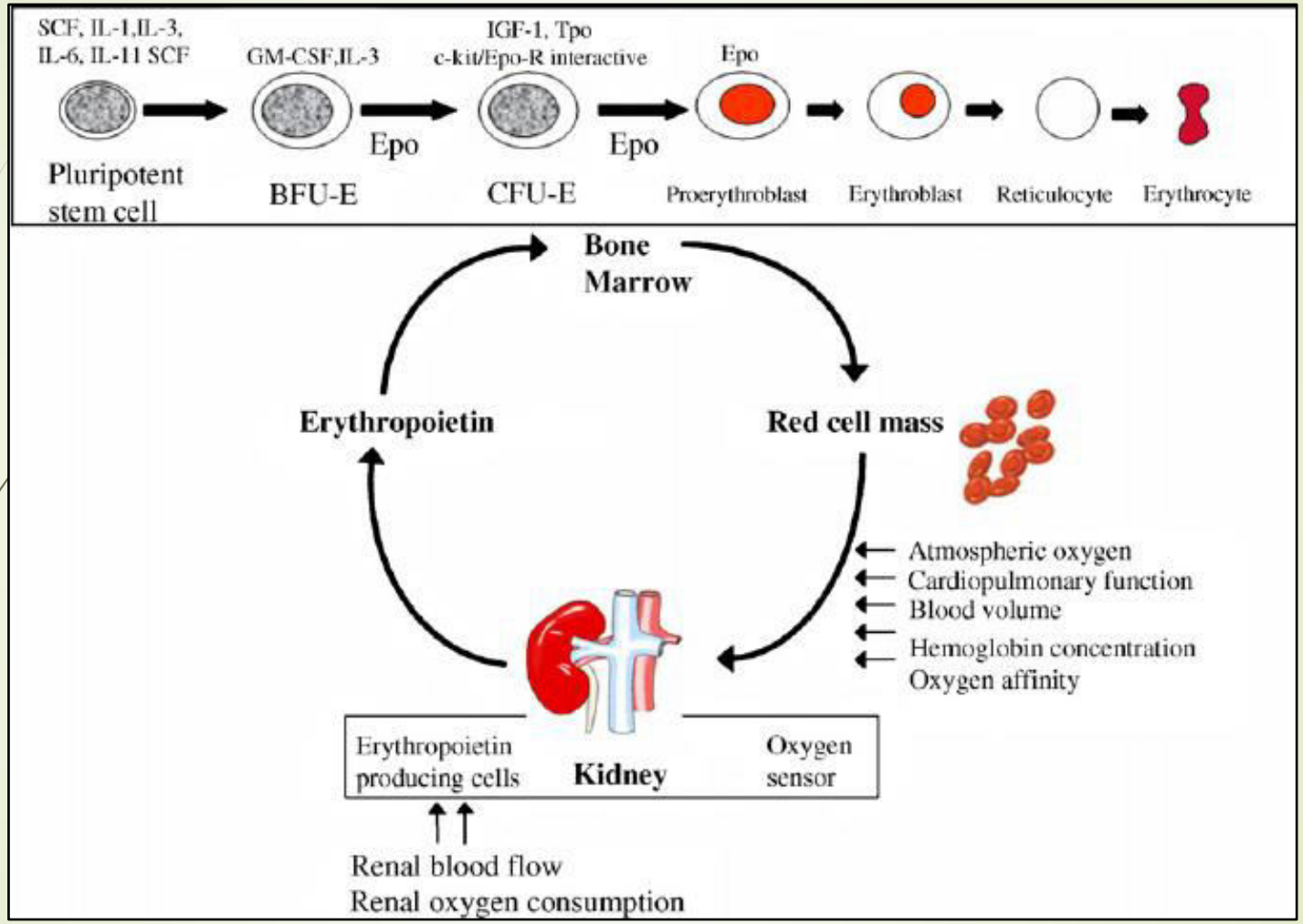
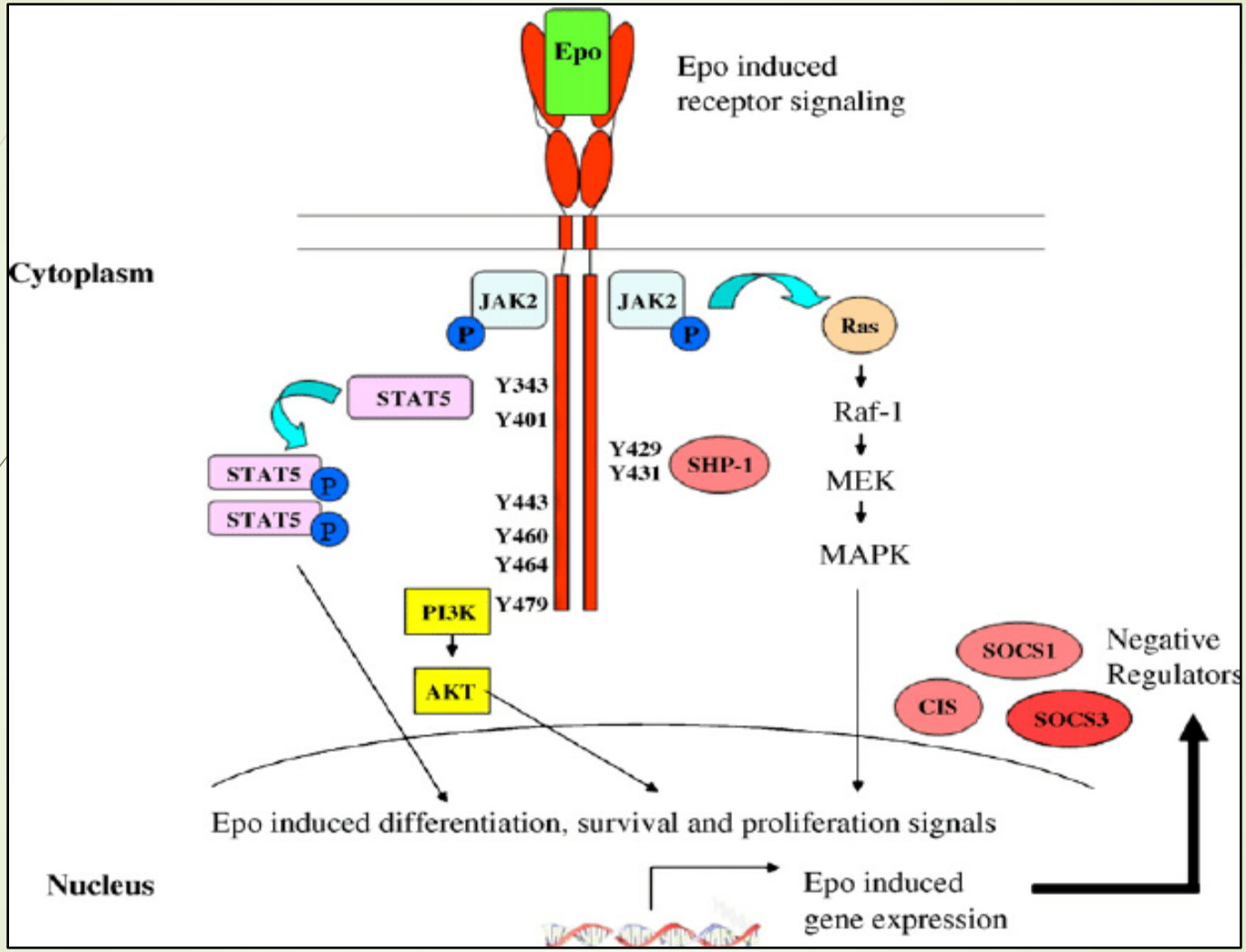
Normal erythropoiesis



Hodges, Vivien & Rainey, Susan & Lappin, Terry & Maxwell, Peter. (2007). Pathophysiology of anemia and erythrocytosis. Critical reviews in oncology/hematology. 64. 139-58. 10.1016/j.critrevonc.2007.06.006

EpoR activation



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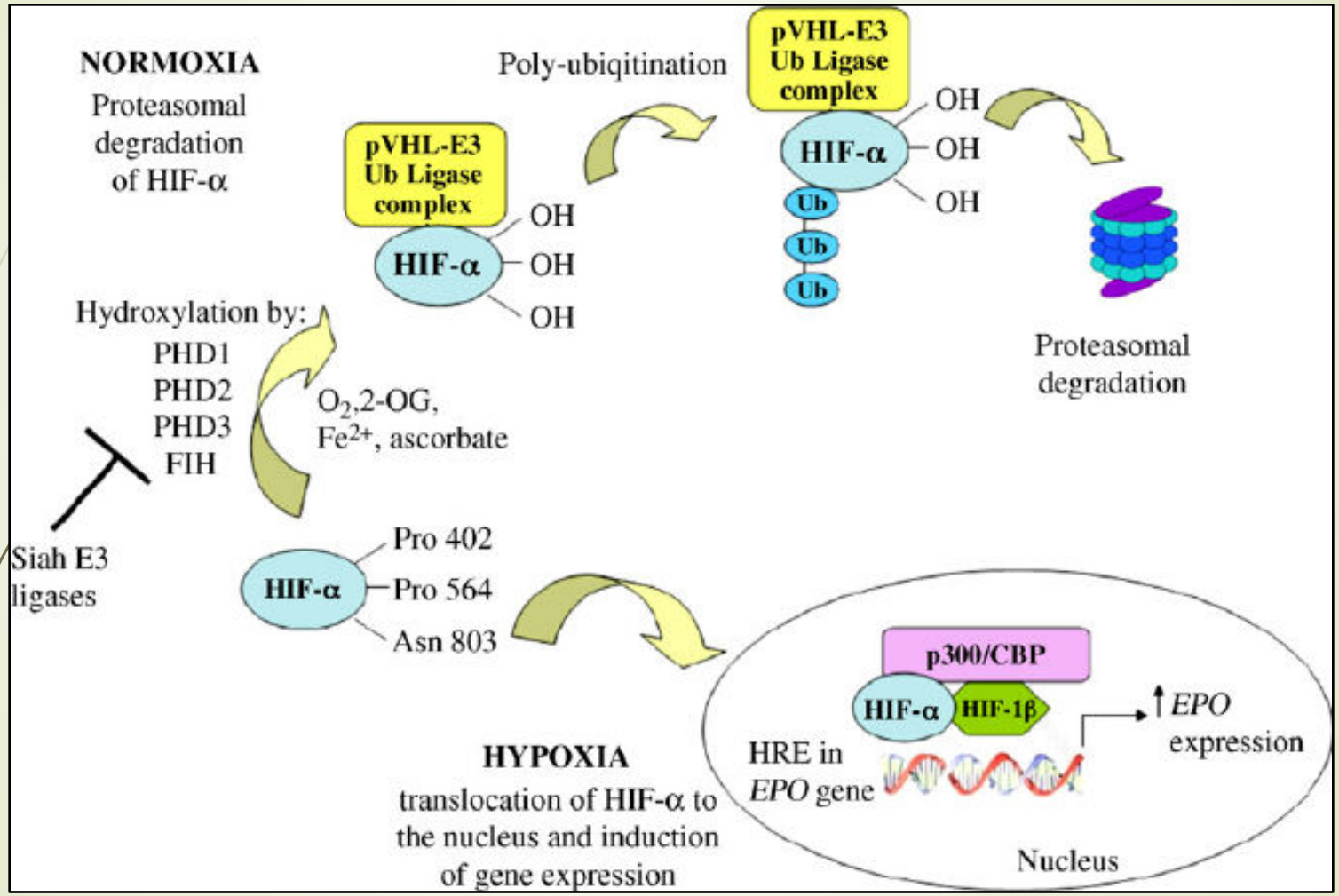
Hypoxia sensing pathway



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Classification

- **Primary erythrocytosis**
  - The molecular defect is intrinsic to the RBC precursors with activated EPO receptor signaling
  - EPO-independent mechanism with reduced EPO levels
- **Secondary erythrocytosis**
  - Defect lying extrinsic to the RBC precursors and driven by EPO
  - Have increased or inappropriately normal plasma EPO levels
- Both can be congenital or acquired
- **Idiopathic erythrocytosis**
  - Etiology remains unexplained after complete investigations

Siraj Mithoowani, Marissa Laureano, Mark A. Crowther and Christopher M. Hillis. Investigation and management of erythrocytosis. CMAJ August 10, 2020 192 (32) E913-E918

Classification	Erythropoietin levels	Cause
Primary		
Congenital	Low	*EPOR* mutation
Acquired	Low	*JAK2* mutations—Polycythemia vera
Secondary		
Congenital	Inappropriately normal or elevated	*VHL* mutations
		PHD2 (*EGLN1*) mutations
		HIF-2α (*EPAS1*) mutations
		High oxygen affinity hemoglobin variants (β/α globin gene mutations)
		2,3-BPGM mutations
Acquired	Elevated (EPO production by renal interstitial pericytes/ fibroblastic cells)	Chronic lung disorders
		Right to left cardiac shunt
		Hypoventilation syndromes
		High altitude residence
	Elevated (EPO production by neoplastic cells or via exogeneous administration)	Tumors (renal cell carcinoma, paraganglioma, cerebellar hemangioblastoma *etc*)
		Iatrogenic erythropoietin administration

Mutations in congenital erythrocytosis

Gene with MIM number	OMIM group	Chromosome locus	Number of exons; transcript size (bp)	Mode of inheritance
EPOR-133171	ECYT1	19p13.2	8; 2056	AD
VHL-608537	ECYT2	3p25.3	3; 4700	AR
EGLN1-606425	ECYT3	1q42.1	5; 7097	AD
EPAS1-603349	ECYT4	2p21	16; 5160	AD
EPO-617907	ECYT5	7q22.1	5; 1662	AD
HBB-141900	ECYT6	11p15.4	3; 754	AD
HBA1-141800	ECYT7	16p13.3	3; 605	AD
HBA2-141850	ECYT7	16p13.3	3; 577	AD
BPGM-613896	ECYT8	7q33	3; 1753	–

Abbreviations: AD, autosomal dominant; AR, autosomal recessive; bp, base pairs;*BPGM*, bisphosphoglycerate mutase; ECYT, erythrocytosis type/group; *EPO*, erythropoietin; *EPOR*, erythropoietin receptor gene; MIM, Mendelian inheritance in man; OMIM, online Mendelian inheritance in man.

Nabhajit Mallik, Reena Das,Pankaj Malhotra,Prashant Sharma .Congenital erythrocytosis. *Eur J Haematol.* 2021;107:29–37

Disease burden (1/2)

- Difficult to estimate since often un-diagnosed
- Polycythemia vera is rare, with an incidence of 0.84 and prevalence of 22 per 100 000
- Secondary erythrocytosis more common
  - 6%–18% of patients with COPD
  - 2%–8% of patients with OSA

Siraj Mithoowani, Marissa Laureano, Mark A. Crowther and Christopher M. Hillis. Investigation and management of erythrocytosis. CMAJ August 10, 2020 192 (32) E913-E918

Disease burden (2/2)

- Primary polycythemia estimated to be 0.6 per 100,000 adults in India [1]
  - In study in South India on PV(n=60)JAK-2 V617F seen in 38%, BM diagnostic in 67%. JAK-2 Exon 14 mutation testing was not available [2]
  - Another study of Primary Erythrocytosis (n=49) from the Armed forces, 20.4% had PV (8/10 were JAK-2 mutated), remaining 79.6% were idiopathic, genetic testing for congenital erythrocytosis was not available [3]
  - Study at PGI-C genetic basis of unexplained erythrocytosis in 18 patients, 11 diagnosed as CP, 3 with high-affinity hemoglobins & in 4, no diagnosis made [4]

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2. Ankita Bist Ajay Kandpal Sumitha A. Dhanasekaran R Article published online: 2022-07-11. DOI https://doi.org/ 10.1055/s-0042-1750702. ISSN 2454-6798. Thieme Medical and Scientific Publishers Pvt. Ltd. Accessed on 16/12/2022.< https://www.thieme-connect.com/products/ejournals/pdf/10.1055/s-0042-1750702.pdf>
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Nguyen et al. *Blood Cancer Journal* (2021)11:75
https://doi.org/10.1038/s41408-021-00463-x

Blood Cancer Journal

CORRESPONDENCE Open Access

Phenotypical differences and thrombosis rates in secondary erythrocytosis versus polycythemia vera

Eliane Nguyen[1,2], Michaël Harnois[3], Lambert Busque[1,2,3], Shireen Sirhan[3,4], Sarit Assouline[3,4,5], Ines Chamaki[2,3,6], Harold Olney[2,3,7], Luigina Mollica[1,2,3] and Natasha Szuber[1,2,3]

- Retrospective study at Quebec 1999-2019 compared SE(n=36) with PV(n=66)
- PV diagnosed with WHO 2016 criteria
- SE- defined as Hct as per WHO 2016, Negative JAK-2, inappropriate normal to elevated Epo
- SE-50% were idiopathic, 22.2% COPD/smoking, 5.6% OSA, 5.6% polycystic kidneys, 8.3% smoking+drugs/endocrine/OSA,5.4% OSA+
- Patients with SE were significantly younger, obese,more likely to be male and smokers, less splenomegaly seen.
- CV risk factors other than BMI and smoking were comparable

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- Epo was significantly different though some SE had low Epo & 25% PV had inappropriately normal Epo
- Hct>0.55 seen in 25% SE vs 48% PV(p=0.02)
- None of the SE patients(50% were tested) showed Endogenous Erythroid Colony formation in in-vitro culture
- Phlebotomy and aspirin use was higher in PV(p=0.004 & <0.0001)
- Pre-diagnosis thrombosis was comparable in both groups(p=0.68)
- Post-diagnosis thrombosis was seen only in PV (14%)

Diagnostic approach

- Erythrocytosis may be a true or apparent increase in red cell mass above normal for sex-, race- and altitude-adjusted values
- Absolute polycythaemia, patients must have an increased red cell mass (RCM) measurement >125% of normal for age, sex, altitude
- Hemoglobin (Hb)/hematocrit (Hct or PCV) are convenient measures
- 2016 WHO Hb/Hct thresholds for PV diagnosis 16.5 g/dL/0.49 in males and 16 g/dL/0.48 in females are used to initiate investigation
- Relative erythrocytosis, normal red cell mass with decreased plasma volume, should be excluded

Acute conditions with dehydration,endocrine conditions, drugs

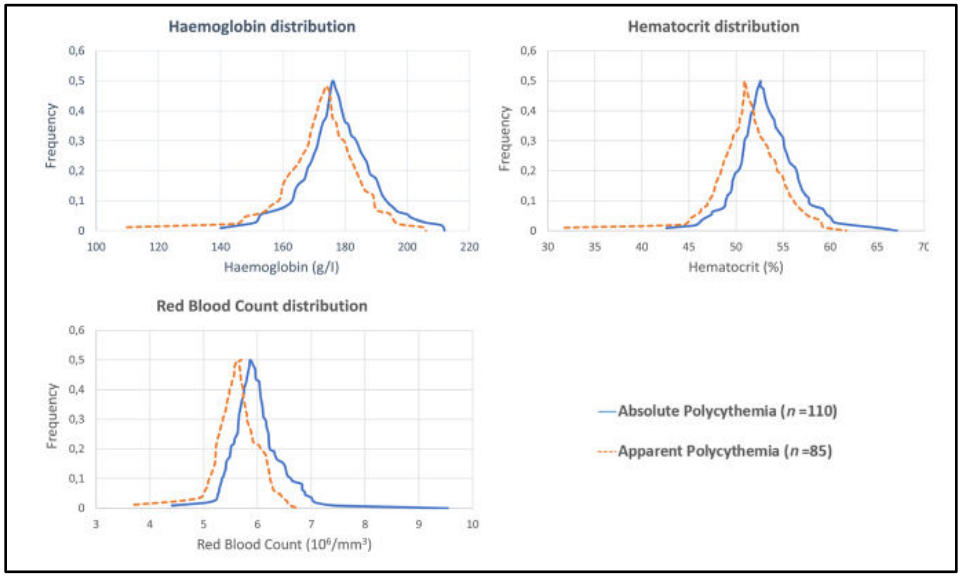
Gaisbock syndrome- sustained rise, 16-18g/dl, PCV 0.48-0.55

1. Siraj Mithoowani, Marisa Lauzon, Mark A. Crowther, and Christopher M. Hillis. Investigation and management of erythrocytosis. CMAJ August 10, 2020 192 (32) E913-E918
2. Mary Frances McMullin, Claire N. Harrison, Sahra Ali, Catherine Cargo, Frederick Chen, Joanne Ewing,et al. . A guideline for the diagnosis and management of polycythaemia vera.. BJH. 2019(184):176-191.

Diagnostic approach

- Considerable overlap in values
- RCM estimation is the gold standard and Hct >0.60 (males) or >0.56 (females) can be assumed to have an absolute erythrocytosis
- WHO 2016 guideline for PV hemoglobin /PCV
  - greater than 18.5 g/dL in men(PCV 0.55)

    Or
  - greater than 16.5 g/dL in women (PCV 0.50)



Grenier, M., Callegarin, D., Nughe, M., Gardie, B., Riedinger, J.M. and Girodon, F. (2020), Can absolute polycythaemia be identified without measurement of the red cell mass?. Br J Haematol, 190: e107-e110.

Clinical approach (1/2)

- Detailed clinical history and examination are essential
- Often incidental diagnosis
- Hyperviscosity symptoms headache, visual disturbances or light-headedness. fatigue, mental fog, tinnitus, chest pain, palpitations, dyspnea, abdominal and bone pain
- Complications –thrombosis, hemorrhage
- PV-pruritus often aquagenic, gout and constitutional symptoms, such as fatigue, erythromelalgia
- Secondary causes- exposure to carbon monoxide and symptoms of obstructive sleep apnea, cardiac or pulmonary disease
- Place of residence
- Drug use- tobacco, alcohol, androgens, ESA
- Family history
- Past history esp cardiovascular risk factors, Review of previous records especially any labs

Clinical approach (2/2)

- Clinical examination
- Vitals- esp BP, oxygen Saturation<92%
- Body habitus
- GPE for diagnostic clues
- Systemic examination-
  - Lung disease
  - Cardiac disease
  - Abdomen- lump, splenomegaly
  - CNS & fundus exam

1. Siraj Mithoowani, Marissa Laureano, Mark A. Crowther and Christopher M. Hillis. Investigation and management of erythrocytosis. CMAJ August 10, 2020 192 (32) E913-E918
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Stage 1 Investigations

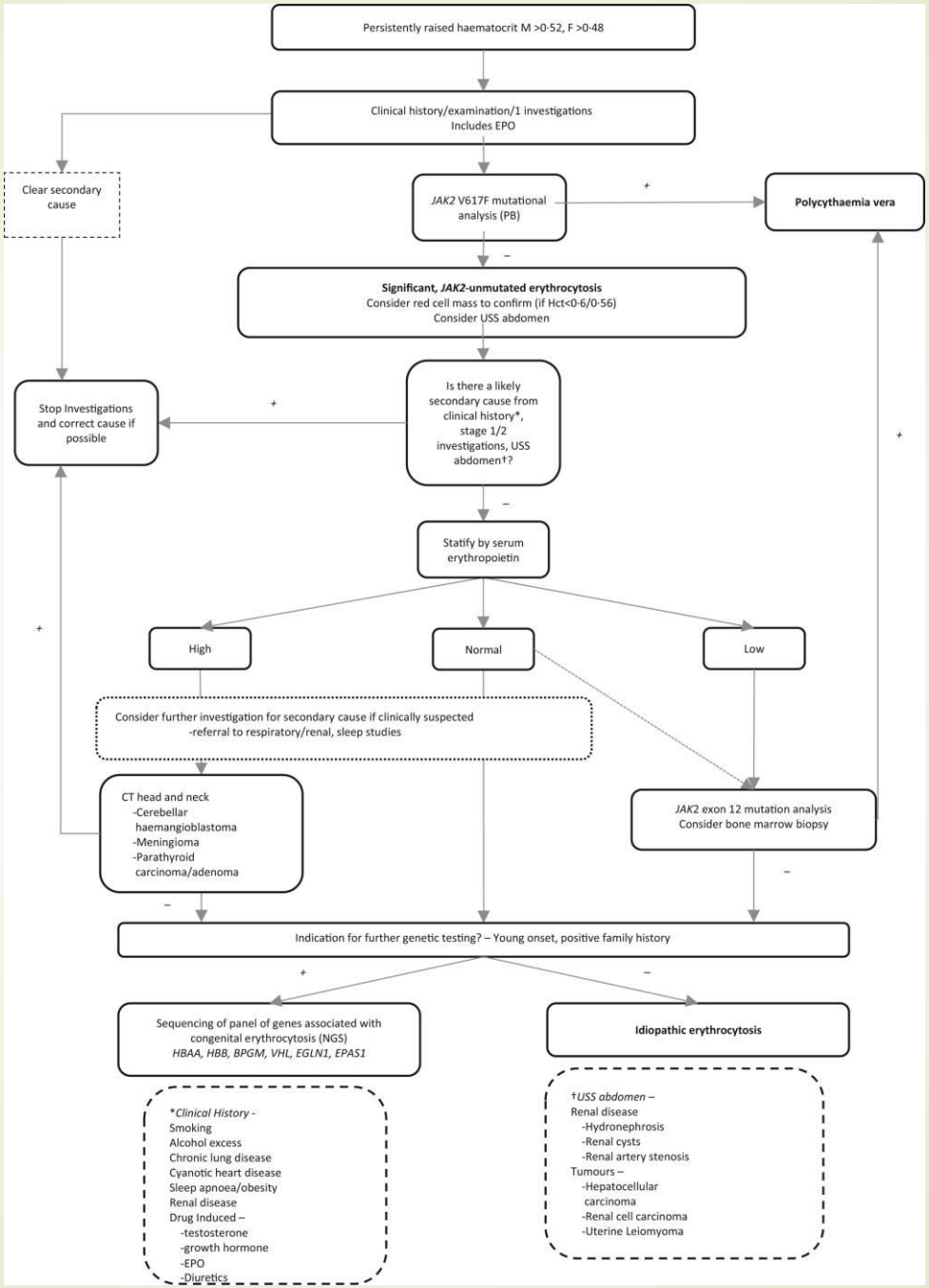
- CBC with blood film
  - Red cells only
  - Multiple lines, neutrophilia and thrombocytosis- favour PV, blasts, leucoerythroblastic features and monocytosis – marrow examination indicated
- RFT/LFT/Calcium
- **ABG** in hypoxic patients, co-oximetry for CO, Methemoglobin levels, determine p50 for high affinity variants
- Iron levels and ferritin– iron deficiency common, masked eythrocytosis
- **Serum Erythropoietin** levels
- **JAK-2 *V617F*** mutation analysis
- Investigations for secondary erythrocytosis
  - symptom directed
  - may include further imaging, overnight oximetry, PFT,ECHO

Stage 2 investigations

- **JAK 2 negative polycythemia**
  - Red Cell mass studies more than 25% above the mean predicted value is diagnostic of an absolute erythrocytosis
  - Abdominal ultrasound- splenomegaly or secondary cause not obvious clinically
- **Stratified as per EPO**
- Normal to low EPO
  - JAK2 exon 12 analysis
  - Bone marrow biopsy
  - Acquired genetic abnormality -abnormal karyotype,SH2B3 (LNK) mutation
  - Germline mutations in EPO receptor gene
- High EPO
  - Head & Neck CT/other imaging
  - Gene sequencing for congenital erythrocytosis

i. Genes in the oxygen sensing pathway-VHL, EGLN1, EPAS1

ii. High oxygen affinity haemoglobins - mutations in the globin genes & BPGM mutations (p50 may be low)


Persistently raised haematocrit M >0·52, F >0·48
Clinical history/examination/1 investigations
Includes EPO
Clear secondary cause
JAK2 V617F mutational analysis (PB)
+
Polycythaemia vera
–
Significant, JAK2-unmutated erythrocytosis
Consider red cell mass to confirm (if Hct<0·6/0·56)
Consider USS abdomen
Is there a likely secondary cause from clinical history*, stage 1/2 investigations, USS abdomen†?
+
Stop Investigations and correct cause if possible
–
Statify by serum erythropoietin
High
Normal
Low
Consider further investigation for secondary cause if clinically suspected
-referral to respiratory/renal, sleep studies
CT head and neck
-Cerebellar haemangioblastoma
-Meningioma
-Parathyroid carcinoma/adenoma
JAK2 exon 12 mutation analysis
Consider bone marrow biopsy
Indication for further genetic testing? – Young onset, positive family history
+
–
Sequencing of panel of genes associated with congenital erythrocytosis (NGS)
HBAA, HBB, BPGM, VHL, EGLN1, EPAS1
Idiopathic erythrocytosis
*Clinical History -
Smoking
Alcohol excess
Chronic lung disease
Cyanotic heart disease
Sleep apnoea/obesity
Renal disease
Drug Induced –
-testosterone
-growth hormone
-EPO
-Diuretics
†USS abdomen –
Renal disease
-Hydronephrosis
-Renal cysts
-Renal artery stenosis
Tumours –
-Hepatocellular carcinoma
-Renal cell carcinoma
-Uterine Leiomyoma

When to refer

- No established criteria
- Low or normal erythropoietin level and unproductive workup for secondary erythrocytosis
- Women with diagnosed PV who desire pregnancy
- Diagnosed with High risk PV
- Patients with diagnosed PV who are refractory or intolerant to treatment with hydroxyurea, Phlebotomy
- Idiopathic erythrocytosis-with no diagnosis despite extensive appropriate investigations

Diagnostic criteria for Polycythemia Vera (WHO 2016)

Diagnosis of polycythemia vera requires all 3 major criteria OR the first 2 major criteria and the minor criterion

Major criteria

- Hemoglobin level > 165 g/L in men, > 160 g/L in women OR hematocrit > 0.49 in men, > 0.48 in women OR increased erythrocyte mass
- Bone marrow biopsy showing hypercellularity for age with trilineage growth (panmyelosis) including prominent erythroid, granulocytic and megakaryocytic proliferation with pleomorphic, mature megakaryocytes*
- Presence of JAK2 V617F or JAK2 exon 12 mutation

Minor criterion

- Subnormal serum erythropoietin level

*Bone marrow biopsy is not required for patients with sustained absolute erythrocytosis, defined as a hemoglobin level greater than 185 g/L in men (hematocrit 0.55) or greater than 165 g/L in women (hematocrit 0.50) if the third major criterion and the minor criterion are met.

Clinical course & management

- Hyper viscosity and risk of thrombosis are major concerns.
- 15% PV present with thrombosis, accounted for 45% deaths over 4000P.Y.
- Risk of secondary myelofibrosis & leukemia in PV
- Accurate diagnosis determines management
  - Primary & Secondary
  - Idiopathic- upto 70% remain undiagnosed, may have rare mutations(*LNK*, Hypoxia sensing pathway, *PEIZO*), typically young male without splenomegaly or thrombosis, asymptomatic, low WBC & platelets, high LDH
- Clinical follow-up and monitoring advisable in idiopathic cases

1. Mary Frances McMullin, Claire N. Harrison, Sahra Ali, Catherine Cargo, Frederick Chen, Joanne Ewing,et al. . A guideline for the diagnosis and management of polycythaemia vera.. BJH. 2019(184):176-191.
2. Mary Frances McMullin, Claire N. Harrison, Sahra Ali, Catherine Cargo, Frederick Chen, Joanne Ewing,et al. A guideline for the management of specific situations in polycythaemia vera and secondary erythrocytosis . BJH. 2019:, (184) :161–175
3. Gangat, N., Szuber, N., Pardanani, A. *et al. JAK2* unmutated erythrocytosis: current diagnostic approach and therapeutic views. *Leukemia* 35, 2166–2181 (2021).

Goals of treatment in PV

- Reduce thrombosis and haemorrhage risk
  - Low risk- age<65 years, no prior thrombosis- aspirin+phlebotomy
  - High risk – age>65 years, prior thrombosis- aspirin±phlebotomy+cytoreduction with hydroxyurea, JAK-2 inhibitor as second line
- Minimise complications and symptomatology
- Minimise risk of transformation to myelofibrosis and acute leukaemia
- Manage specific situations such as pregnancy and surgery
- Achieve good haematocrit control to <0.45

Treatment in other forms of erythrocytosis

- Limited evidence due to heterogeneity, extrapolation from PV studies
- Smoking cessation, control of CV risk
- Low dose aspirin in patients with CV risk factors, Chuvash polycythemia
- Arterial & venous thrombosis managed on established lines
- LTOT & CPAP as indicated
- Review drugs when causative
- ACEi or ARB for post transplant causes
- Cytoreduction is not indicated

Treatment in other forms of erythrocytosis

- Phlebotomy - risk-benefit analysis needed
  - Use only for relief of symptoms with documented response
  - Depletes iron stores & stabilizes HIF and Epo mediated erythropoiesis, microcytic cells are rigid
  - Avoid in physiologic erythrocytosis or cardiopulmonary disease where symptoms rather than a Hct target addressed
  - Arbitrary target of 0.45-0.55 in idiopathic cases with symptoms or history of thrombosis

Treatment in congenital erythrocytosis

- Young age, family history ,rare & poorly described
- Major thrombosis & PAH described in CP and other congenital erythrocytosis
- **Chuvash polycythemia-** increased thrombosis in homozygous patients, phlebotomy may be detrimental, JAK-2 inhibition under trial
- **Congenital EPAS1** gof mutations should be screened for neuroendocrine tumors since association described with somatic mutations in patients with these tumors and erythrocytosis
- Phlebotomy may be considered in following situation(Grade 2C)
  - Symptomatic polycythemia
  - Previous thrombotic episode
  - Asymptomatic individual in whom affected family member with the same genetic lesion has had thrombotic episode (esp. high affinity hemoglobins)
  - Hct target of 0.52 is suggested

CONCLUSION

- Not always Polycythemia Vera
- Meticulous history & examination can exclude relative polycythemia & differentiate secondary causes.
- Rational diagnostic approach and step1 investigations investigation(Epo, ABG+p50, JAK-2) can distinguish Primary from Secondary causes
- Treatment modalities are limited
- Phlebotomy should be applied judiciously
- Many cases may remain undiagnosed and should be kept in follow-up.

THANK YOU