Pathological evaluation of haematological diseases on bone marrow biopsies

Educational course, Belgian Society of Hematology

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(Bone marrow Particle clot)

BONE MARROW

3 main indications :
-Diagnostic purposes
-Staging for malignant diseases
-Monitoring

Bone marrow aspirate

Bacteriology if necessary

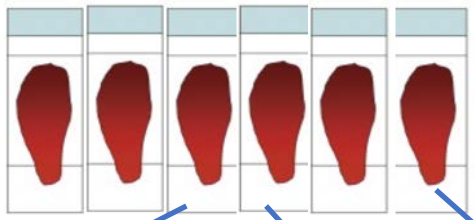
Bone marrow trephine biopsy

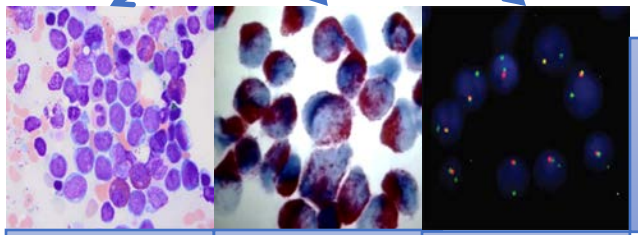
Smear Preparation

+ Heparin

+EDTA

Heparin

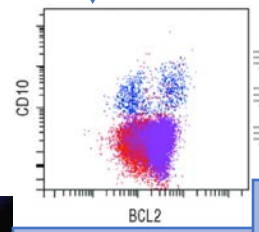




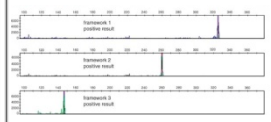
MGG cytology (hours)

Cyto-chemistry (hours)

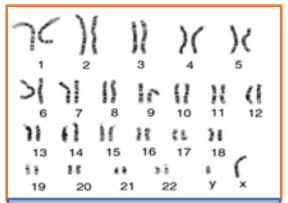
FISH If necessary (days)



Flow cytometry (hours)



Molecular biology (clonal expansion, NGS, …) Days-Weeks



Cytogenetics days-weeks

Karyotyping FISH

FFPE 2-10 days

Morpho, IHC, FISH, (clonal expansion, NGS)

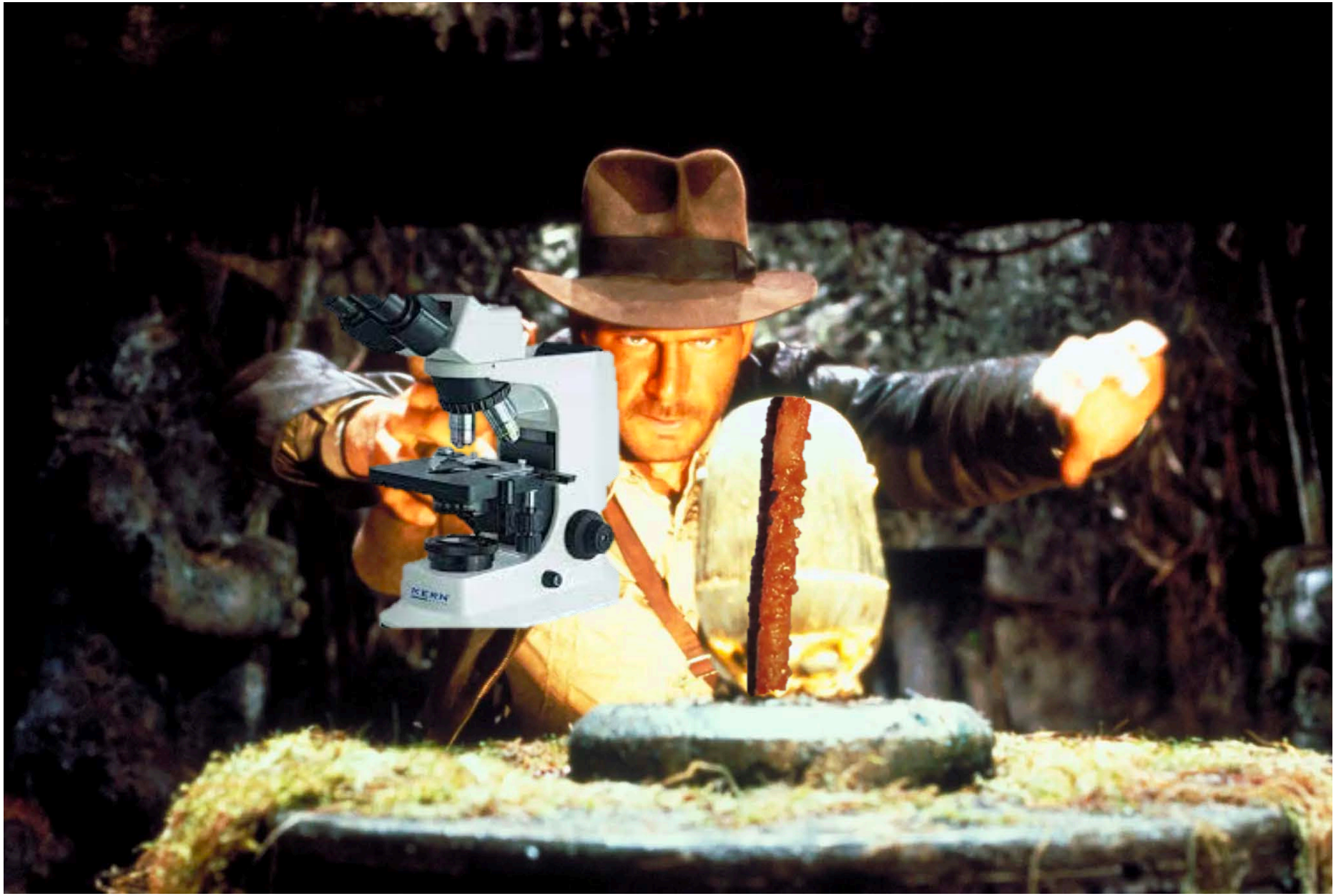
BM biopsy touch imprint (hours)
Usefull if dry tap

BM aspirate or trephine biopsy ?

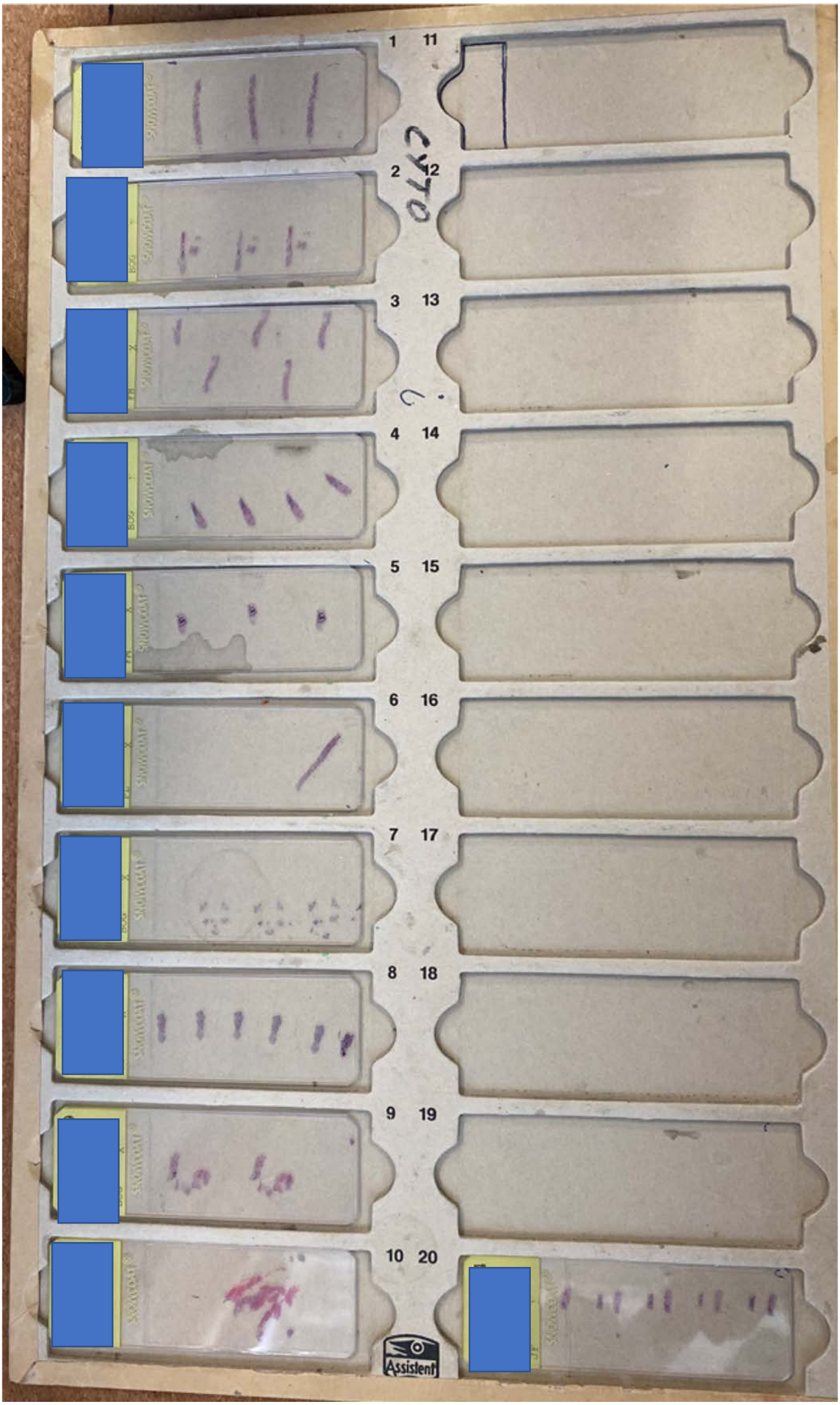
A thorough bone marrow examination includes ***both*** BM aspiration and trephine biopsy.

- **MDS:** BM aspirate >>> BM biopsy
- **MPN:** BM aspirate << **BM biopsy**
- **MPN/MDS** BM aspirate > BM biopsy
- **AML:** BM aspirate >>> BM biopsy
- **NHL:** BM aspirate << **BM biopsy**
- **HL:** BM aspirate <<< **BM biopsy**
- **MM:** BM aspirate < **BM biopsy**
- **Carcinoma:** BM aspirate << **BM biopsy**

Pathologist POV




LEICA
HOW YA DOIN?
DELL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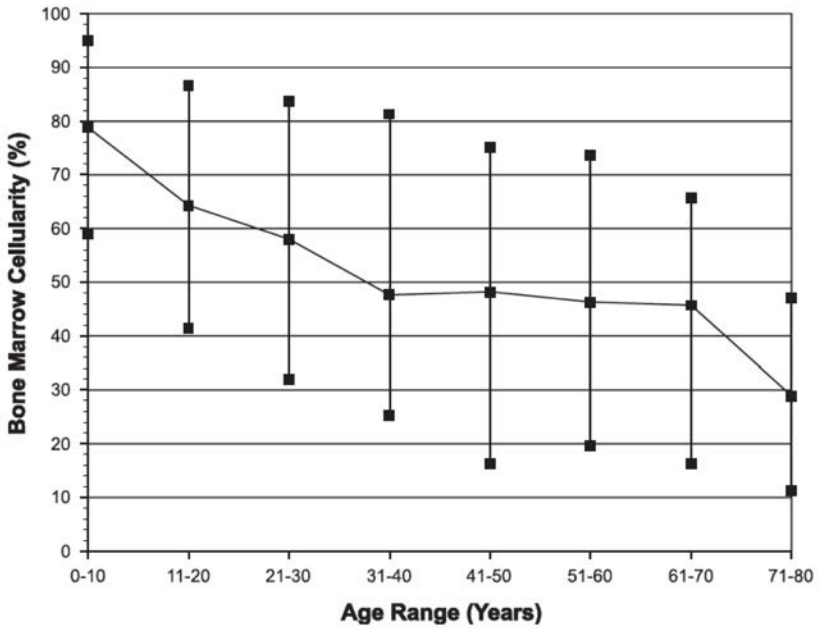
Assistent

BM biopsy analysis : Basics

Cellularity

100-age = expected cellularity for age



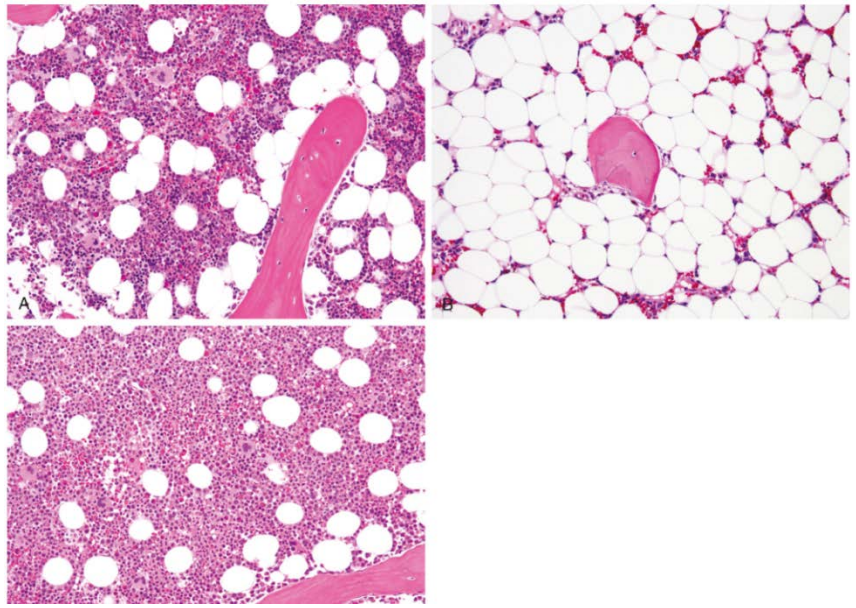


Fig. 6. Normal bone marrow cellularity relative to age. Data from (37).

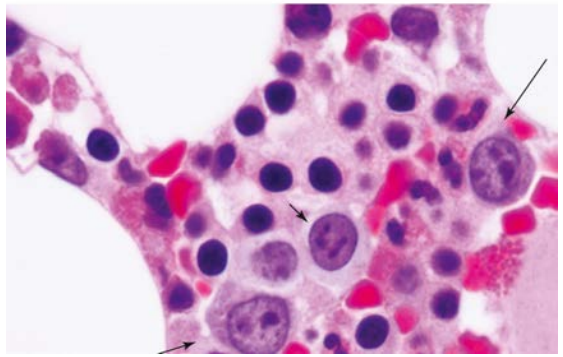
Severe AA is characterized by a markedly hypocellular bone marrow (<25% of normal for age or 25 to 50% of normal with <30% hematopoietic cells) accompanied by two of the following: granulocytes <0.5 × 10^9/L; platelets <20 × 10^9/L; or corrected reticulocyte count <1%

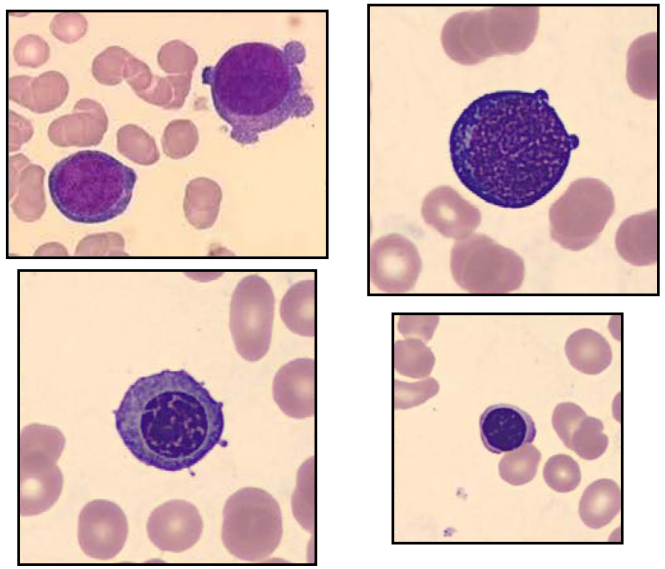
Riley et al., JCLA, 2009

Jaffe, Hematopathology, second edition, 2017

Haematopoesis

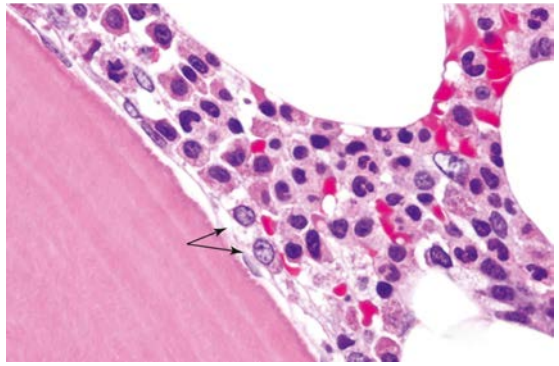
Erythropoïesis

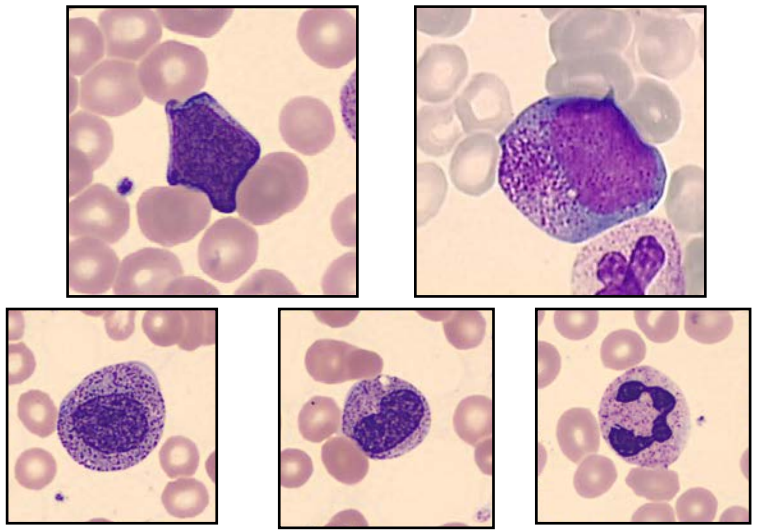




Courtesy of Pr. Tassin and Dr. Keutgens

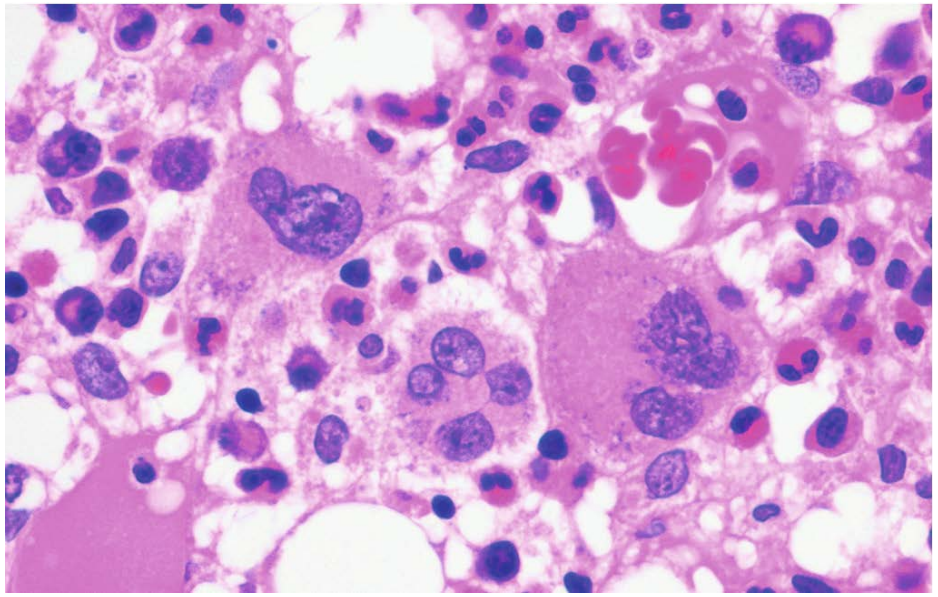
Granulopoïesis

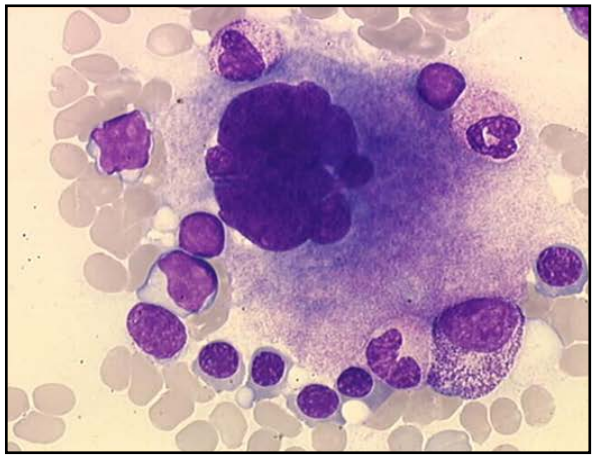




Jaffe, Hematopathology, second edition, 2017

Mégacaryocytes





Courtesy of Pr. Tassin and Dr. Keutgens

Other?

- Monocytes
- Macrophages
- Plasma cells
- Lymphoid cells
- Mast cells
- Oestoclasts
- **Bone**
- Iron
- …

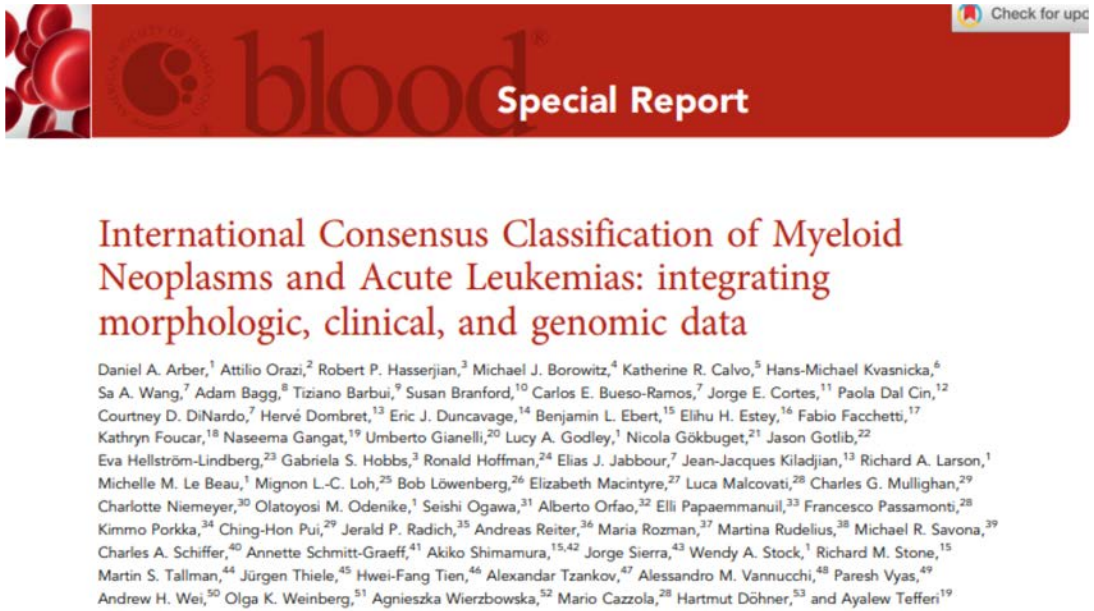
Jaffe, Hematopathology, second edition, 2017

Immunohistochemistry

- Erythroid: GlycophorinA, LMO2, **CD71**
- Myeloid: **MPO**
- Megacaryocytes: **CD61**, Factor VIII
- Blasts: **CD34, CD117**, CD33
  - CD34+ cells are rare in normal marrow
  - CD34 does not equal blast
    - not all blasts are CD34+
    - not all CD34+ cells are blasts
  - Not all AML's are CD34+
  - CD34 is not lineage specific
- Mastocytes: Tryptase, CD117, CD25, CD2, CD30
- Plasma cells: CD138, IgKappa, IgLambda
- Lymphocytes: CD20, CD3, CD30, …
- …

Classifications : BM Pathology

ICC, 2022 Myeloid classification



blood
Special Report

International Consensus Classification of Myeloid Neoplasms and Acute Leukemias: integrating morphologic, clinical, and genomic data

Daniel A. Arber,[1] Attilio Orazi,[2] Robert P. Hasserjian,[3] Michael J. Borowitz,[4] Katherine R. Calvo,[5] Hans-Michael Kvasnicka,[6] Sa A. Wang,[7] Adam Bagg,[8] Tiziano Barbui,[9] Susan Branford,[10] Carlos E. Bueso-Ramos,[7] Jorge E. Cortes,[11] Paola Dal Cin,[12] Courtney D. DiNardo,[7] Hervé Dombret,[13] Eric J. Duncavage,[14] Benjamin L. Ebert,[15] Elihu H. Estey,[16] Fabio Facchetti,[17] Kathryn Foucar,[18] Naseema Gangat,[19] Umberto Gianelli,[20] Lucy A. Godley,[1] Nicola Gökbuget,[21] Jason Gotlib,[22] Eva Hellström-Lindberg,[23] Gabriela S. Hobbs,[3] Ronald Hoffman,[24] Elias J. Jabbour,[7] Jean-Jacques Kiladjian,[13] Richard A. Larson,[1] Michelle M. Le Beau,[1] Mignon L.-C. Loh,[25] Bob Löwenberg,[26] Elizabeth Macintyre,[27] Luca Malcovati,[28] Charles G. Mullighan,[29] Charlotte Niemeyer,[30] Olatoyosi M. Odenike,[1] Seishi Ogawa,[31] Alberto Orfao,[32] Elli Papaemmanuil,[33] Francesco Passamonti,[28] Kimmo Porkka,[34] Ching-Hon Pui,[29] Jerald P. Radich,[35] Andreas Reiter,[36] Maria Rozman,[37] Martina Rudelius,[38] Michael R. Savona,[39] Charles A. Schiffer,[40] Annette Schmitt-Graeff,[41] Akiko Shimamura,[15,42] Jorge Sierra,[43] Wendy A. Stock,[1] Richard M. Stone,[15] Martin S. Tallman,[44] Jürgen Thiele,[45] Hwei-Fang Tien,[46] Alexandar Tzankov,[47] Alessandro M. Vannucchi,[48] Paresh Vyas,[49] Andrew H. Wei,[50] Olga K. Weinberg,[51] Agnieszka Wierzbowska,[52] Mario Cazzola,[28] Hartmut Döhner,[53] and Ayalew Tefferi[19]

WHO 5th edition, 2022

Leukemia www.nature.com/leu

REVIEW ARTICLE OPEN

The 5th edition of the World Health Organization Classification of Haematolymphoid Tumours: Myeloid and Histiocytic/ Dendritic Neoplasms

Joseph D. Khoury[1], Eric Solary[2], Oussama Abla[3], Yassmine Akkari[4], Rita Alaggio[5], Jane F. Apperley[6], Rafael Bejar[7], Emilio Berti[8], Lambert Busque[9], John K. C. Chan[10], Weina Chen[11], Xueyan Chen[12], Wee-Joo Chng[13], John K. Choi[14], Isabel Colmenero[15], Sarah E. Coupland[16], Nicholas C. P. Cross[17], Daphne De Jong[18], M. Tarek Elghetany[19], Emiko Takahashi[20], Jean-Francois Emile[21], Judith Ferry[22], Linda Fogelstrand[23], Michaela Fontenay[24], Ulrich Germing[25], Sumeet Gujral[26], Torsten Haferlach[27], Claire Harrison[28], Jennelle C. Hodge[29], Shimin Hu[1], Joop H. Jansen[30], Rashmi Kanagal-Shamanna[1], Hagop M. Kantarjian[31], Christian P. Kratz[32], Xiao-Qiu Li[33], Megan S. Lim[34], Keith Loeb[35], Sanam Loghavi[1], Andrea Marcogliese[19], Soheil Meshinchi[36], Phillip Michaels[37], Kikkeri N. Naresh[35], Yasodha Natkunam[38], Reza Nejati[39], German Ott[40], Eric Padron[41], Keyur P. Patel[1], Nikhil Patkar[42], Jennifer Picarsic[43], Uwe Platzbecker[44], Irene Roberts[45], Anna Schuh[46], William Sewell[47], Reiner Siebert[48], Prashant Tembhare[42], Jeffrey Tyner[49], Srdan Verstovsek[31], Wei Wang[1], Brent Wood[50], Wenbin Xiao[51], Cecilia Yeung[35] and Andreas Hochhaus[52]

- ICC, 2022 Myeloid classification

Major ICC categories of myeloid neoplasms and acute leukemias

MPNs
Chronic myeloid leukemia
Polycythemia vera
Essential thrombocythemia
Primary myelofibrosis
Early/prefibrotic primary myelofibrosis
Overt primary myelofibrosis
Chronic neutrophilic leukemia
Chronic eosinophilic leukemia, not otherwise specified
MPN, unclassifiable
Myeloid/lymphoid neoplasms with eosinophilia and tyrosine kinase gene fusions
Myeloid/lymphoid neoplasm with *PDGFRA* rearrangement
Myeloid/lymphoid neoplasm with *PDGFRB* rearrangement
Myeloid/lymphoid neoplasm with *FGFR1* rearrangement
Myeloid/lymphoid neoplasm with *JAK2* rearrangement
Myeloid/lymphoid neoplasm with *FLT3* rearrangement
Myeloid/lymphoid neoplasm with *ETV6::ABL1*
Mastocytosis
Myelodysplastic/myeloproliferative neoplasms
Chronic myelomonocytic leukemia
Clonal cytopenia with monocytosis of undetermined significance
Clonal monocytosis of undetermined significance
Atypical chronic myeloid leukemia
Myelodysplastic/myeloproliferative neoplasm with thrombocytosis and *SF3B1* mutation
Myelodysplastic/myeloproliferative neoplasm with ring sideroblasts and thrombocytosis, not otherwise specified
Myelodysplastic/myeloproliferative neoplasm, not otherwise specified

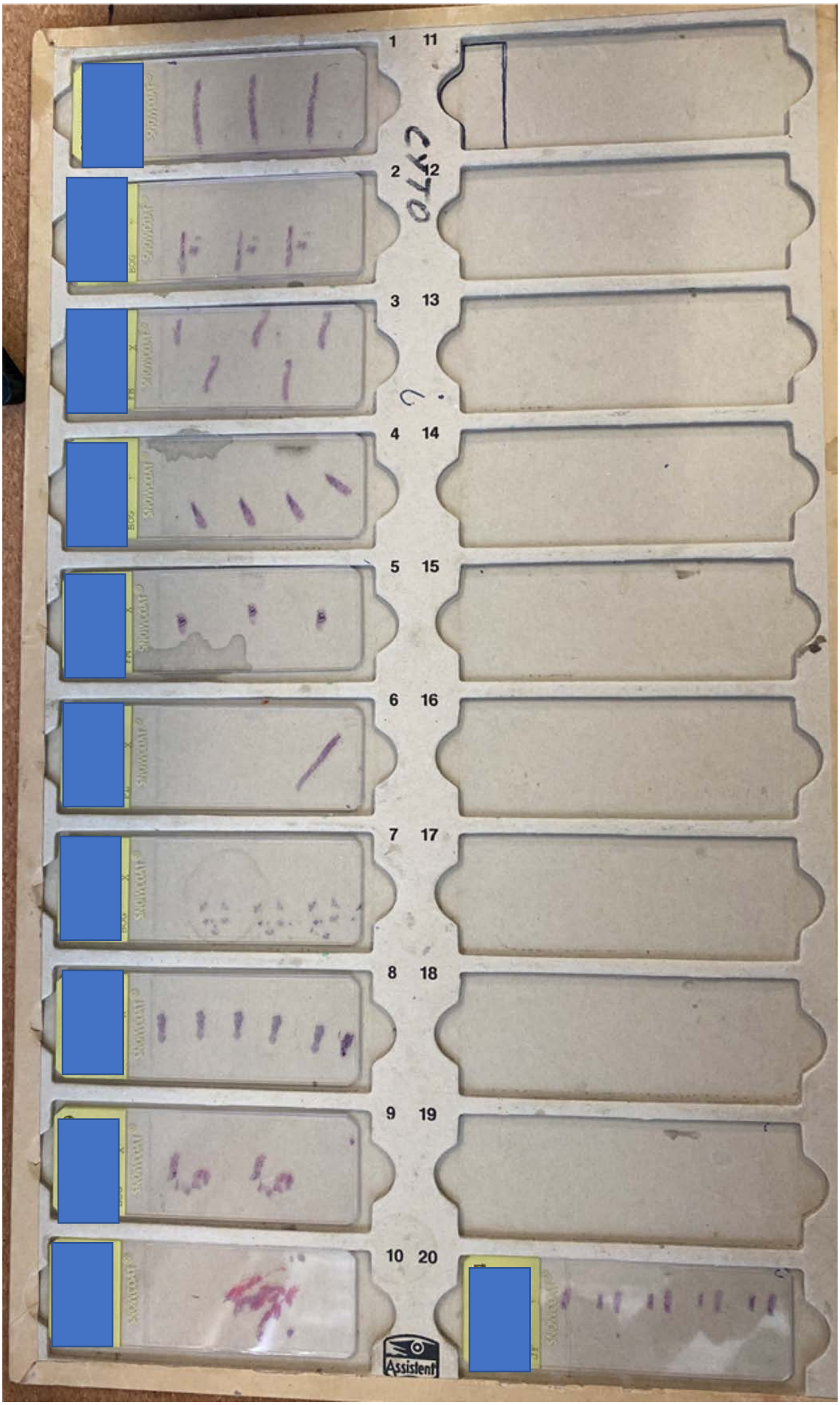
Premalignant clonal cytopenias and myelodysplastic syndromes
Clonal cytopenia of undetermined significance
Myelodysplastic syndrome with mutated *SF3B1*
Myelodysplastic syndrome with del(5q)
Myelodysplastic syndrome with mutated *TP53*
Myelodysplastic syndrome, not otherwise specified (MDS, NOS)
MDS, NOS without dysplasia
MDS, NOS with single lineage dysplasia
MDS, NOS with multilineage dysplasia
Myelodysplastic syndrome with excess blasts
Myelodysplastic syndrome/acute myeloid leukemia (MDS/AML)
MDS/AML with mutated *TP53*
MDS/AML with myelodysplasia-related gene mutations
MDS/AML with myelodysplasia-related cytogenetic abnormalities
MDS/AML, not otherwise specified
Pediatric and/or germline mutation-associated disorders
Juvenile myelomonocytic leukemia
Juvenile myelomonocytic leukemia-like neoplasms
Noonan syndrome-associated myeloproliferative disorder
Refractory cytopenia of childhood
Hematologic neoplasms with germline predisposition
Acute myeloid leukemias (Tables 25 and 26)
Myeloid proliferations associated with Down syndrome
Blastic plasmacytoid dendritic cell neoplasm

Acute leukemia of ambiguous lineage
Acute undifferentiated leukemia
Mixed phenotype acute leukemia (MPAL) with t(9;22)(q34.1;q11.2); *BCR::ABL1*
MPAL, with t(v;11q23.3); *KMT2A* rearranged
MPAL, B/myeloid, NOS
MPAL, T/myeloid, NOS
B-lymphoblastic leukemia/lymphoma (Tables 27 and 28; supplemental Table 6)
T-lymphoblastic leukemia/lymphoma (Table 27; supplemental Table 6)

- WHO 5th edition, 2022

2. Myeloid proliferations and neoplasms

Myeloid precursor lesions
Clonal Haematopoiesis
Introduction
Clonal haematopoiesis
Clonal cytopenias of undetermined significance
Myeloproliferative neoplasms
Myeloproliferative neoplasms
Introduction
Chronic myeloid leukaemia
Chronic neutrophilic leukaemia
Chronic eosinophilic leukaemia
Polycythaemia vera
Essential thrombocythaemia
Primary myelofibrosis
Juvenile myelomonocytic leukaemia
Myeloproliferative neoplasm, NOS
Mastocytosis
Introduction
Cutaneous mastocytosis
Systemic mastocytosis
Mast cell sarcoma
Myelodysplastic neoplasms
Introduction
Myelodysplastic neoplasms, with defining genetic abnormalities
Myelodysplastic neoplasm with low blasts and 5q deletion
Myelodysplastic neoplasm with low blasts and SF3B1 mutation
Myelodysplastic neoplasm with biallelic TP53 inactivation
Myelodysplastic neoplasms, morphologically defined
Myelodysplastic neoplasm with low blasts
Myelodysplastic neoplasm, hypoplastic
Myelodysplastic neoplasm with increased blasts
Myelodysplastic neoplasms of childhood
Childhood myelodysplastic neoplasm with low blasts
Childhood myelodysplastic neoplasm with increased blasts
Myelodysplastic/myeloproliferative neoplasms
Introduction
Chronic myelomonocytic leukaemia
Myelodysplastic/myeloproliferative neoplasm with neutrophilia
Myelodysplastic/myeloproliferative neoplasm with SF3B1 mutation and thrombocytosis
Myelodysplastic/myeloproliferative neoplasm, NOS
Acute myeloid leukaemia
Introduction
Acute myeloid leukaemia with defining genetic abnormalities
Acute promyelocytic leukaemia with PML::RARA fusion
Acute myeloid leukaemia with RUNX1::RUNX1T1 fusion
Acute myeloid leukaemia with CBFB::MYH11 fusion
Acute myeloid leukaemia with DEK::NUP214 fusion
Acute myeloid leukaemia with RBM15::MRTFA fusion
Acute myeloid leukaemia with BCR::ABL1 fusion
Acute myeloid leukaemia with KMT2A rearrangement
Acute myeloid leukaemia with MECOM rearrangement
Acute myeloid leukaemia with NUP98 rearrangement
Acute myeloid leukaemia with NPM1 mutation
Acute myeloid leukaemia with CEBPA mutation
Acute myeloid leukaemia, myelodysplasia-related
Acute myeloid leukaemia with other defined genetic alterations
Acute myeloid leukaemia, defined by differentiation
Acute myeloid leukaemia with minimal differentiation
Acute myeloid leukaemia without maturation
Acute myeloid leukaemia with maturation
Acute basophilic leukaemia
Acute myelomonocytic leukaemia
Acute monocytic leukaemia
Acute erythroid leukaemia
Acute megakaryoblastic leukaemia
Myeloid sarcoma
Myeloid sarcoma
Myeloid neoplasms, secondary
Myeloid neoplasms and proliferations associated with antecedent or predisposing conditions
Introduction
Myeloid neoplasm post cytotoxic therapy
Myeloid neoplasms associated with germline predisposition
Myeloid proliferations associated with Down syndrome
Myeloid/lymphoid neoplasms
Myeloid/lymphoid neoplasms with eosinophilia and defining gene rearrangement
Introduction
Myeloid/lymphoid neoplasm with PDGFRA rearrangement
Myeloid/lymphoid neoplasm with PDGFRB rearrangement
Myeloid/lymphoid neoplasm with FGFR1 rearrangement
Myeloid/lymphoid neoplasm with JAK2 rearrangement
Myeloid/lymphoid neoplasm with FLT3 rearrangement
Myeloid/lymphoid neoplasm with ETV6::ABL1 fusion
Myeloid/lymphoid neoplasms with other tyrosine kinase gene fusions
Acute leukaemias of mixed or ambiguous lineage
Introduction
Acute leukaemia of ambiguous lineage with defining genetic abnormalities
Mixed-phenotype acute leukaemia with BCR::ABL1 fusion
Mixed-phenotype acute leukaemia with KMT2A rearrangement
Acute leukaemia of ambiguous lineage with other defined genetic alterations
Acute leukaemia of ambiguous lineage, immunophenotypically defined
Mixed-phenotype acute leukaemia, B/myeloid
Mixed-phenotype acute leukaemia, T/myeloid
Mixed-phenotype acute leukaemia, rare types
Acute leukaemia of ambiguous lineage, NOS
Acute undifferentiated leukaemia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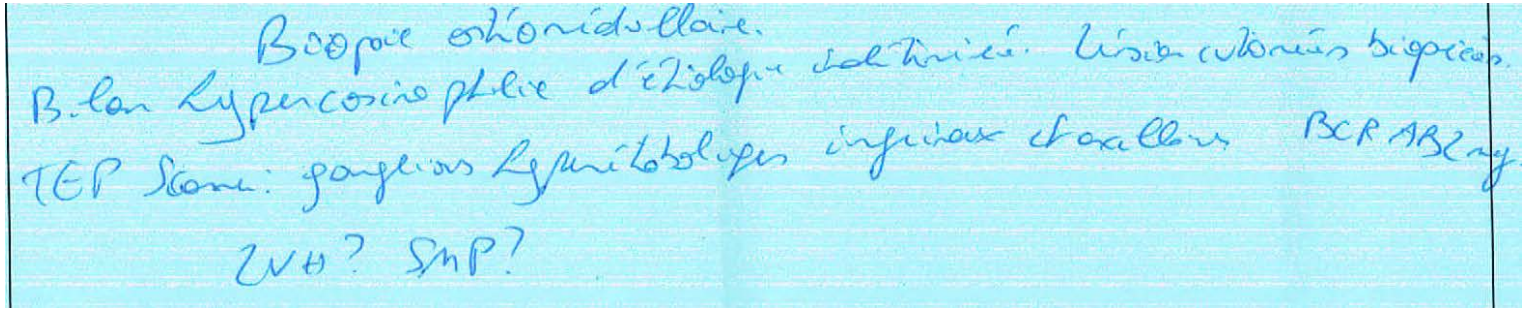
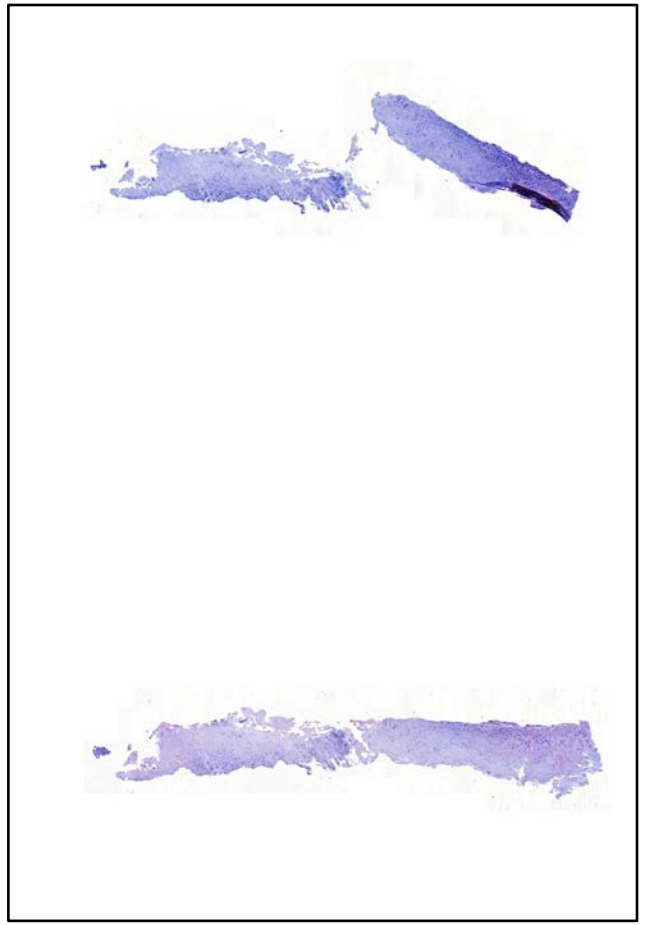
Assistent

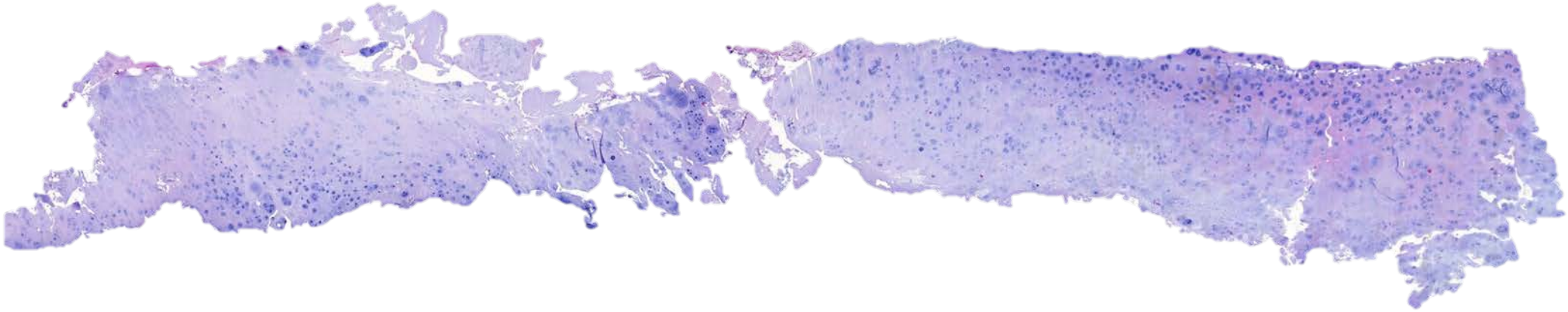
Case 1 : 76 y, M

- Bone marrow biopsy
- Hypereosinophilia of unknown aetiology
- Biopsied cutaneous lesions
- BCR-ABL –
- PET scan : axillary and inguinal hypermetabolic lymph nodes
- NHL ? MPN ?



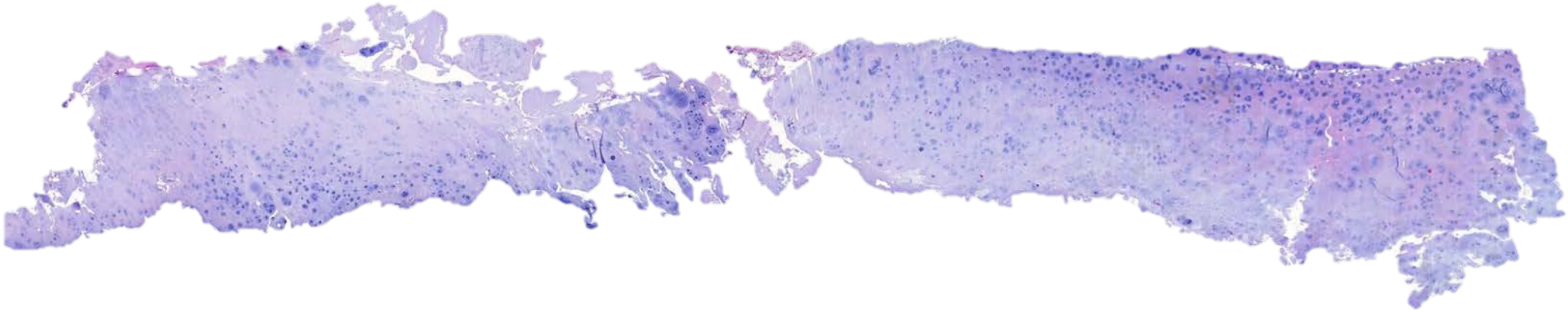


What's your diagnosis/reaction ?



- Non contributory
- Crying in your office
- Avoiding phone calls asking you if you're sure because the patient is very difficult to biopsy
- All of the above

What's your diagnosis/reaction ?

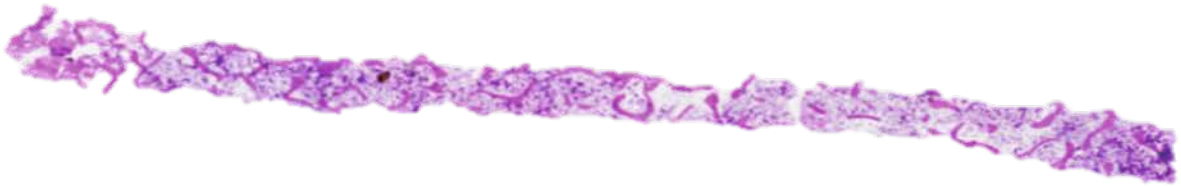


- **Non contributory**
- **Crying in your office**
- **Avoiding phone calls asking you if you're sure because the patient is very difficult to biopsy**
- **All of the above**

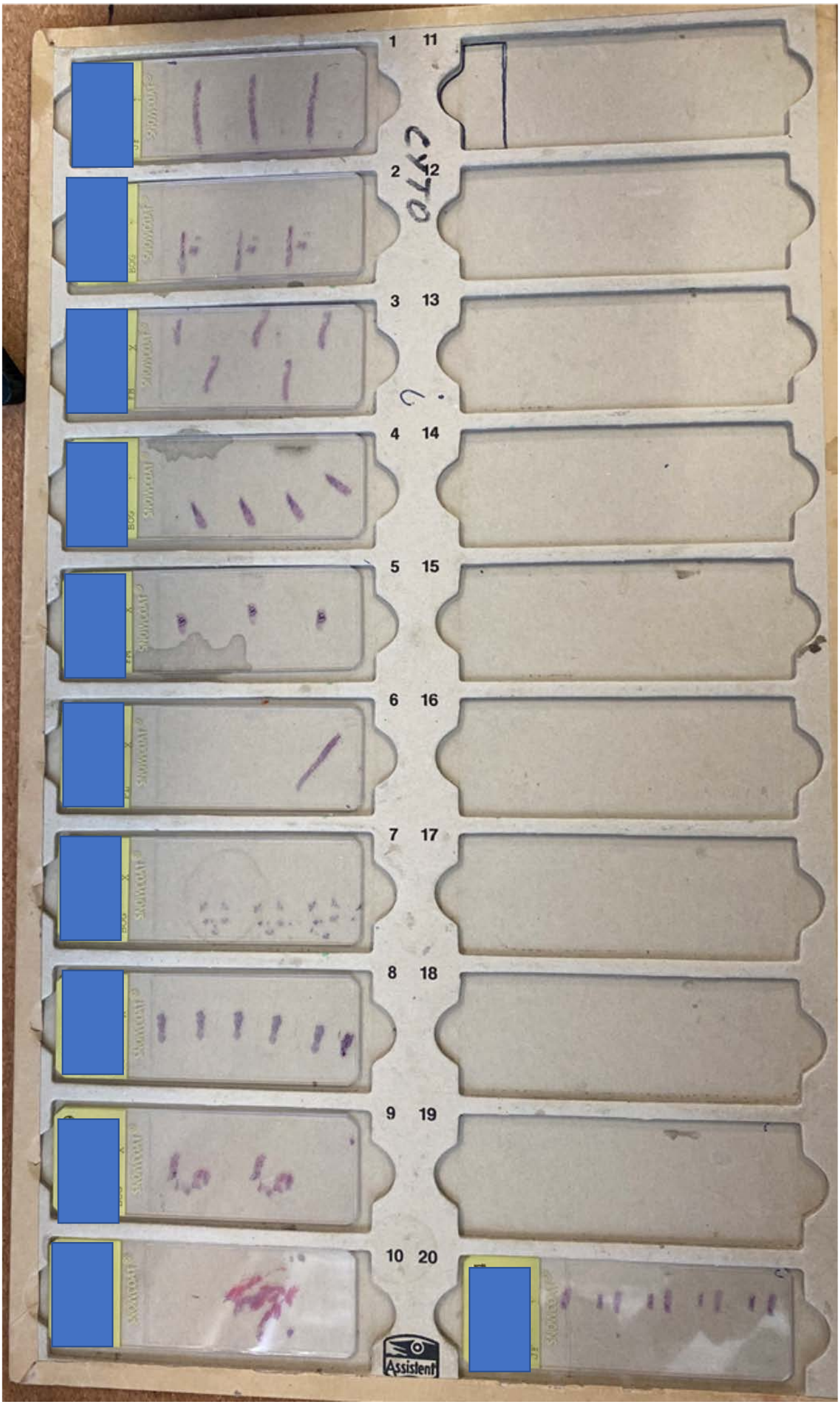
BM trephine biopsy adequacy

Collection procedure

- **11-gauge needle** AT LEAST
- If osteopaenic, a 8-gauge needle allows the collection of an intact core biopsy with minimal crush artifact
- 13-gauge biopsy needle for paediatric patients



- Adequate core biopsy,
  - At least 1.6 cm to 2 cm long
    - Prior to fixation
    - Exclusive of cortical bone, cartilage, or periosteum
    - **Free of crush artifact or interstitial hemorrhage or fragmentation**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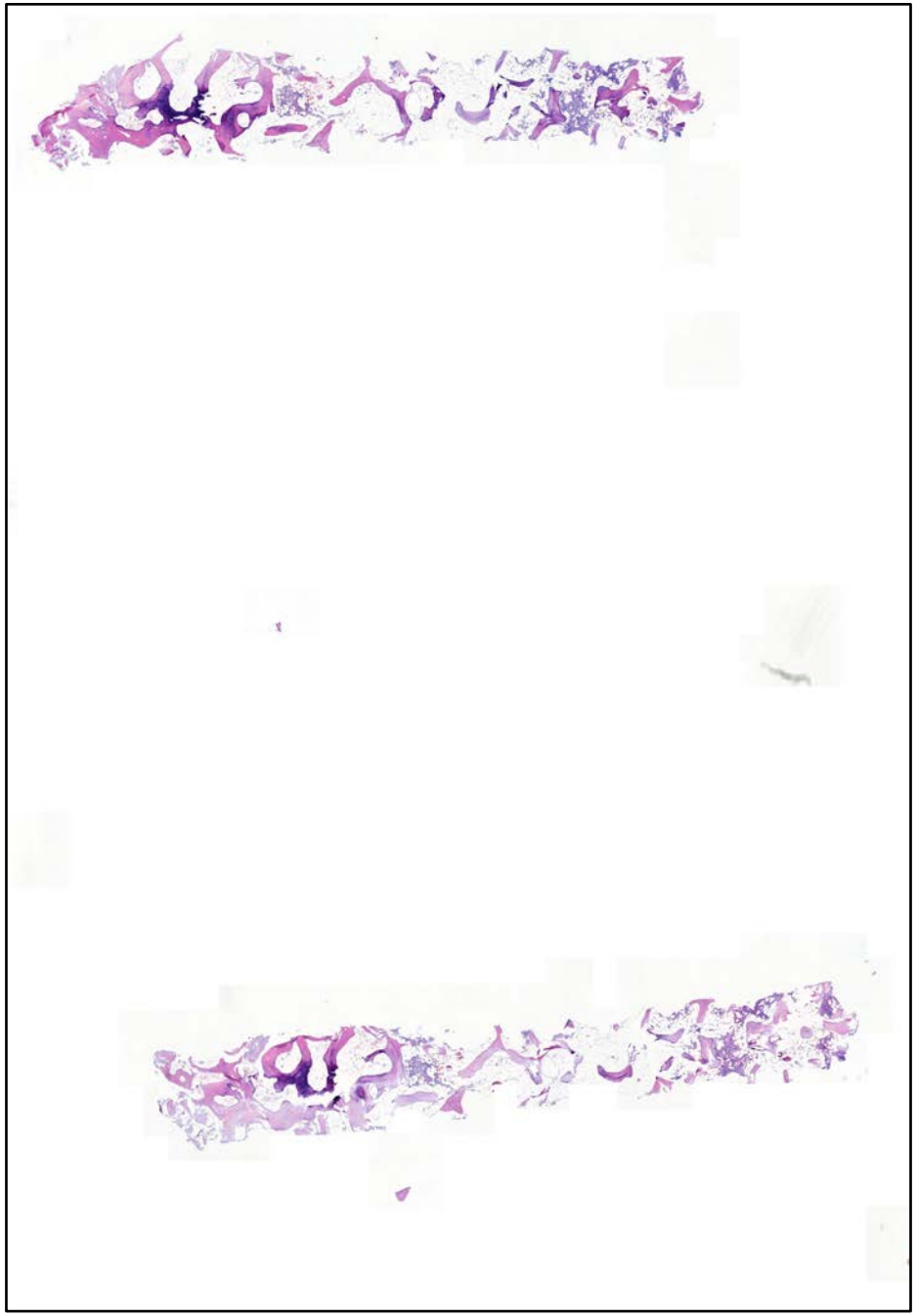
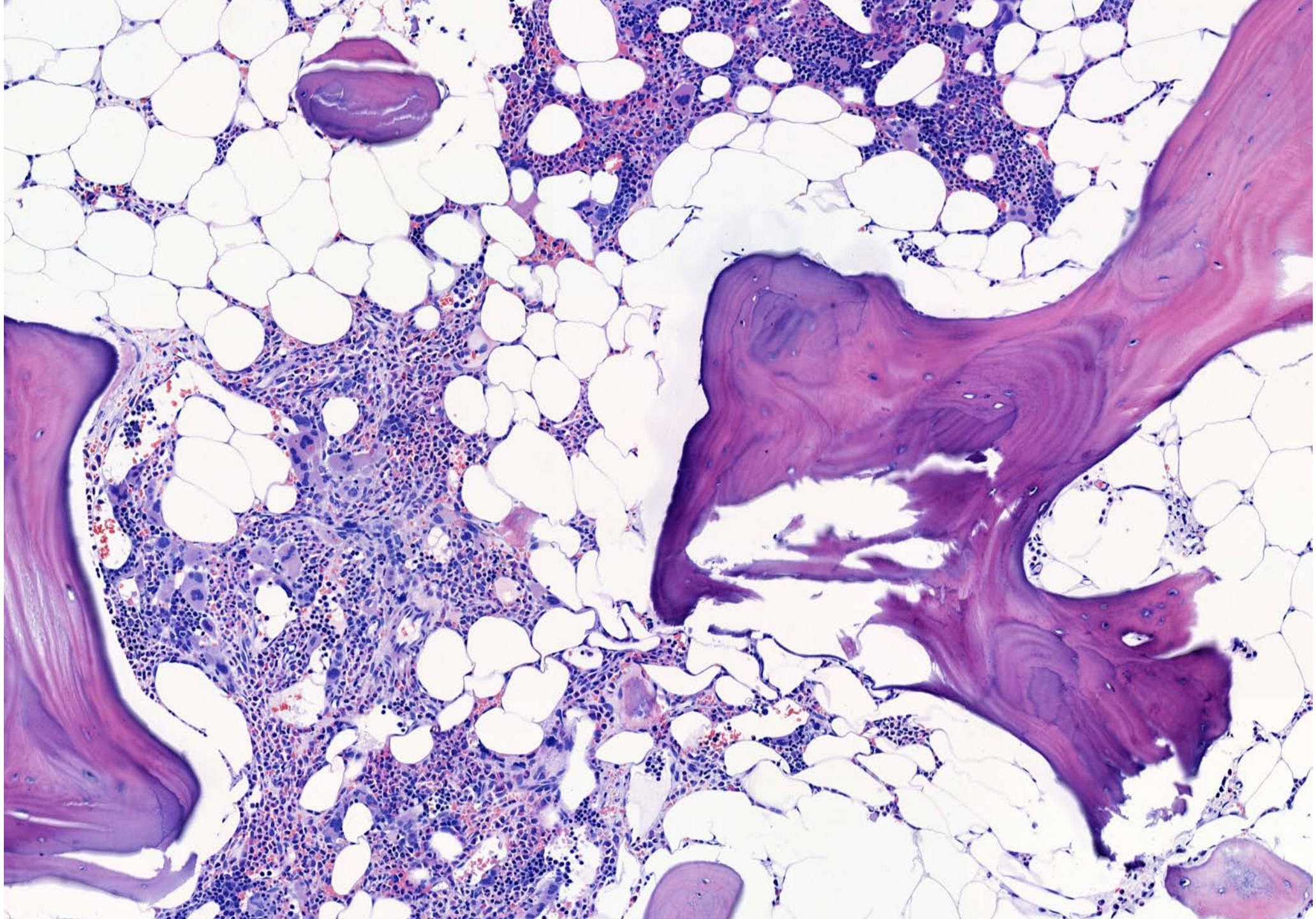
Assistent

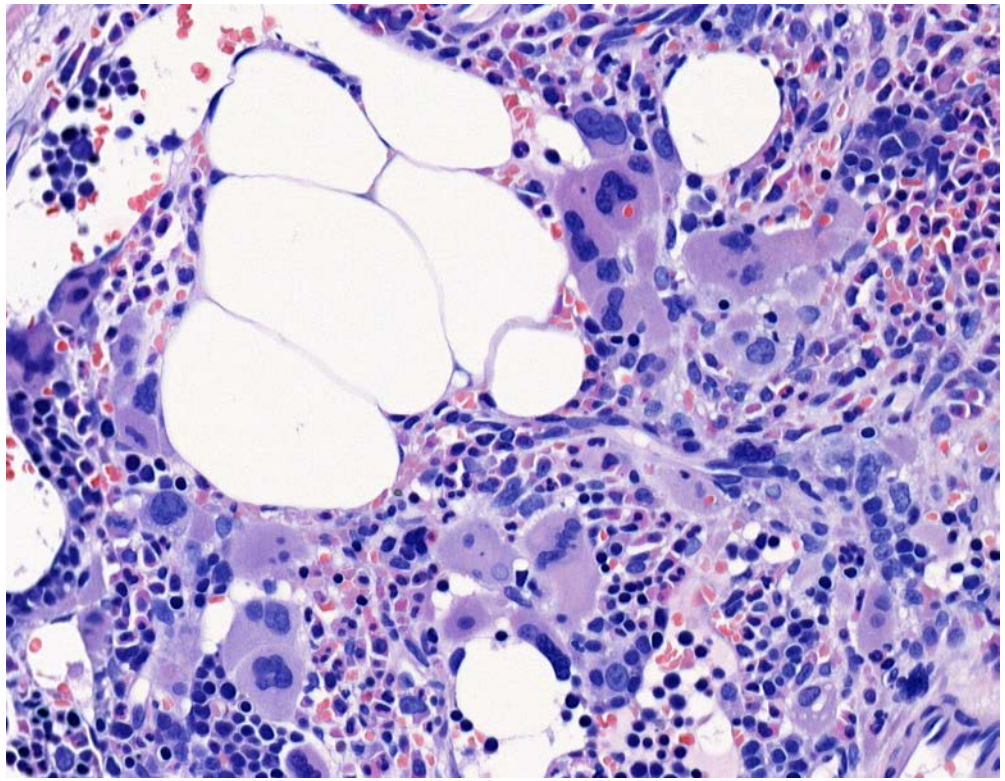
Case 2 : 66 y, M

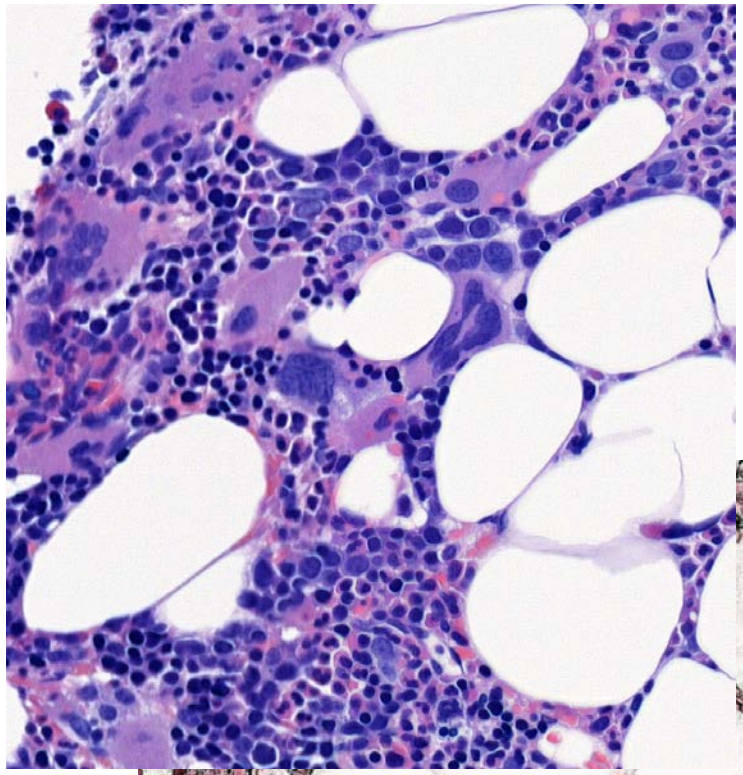
- JAK2 +
- Non contributory
- Search for associated fibrosis

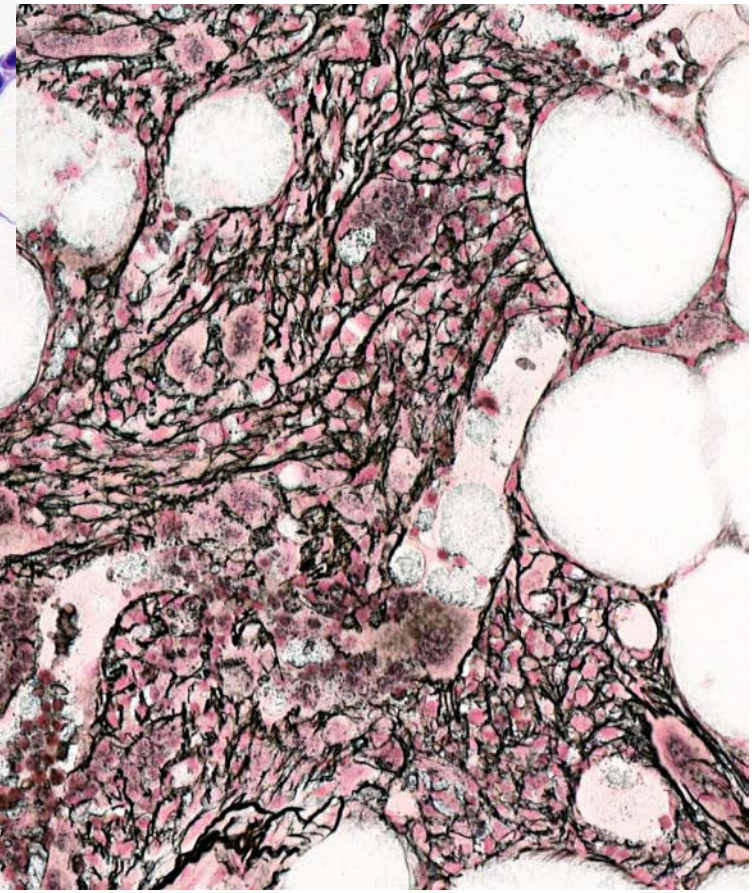
JAK 2 ⊕.
non contributive → ctrl pour recherche fibrose associé.
Merci.

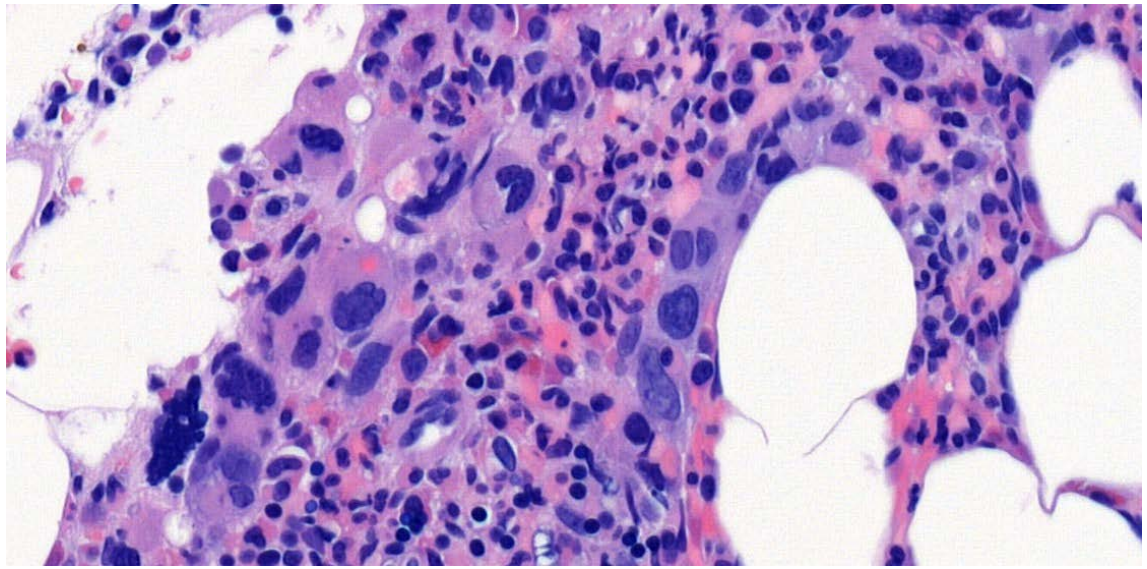












What’s your diagnosis ?

- Essential Thrombocythaemia
- Primary myelofibrosis
  - Overt
  - Pre-fibrotic
- Polycythaemia vera
- Please correlate with clinical informations
- Crying in your office (again)
- All of the above

What’s your diagnosis ?

- Essential Thrombocythaemia
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  - **Overt**
  - Pre-fibrotic
- Polycythaemia vera
- Please correlate with clinical informations
- Crying in your office (again)
- All of the above

Myeloproliferative neoplasms

- Clonal hematopoietic disorders

- Characterized by proliferation of cells of one or more of the myeloid lineages; erythroid, granulocytic, or megakaryocytic

- Initially, the proliferation in the bone marrow is effective and associated with maturation of the neoplastic cells

- Leads to increased numbers of mature granulocytes, red blood cells (RBCs), and platelets in the peripheral blood

- Splenomegaly and hepatomegaly are common and caused by the sequestration of excess blood cells, extramedullary hematopoiesis or both in these organs.

World Health Organization Classification of Myeloproliferative Neoplasms

- Chronic myeloid leukemia, *BCR-ABL1* positive
  - Chronic neutrophilic leukemia
- Polycythemia vera
- Primary myelofibrosis
- Essential thrombocythemia
  - Chronic eosinophilic leukemia, not otherwise specified*
  - Myeloproliferative neoplasm, unclassifiable

From Swerdlow SH, Campo E, Harris NL, et al, eds. *WHO Classification of Tumours of Haemato[illegible] and Lymphoid Tissues.* Revised 4th ed. Lyon, France: IARC Press; 2017.

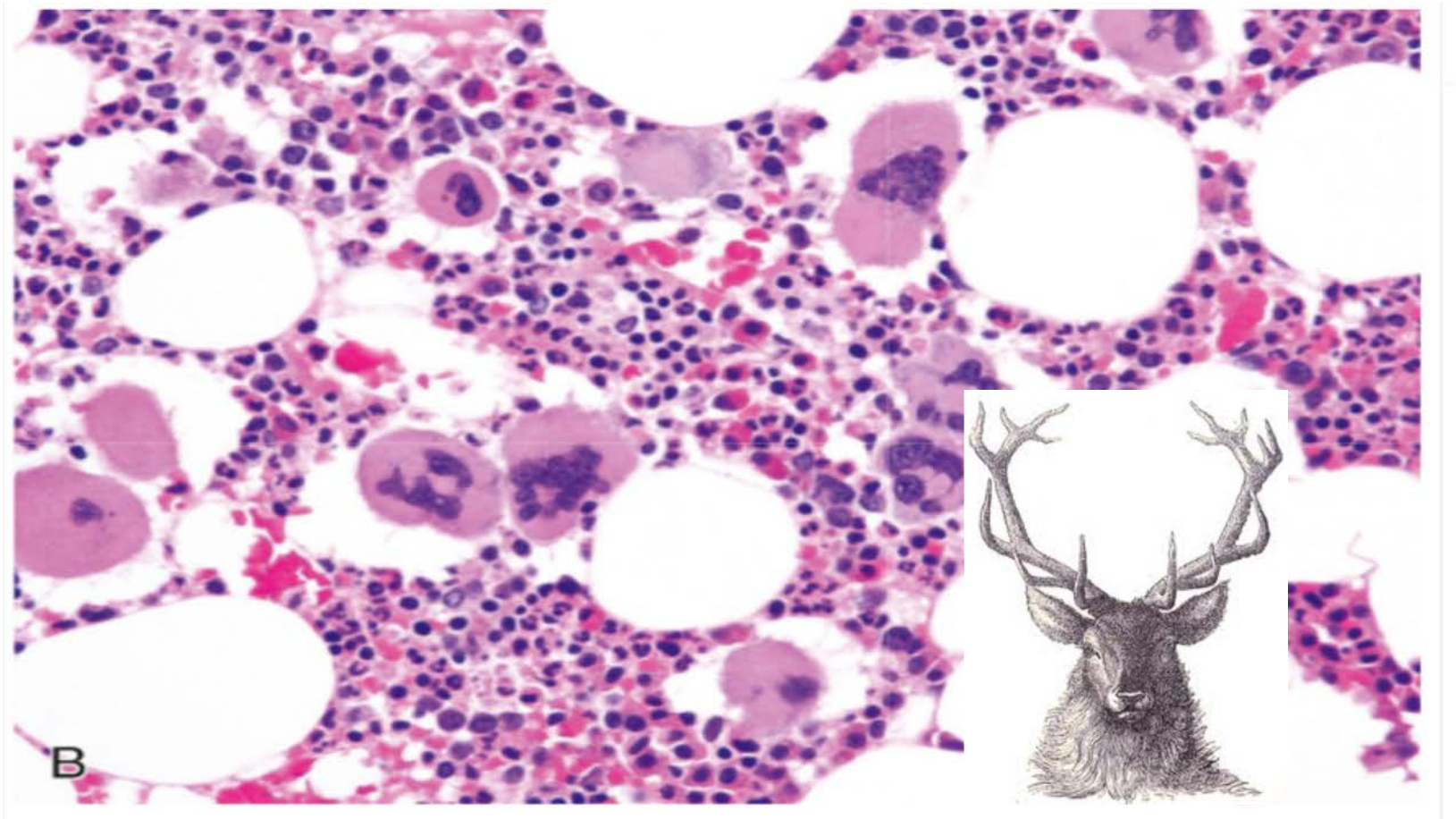
MPNs
Chronic myeloid leukemia
Polycythemia vera
Essential thrombocythemia
Primary myelofibrosis
Early/prefibrotic primary myelofibrosis
Overt primary myelofibrosis
Chronic neutrophilic leukemia
Chronic eosinophilic leukemia, not otherwise specified
MPN, unclassifiable

Essential Thrombocythaemia : WHO 2022

The diagnosis of essential thrombocythaemia requires that either **all major criteria** or the **first 3 major criteria plus the minor criterion are met.**

- **Major criteria**
  - Platelet count ≥ 450 × 10^9/ L
  - Bone marrow biopsy showing proliferation mainly of the megakaryocytic lineage, with increased numbers of enlarged, mature megakaryocytes with hyperlobulated nuclei; no significant increase or left shift in neutrophil granulopoiesis or erythropoiesis; very rarely a minor (grade 1) increase in reticulin fibres
  - WHO criteria for *BCR*::*ABL1*–positive chronic myeloid leukaemia, polycythaemia vera, primary myelofibrosis, or other myeloid neoplasms are not met
  - *JAK2*, *CALR*, or *MPL* mutation
- **Minor criterion**
  - Presence of a clonal marker or
  - Exclusion of reactive thrombocytosis

ICC 2022 : one single minor criteria



Jaffe, 2017

Polycythaemia Vera : WHO 2022

The diagnosis of polycythaemia vera requires either all 3 major criteria or the first 2 major criteria plus the minor criterion.

- **Major criteria**
  - Elevated haemoglobin concentration (> 16.5 g/dL in men; > 16.0 g/dL in women) or elevated haematocrit (>49%[a] in men; >48% in women)
  - Bone marrow biopsy showing age-adjusted hypercellularity with trilineage growth (panmyelosis), including prominent erythroid, granulocytic, and megakaryocytic proliferation with pleomorphic, mature megakaryocytes (differences in size)[b]
  - Presence of *JAK2* V617F or *JAK2* exon 12 mutation
- **Minor criterion**
  - Subnormal serum erythropoietin level.

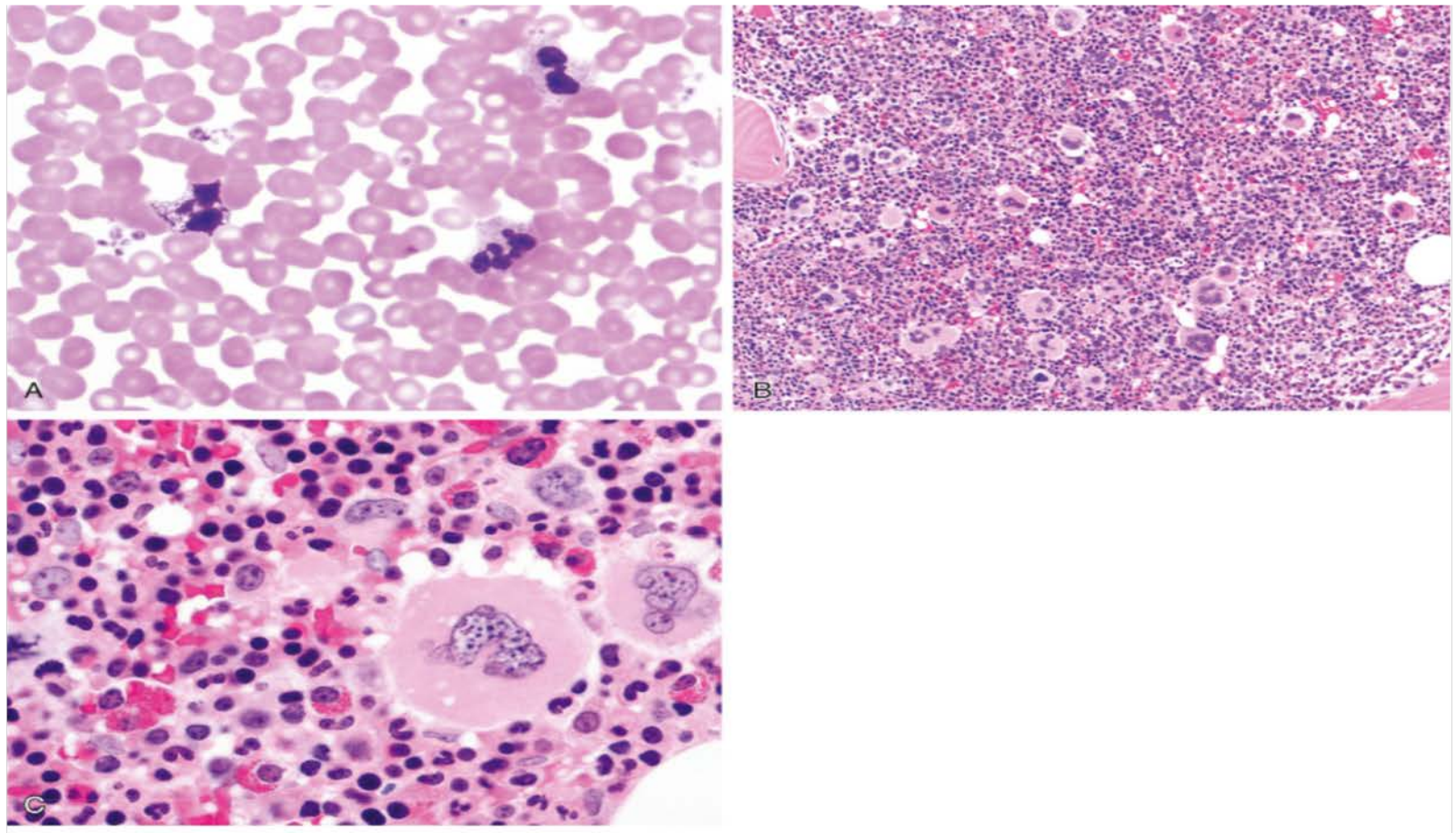
---

[a] Haematocrit for diagnosis in the absence of a *JAK2* mutation. A higher haematocrit target could be considered (e.g., 0.52) in men before further investigation may be required.

[b] Major criterion 2 (bone marrow biopsy) may not be required in patients with sustained absolute erythrocytosis (haemoglobin concentrations of > 18.5 g/dL in men or > 16.5 g/dL in women or haematocrit values of > 0.555 in men or > 0.495 in women), if major criterion 3 and the minor criterion are present.

The determination of the red cell mass with ^{51}Cr-labeled red cells allows the differentiation between true polyglobulia and pseudopolyglobulia. This is not a method for routine clinical use.

ICC 2022 : Bone marrow biopsy may not be required in patients with sustained absolute erythrocytosis (hemoglobin concentrations of >18.5 g/dL in men or >16.5 g/dL in women and hematocrit values of >55.5% in men or >49.5% in women) and the presence of a *JAK2* V617F or *JAK2* exon 12 mutation.



Jaffe, 2017

Primary Myelofibrosis : WHO 2022

The diagnosis of pre-fibrotic primary myelofibrosis requires that all 3 major criteria and at least 1 minor criterion are met.

Major criteria

1. Megakaryocytic proliferation and atypia, without reticulin fibrosis grade > 1[a], accompanied by increased age-adjusted bone marrow cellularity, granulocytic proliferation, and (often) decreased erythropoiesis
2. WHO criteria for *BCR-ABL1*–positive chronic myeloid leukaemia, polycythaemia vera, essential thrombocythaemia, myelodysplastic syndromes, or other myeloid neoplasms are not met
3. *JAK2*, *CALR*, or *MPL* mutation

   OR

   Presence of another clonal marker[b]

   OR

   Absence of minor reactive bone marrow reticulin fibrosis[c]

Minor criteria

Presence of at least one of the following, confirmed in 2 consecutive determinations:

- Anaemia not attributed to a comorbid condition
- Leukocytosis ≥ 11 × 10^9/L
- Splenomegaly detected clinically and/or by imaging
- Lactate dehydrogenase level above the upper limit of the institutional reference range
- Leukoerythroblastosis

[a] See Table <<30704>>

[b] In the absence of any of the 3 major clonal mutations, a search for other mutations associated with myeloid neoplasms (e.g. *ASXL1*, *EZH2*, *TET2*, *IDH1*, *IDH2*, *SRSF2*, and *SF3B1* mutations) may be of help in determining the clonal nature of the disease.

[c] Minor (grade 1) reticulin fibrosis secondary to infection, autoimmune disorder or other chronic inflammatory conditions, hairy cell leukaemia or another lymphoid neoplasm, metastatic malignancy, or toxic (chronic) myelopathies.

The diagnosis of overt primary myelofibrosis requires that all 3 major criteria and at least 1 minor criterion are met.

Major criteria

1. Megakaryocytic proliferation and atypia, accompanied by reticulin and/or collagen fibrosis grades 2 or 3[a]
2. WHO criteria for essential thrombocythaemia, polycythaemia vera, *BCR-ABL1*–positive chronic myeloid leukaemia, myelodysplastic syndrome, or other myeloid neoplasms[b] are not met
3. JAK2, CALR, or MPL mutation

   OR

   Presence of another clonal marker[C]

   OR

   Absence of reactive myelofibrosis[d]

Minor criteria

Presence of at least one of the following, confirmed in 2 consecutive determinations:

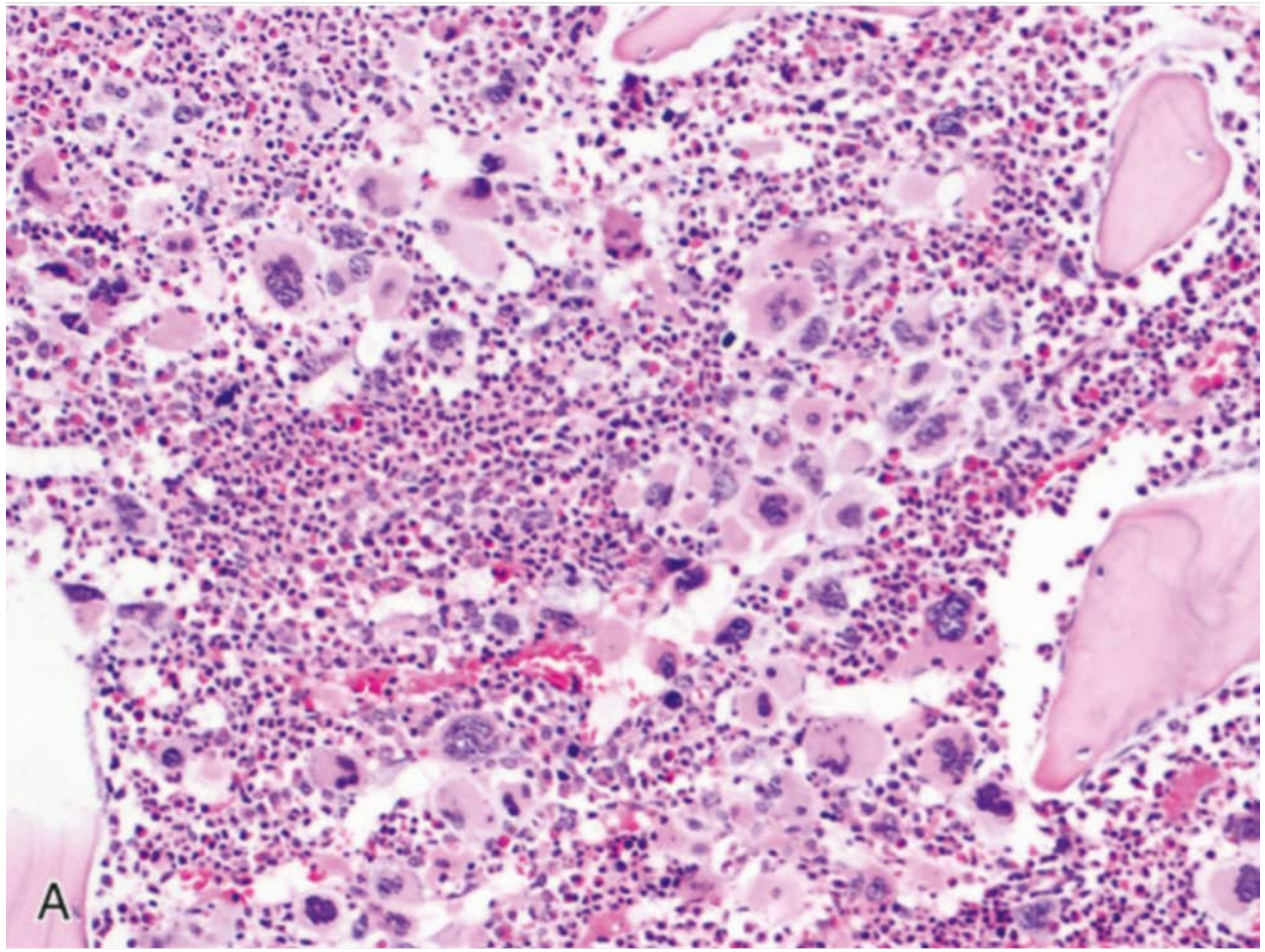
- Anaemia not attributed to a comorbid condition
- Leukocytosis ≥ 11 × 10^9/L
- Splenomegaly detected clinically and/or by imaging
- Lactate dehydrogenase level above the upper limit of the institutional reference range
- Leukoerythroblastosis

[a] See Table <<30704>>

[b] Myeloproliferative neoplasms can be associated with monocytosis or they can develop it during the course of the disease; these cases may mimic chronic myelomonocytic leukaemia (CMML); in these rare instances,
a history of MPN excludes CMML, whereas the presence of MPN features in the bone marrow and/or MPN-associated mutations (in *JAK2*, *CALR*, or *MPL*) tend to support the diagnosis of MPN with monocytosis rather than CMML.

[c] In the absence of any of the 3 major clonal mutations, a search for other mutations associated with myeloid neoplasms (e.g. *ASXL1*, *EZH2*, *TET2*, *IDH1*, *IDH2*, *SRSF2*, and *SF3B1* mutations) may be of help in determining the clonal nature of the disease.

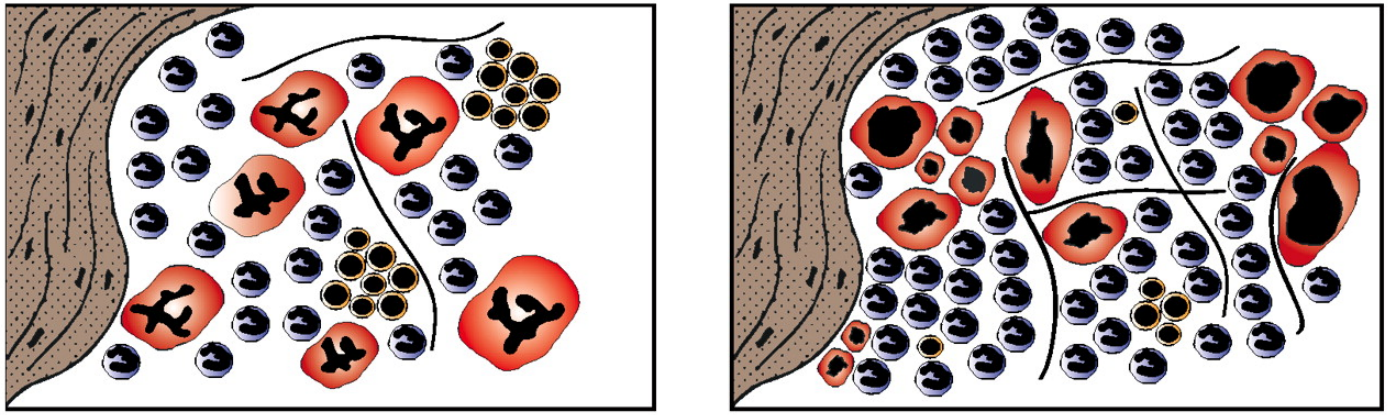
[d] Bone marrow fibrosis secondary to infection, autoimmune disorder or another chronic inflammatory condition, hairy cell leukaemia or another lymphoid neoplasm, metastatic malignancy, or toxic (chronic) myelopathy.



Jaffe, 2017

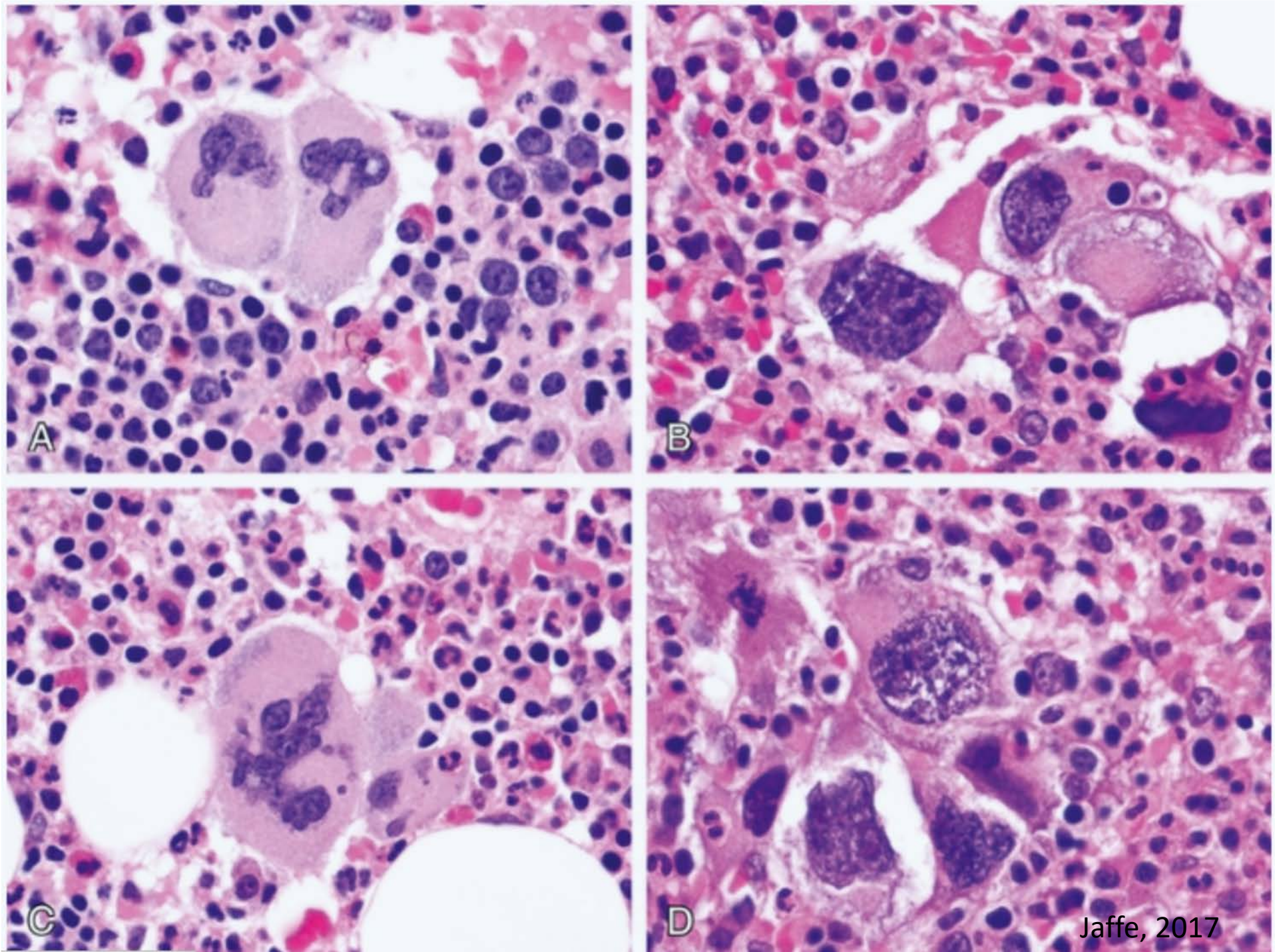
Diagnostic criteria of distinctive value regarding WHO-defined ET (left) versus early-prefibrotic stage of PMF (right), including standardized morphologic features (Table 1 contains more details), allowing the generation of characteristic histologic BM patterns

ET	PMF (early-prefibrotic stage)
• no or only slight increase in age-matched cellularity	• marked increase in age-matched cellularity
• no significant increase in granulo- and erythropoiesis	• pronounced proliferation of granulopoiesis and reduction of erythroid precursors
• prominent large to giant mature megakaryocytes with hyperlobulated or deeply folded nuclei, dispersed or loosely clustered in the marrow space	• dense or loose clustering and frequent endosteal translocation of medium sized to giant megakaryocytes showing hyperchromatic, hypolobulated, bulbous, or irregularly folded nuclei and an aberrant nuclear/cytoplasmic ratio
• no or very rarely minor increase in reticulin fibers	• no or no significant increase in reticulin fibers

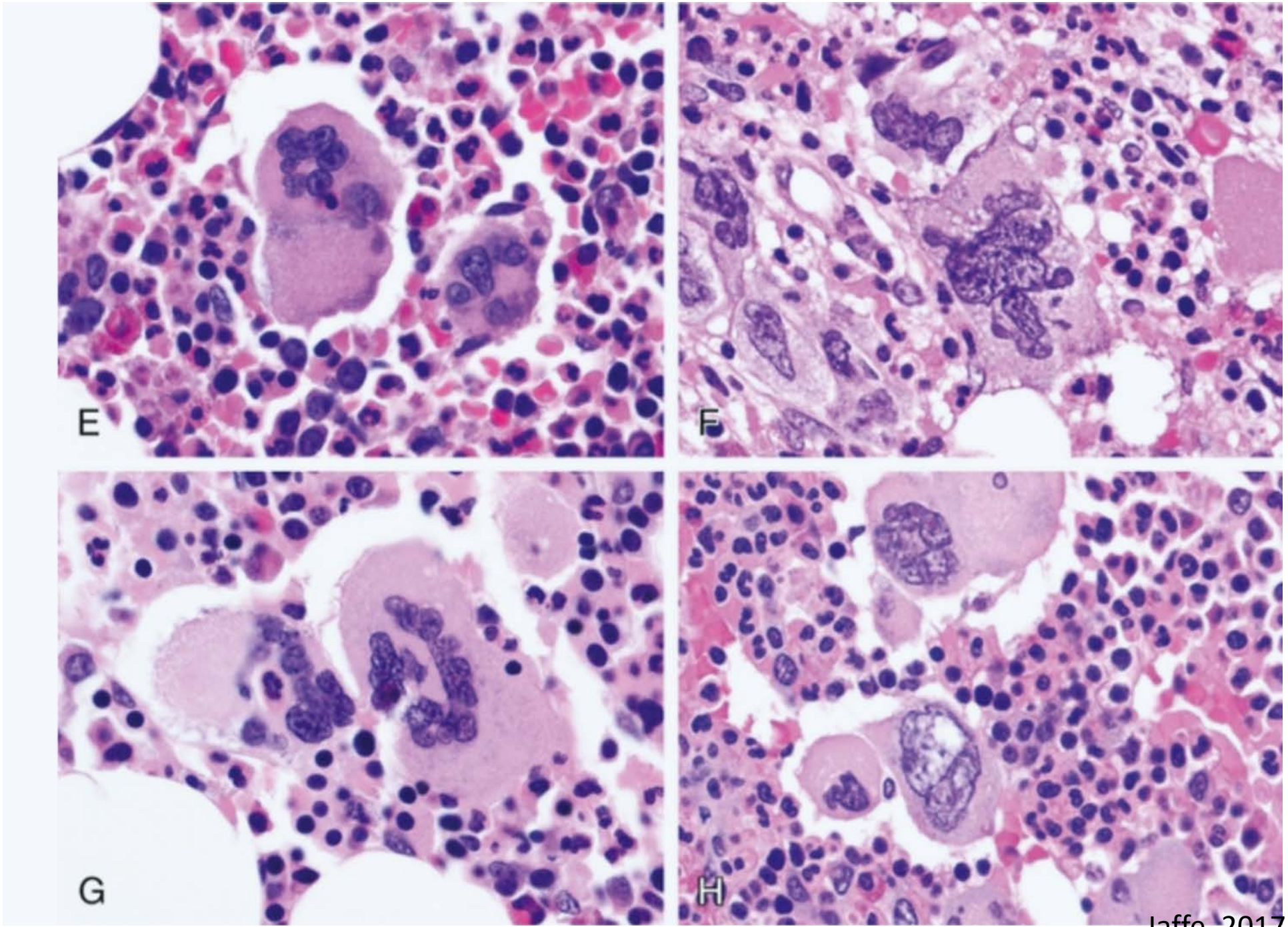


Megakaryopoiesis; Granulopoiesis; Erythropoiesis; Reticulin fibers

Jürgen Thiele et al. Blood 2011;117:5710-5718

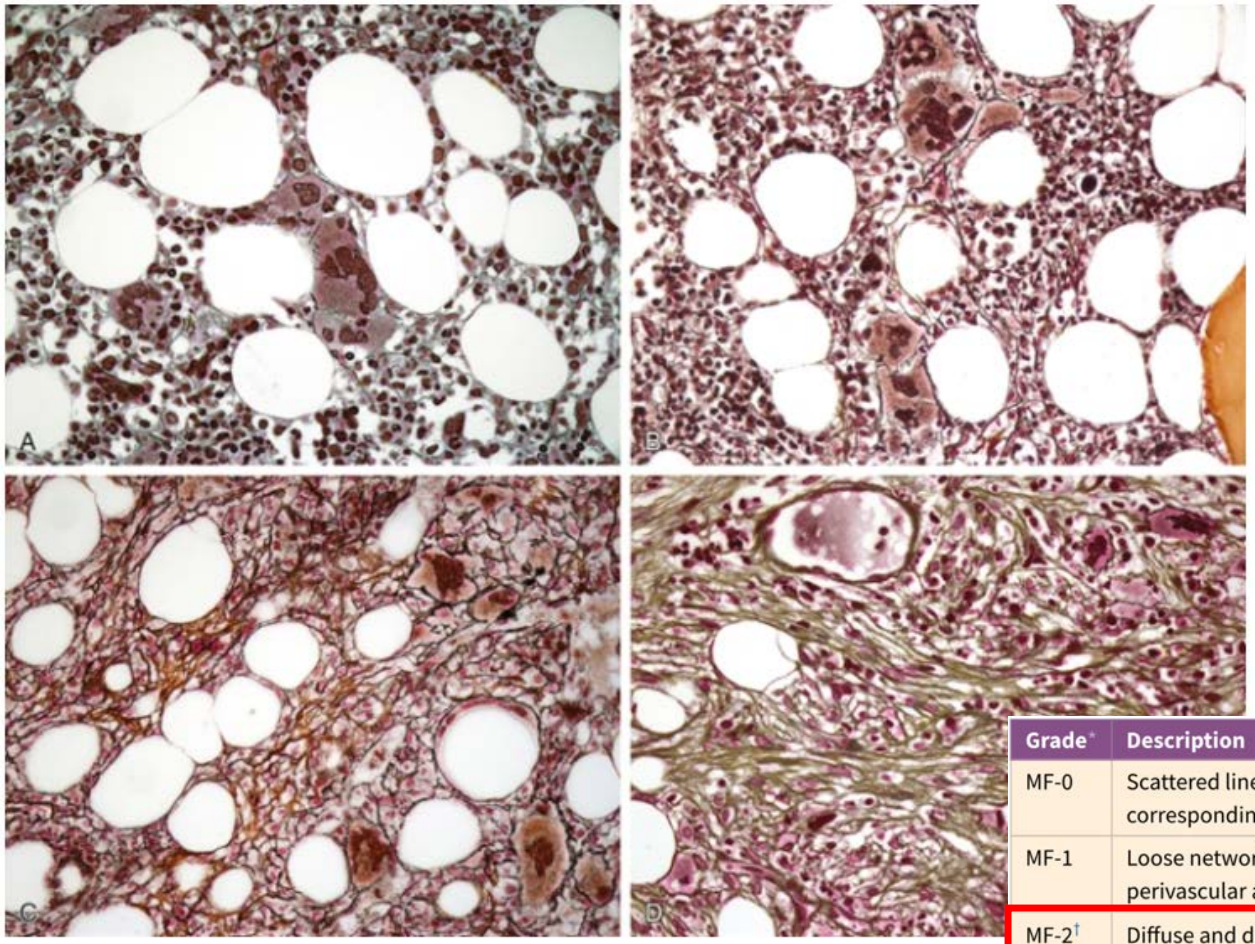


Jaffe, 2017



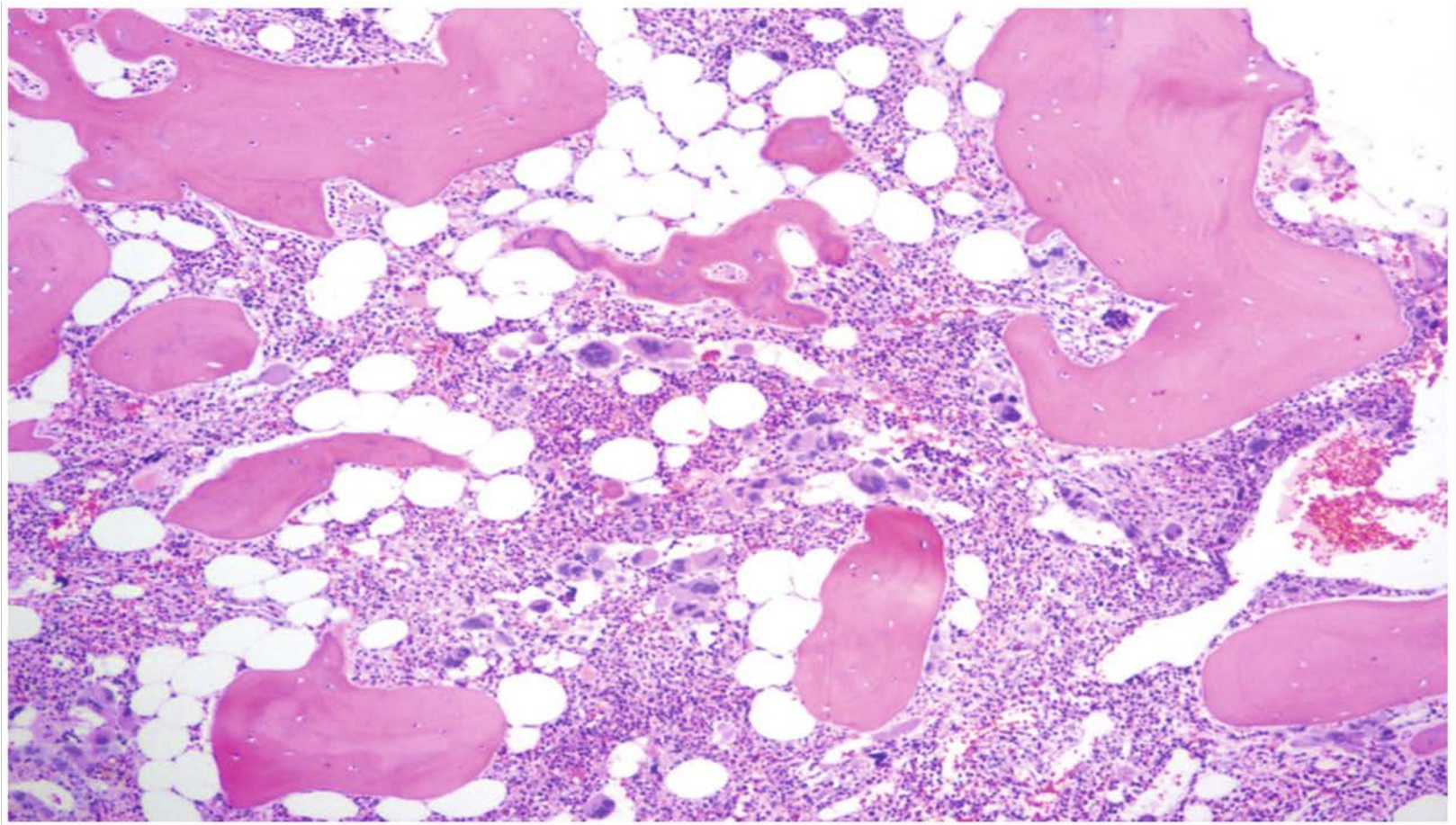
Jaffe, 2017

Bone marrow fibrosis

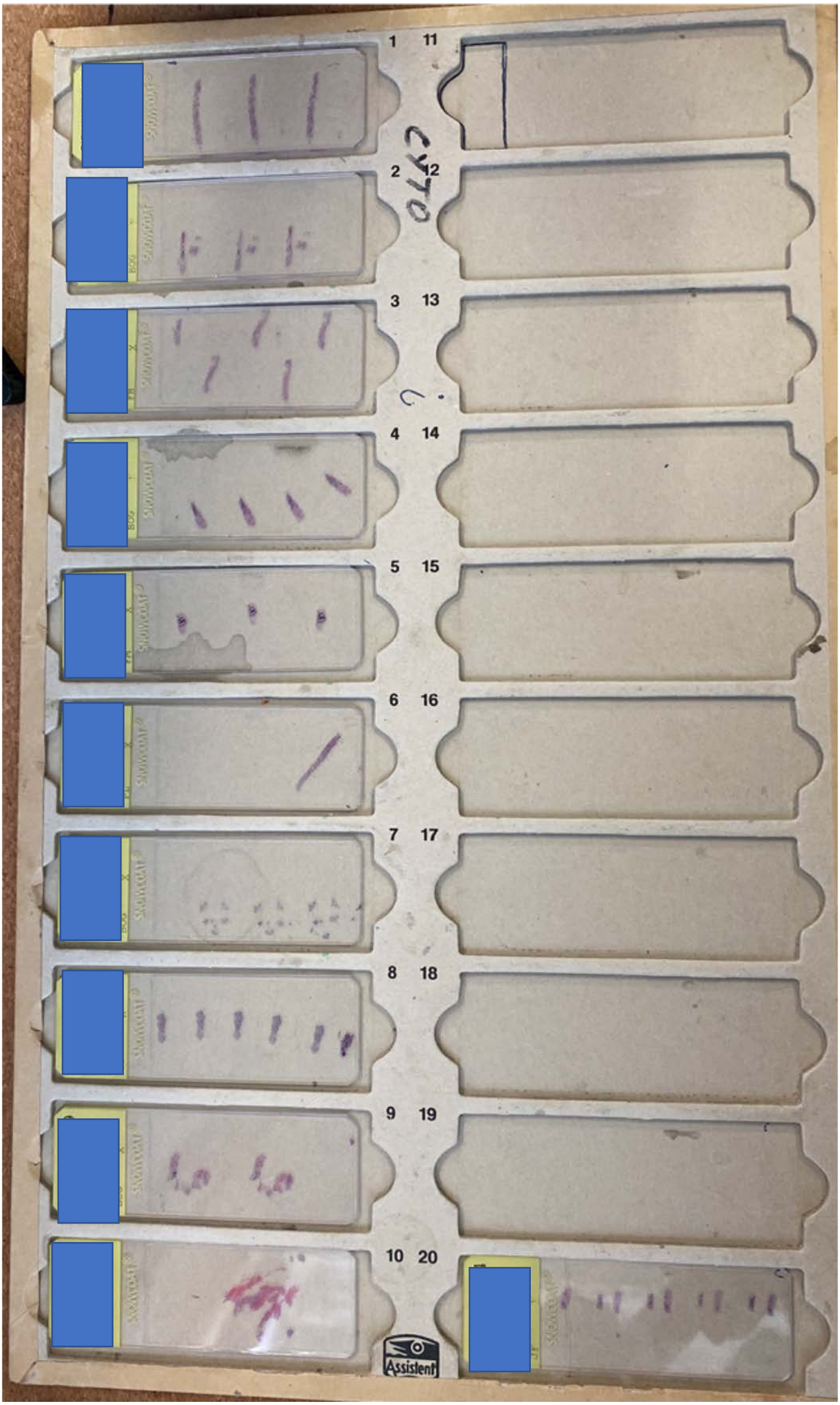


European consensus on grading bone marrow fibrosis and assessment of cellularity. *J.Thiele et al. Haematologica 2005; 90:1128-1132*

Grade*	Description
MF-0	Scattered linear reticulin fibers with no intersections (crossovers), corresponding to normal bone marrow
MF-1	Loose network of reticulin with many intersections, especially in perivascular areas
MF-2†	Diffuse and dense increase in reticulin fibers with extensive intersections, occasionally with focal bundles of thick fibers mostly consistent with collagen or focal osteosclerosis
MF-3†	Diffuse and dense increase in reticulin with extensive intersections and coarse bundles of thick fibers consistent with collagen, usually associated with osteosclerosis



Jaffe, 2017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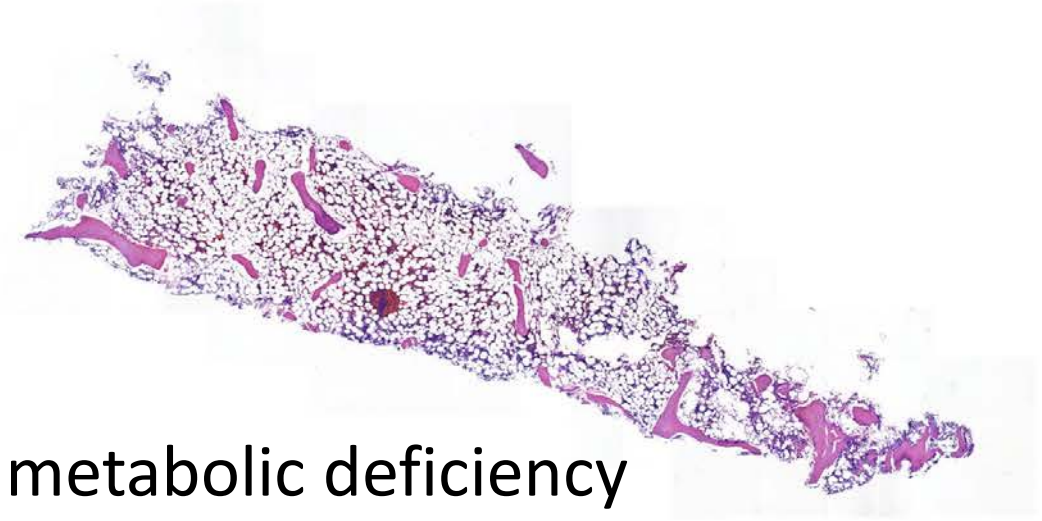
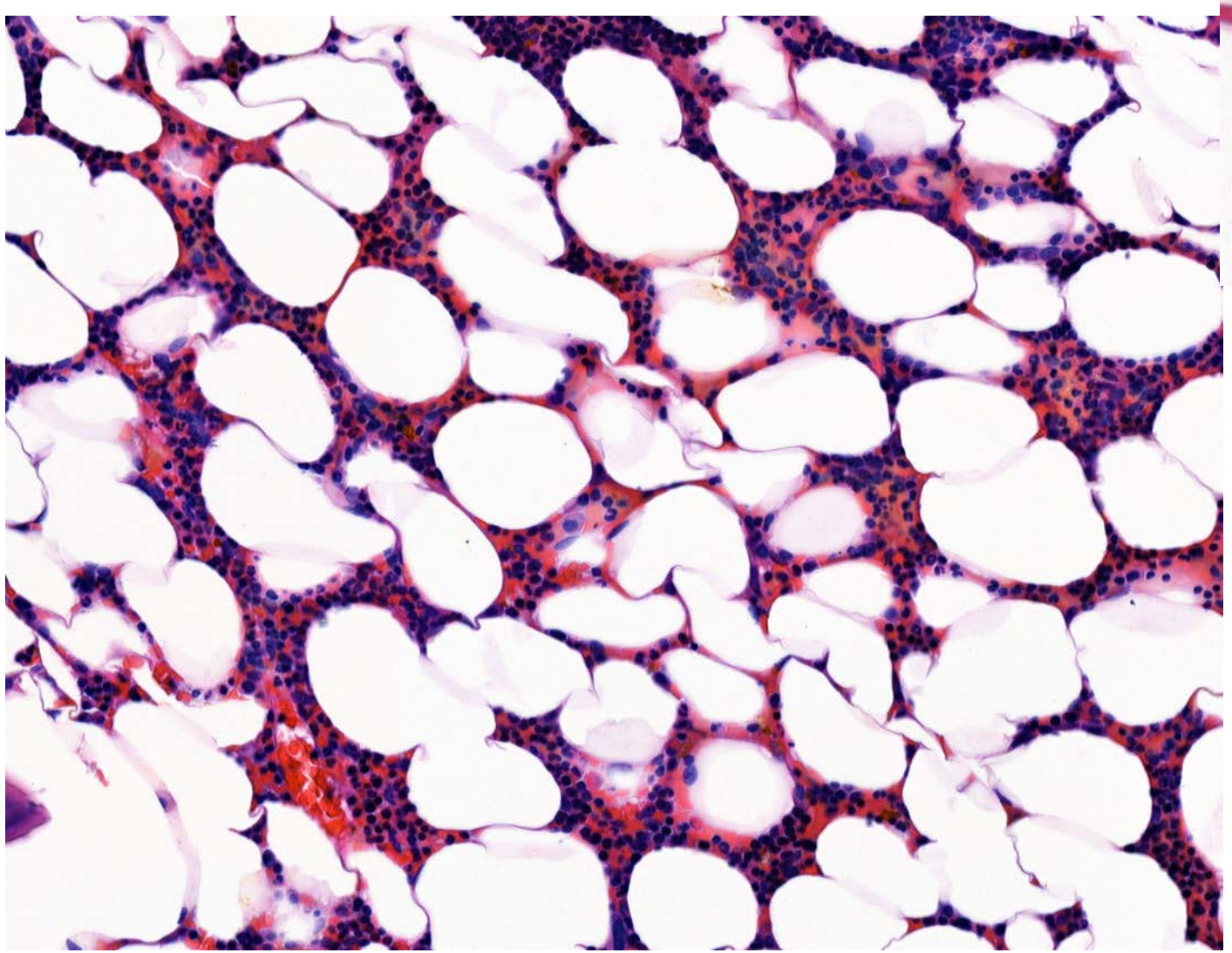
Case 3 : 83y, M

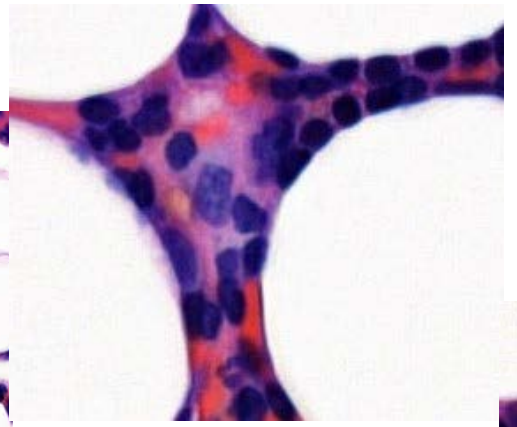


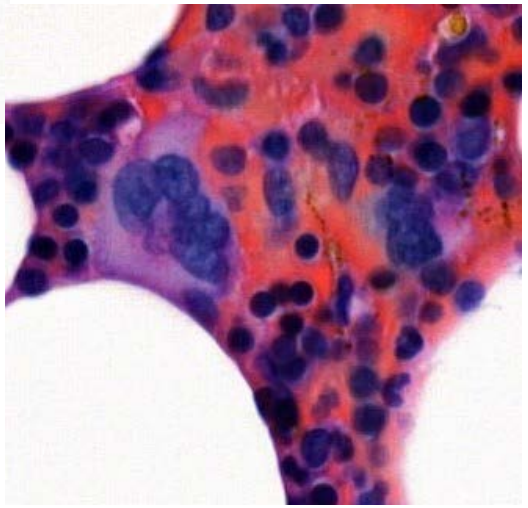
- Bone marrow biopsy

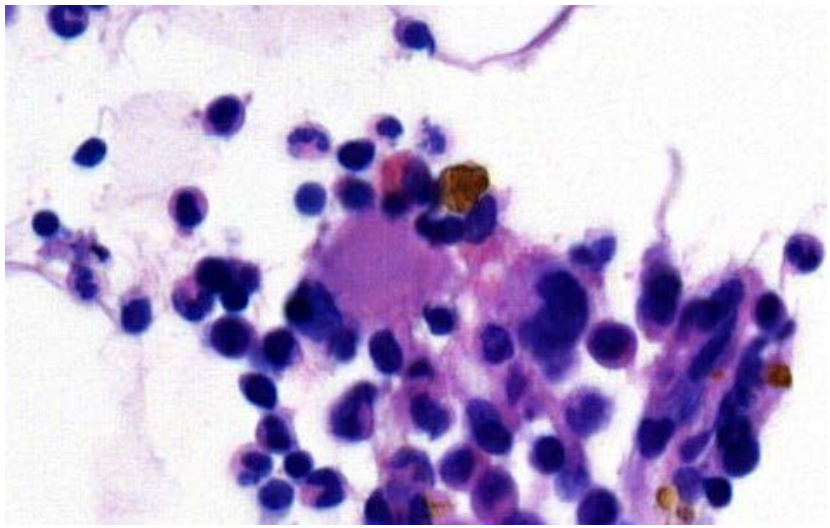
- Anaemia … not associated with metabolic deficiency
- Thrombopaenia gr 0
- MDS ?

Biopsie ost'onidullaire.
Bilan anémie normocytaire non carentielle + thrombopénie gr 0.
SMD?









What’s your diagnosis ?

- Myelodysplatic syndrome
- Lymphoma
- Normal marrow
- Maybe myelodysplastic/ Maybe not myelodysplastic
- Crying in your office (again)
- All of the above

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- Myelodysplatic syndrome
- Lymphoma
- Normal marrow
- **Maybe myelodysplastic/ Maybe not myelodysplastic**
- Crying in your office (again)
- All of the above

Myelodysplastic syndromes

Definition

- Sustained unexplained anemia, neutropenia or thrombocyopenia
  (Hb<10g/dL; Abs. Neutrophil count <1.8 x10^9 or platelets <100 x10^9/L)
- And at least one of the following
  - Dysplastic morphology in erythroid cells, granulocytes or megacaryocytes, affecting at least 10% of the cells of at least one of these lineages
  - Acquired conal MDS-associated cytogenetic abnormality in hematopoïetic cells and absence of de novo AML-defining cytogenetic abnormalities
  - Increased blasts (at least 5% of marrow cells) not attributable to exogenous GF administration or transient marrow recovery

MDS : WHO 2022

Blasts	Cytogenetics	Mutations	
MDS with defining genetic abnormalities			
MDS with low blasts and 5q deletion (MDS-5q)		<5% BM and <2% PB	5q deletion alone, or with 1 other abnormality other than monosomy 7 or 7q deletion
MDS with low blasts and *SF3B1* mutation* (MDS-*SF3B1*)	Absence of 5q deletion, monosomy 7, or complex karyotype	*SF3B1*	
MDS with biallelic *TP53* inactivation (MDS-bi*TP53*)	<20% BM and PB	Usually complex	Two or more *TP53* mutations, or 1 mutation with evidence of *TP53* copy number loss or cnLOH
MDS, morphologically defined			
MDS with low blasts (MDS-LB)		<5% BM and <2% PB	
MDS, hypoplastic† (MDS-h)			
MDS with increased blasts (MDS-IB)			
MDS-IB1	5-9% BM or 2-4% PB		
MDS-IB2	10-19% BM or 5-19% PB or Auer rods		
MDS with fibrosis (MDS-f)	5-19% BM; 2-19% PB		

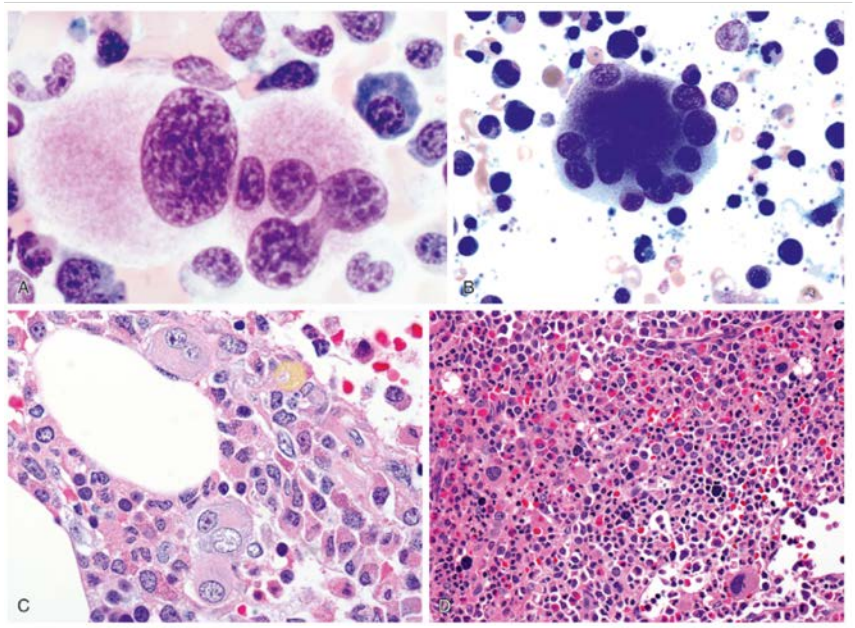
*Detection of ≥15% ring sideroblasts may substitute for *SF3B1* mutation. Acceptable related terminology: MDS with low blasts and ring sideroblasts.
†By definition, ≤25% bone marrow cellularity, age adjusted.
Abbreviations: BM: bone marrow; PB: peripheral blood; cnLOH: copy neutral loss of heterozygosity

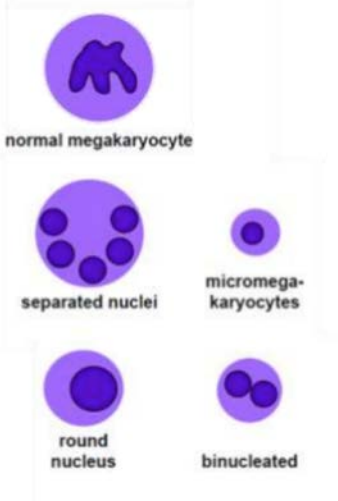
Myelodysplastic syndromes (MDS) and myelodysplastic syndrome/acute myeloid leukemia (MDS/AML)

ICC 2022

	Dysplastic lineages	Cytopenias	Cytoses*	BM and PB Blasts	Cytogenetics†	Mutations
MDS with mutated *SF3B1* (MDS-*SF3B1*)	Typically ≥1‡	≥1	0	<5% BM <2% PB	Any, except isolated del(5q), −7/del(7q), abn3q26.2, or complex	*SF3B1* (≥ 10% VAF), without multi-hit *TP53*, or *RUNX1*
MDS with del(5q) [MDS-del(5q)]	Typically ≥1‡	≥1	Thrombocytosis allowed	<5% BM <2% PB§	del(5q), with up to 1 additional, except −7/del(7q)	Any, except multi-hit *TP53*
MDS, NOS without dysplasia	0	≥1	0	<5% BM <2% PB§	−7/del(7q) or complex	Any, except multi-hit *TP53* or *SF3B1* (≥ 10% VAF)
MDS, NOS with single lineage dysplasia	1	≥1	0	<5% BM <2% PB§	Any, except not meeting criteria for MDS-del(5q)	Any, except multi-hit *TP53;*not meeting criteria for MDS-*SF3B1*
MDS, NOS with multilineage dysplasia	≥2	≥1	0	<5% BM <2% PB§	Any, except not meeting criteria for MDS-del(5q)	Any, except multi-hit *TP53,*; not meeting criteria for MDS-*SF3B1*
MDS with excess blasts (MDS-EB)	Typically ≥1‡	≥1	0	5-9% BM, 2-9% PB§	Any	Any, except multi-hit *TP53*
MDS/AML	Typically ≥1‡	≥1	0	10-19% BM or PB‖	Any, except AML-defining¶	Any, except *NPM1*, bZIP *CEBPA* or *TP53*

Dysplastic Megacaryotes





- Micromegacaryocytes
- Nuclear hypolobulation
- Multinucleation
  - Normal megacaryocytes are uninucleate with lobulated nuclei

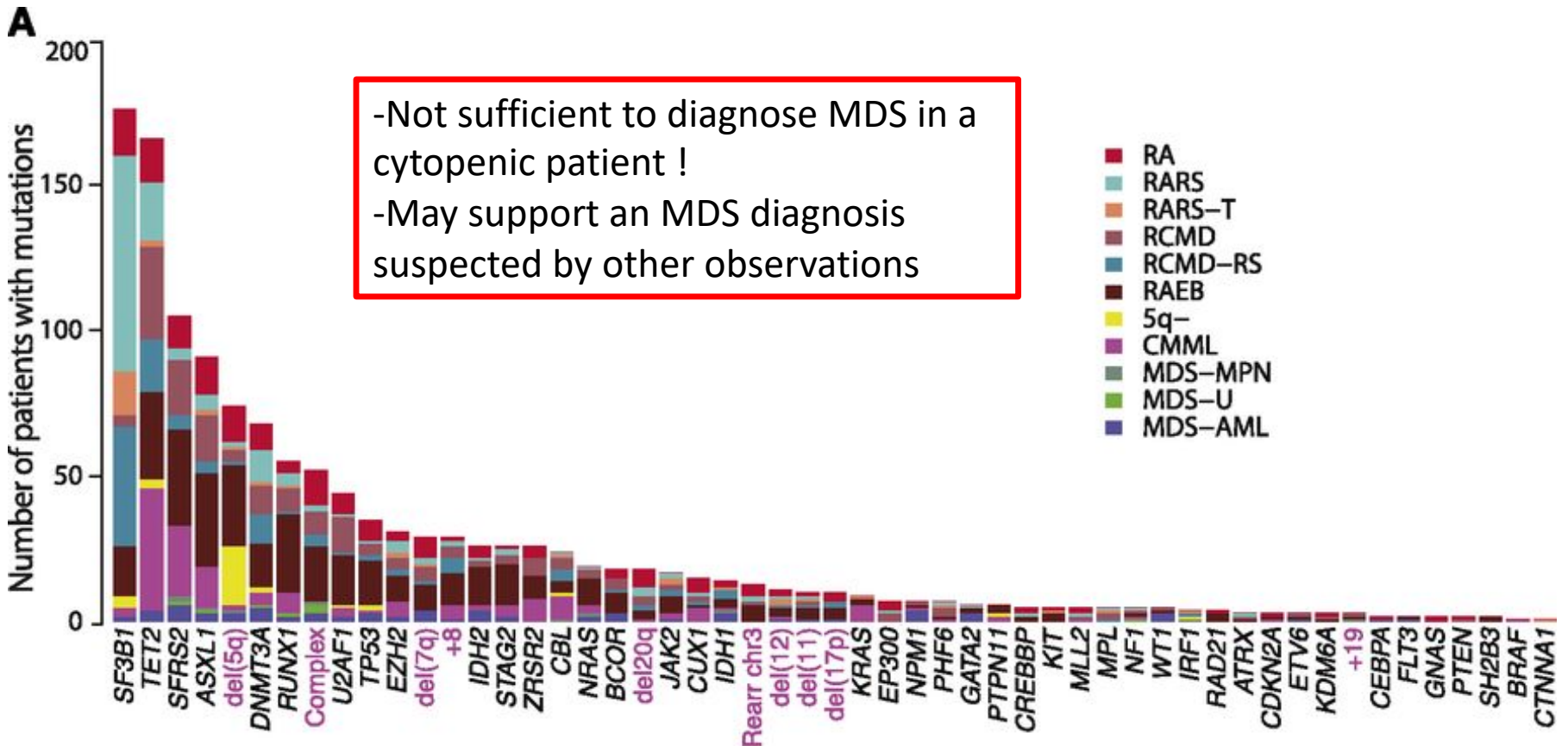
Jaffe, Hematopathology, 2017
Cantù Rajnoldi et al., Ann Hematol, 2005

MDS : Pitfalls
Non-MDS conditions associated with cytopaenia and >10% dysplasia

- Drugs/toxins
  - Recent (<6 m) chemotherapy
  - Heavy alcohol intake
- Metabolic deficiencies: B12, folate, copper
- « Stress erythropoiesis » due to haemoglobinopathy or acquired/congenital haemolytic aenemia
- Infections (HIV, HepC, …)
- Autoimmune diseases
- Concurrent neoplasm
  - Infiltrating marrow (especially MM and HCL)
  - Rare paraneoplastic dysplasia for remote tumour

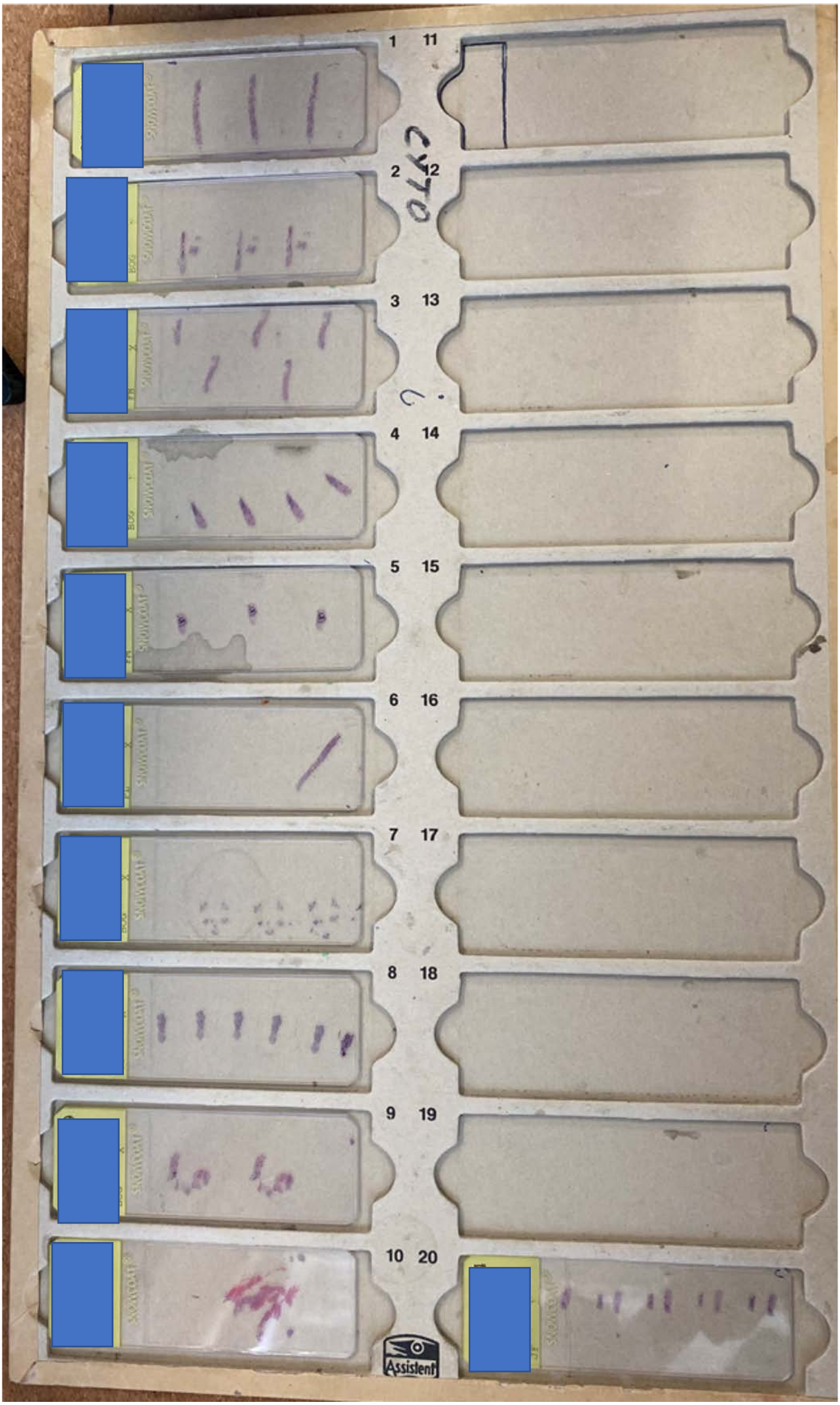
Castello A et al., Haematologica, 1992

MDS :
Recurrent somatic genetic mutations



Genomic architecture of MDS. (A) Frequency of driver mutations identified in the sequencing screen or by cytogenetics in the cohort of 738 patients, broken down by MDS subtype.

Elli Papaemmanuil et al. Blood 2013;122:3616-3627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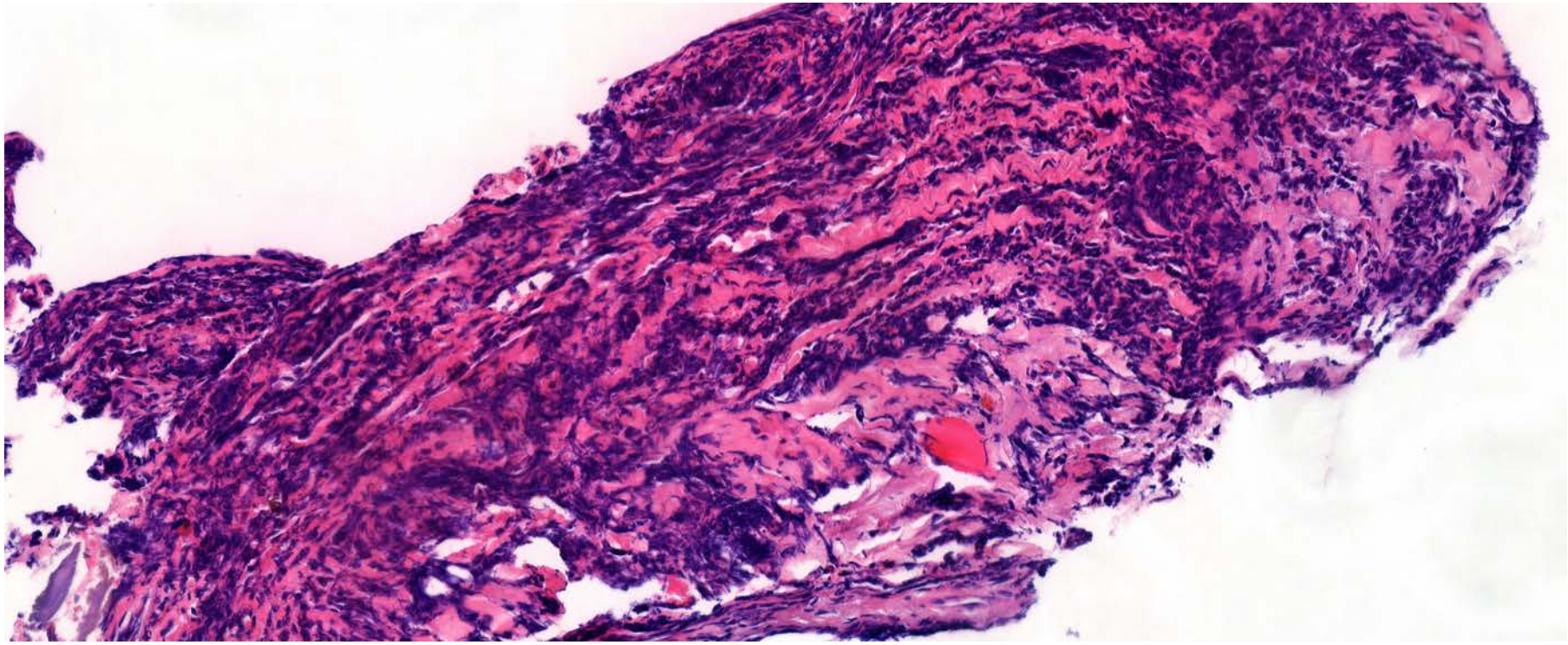
Case 4 : 37 y, M

- Medullary hypermetabolism
- Cervical mass
- Lymphoma ?

Hypermétabolisme médullaire + adénopathies
+ masse cervicale. Lymphome ? MEC.

What’s your diagnosis ?

- Lymphoma
- Myeloma
- Carcinoma
- Melanoma
- All of the above



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- Myeloma
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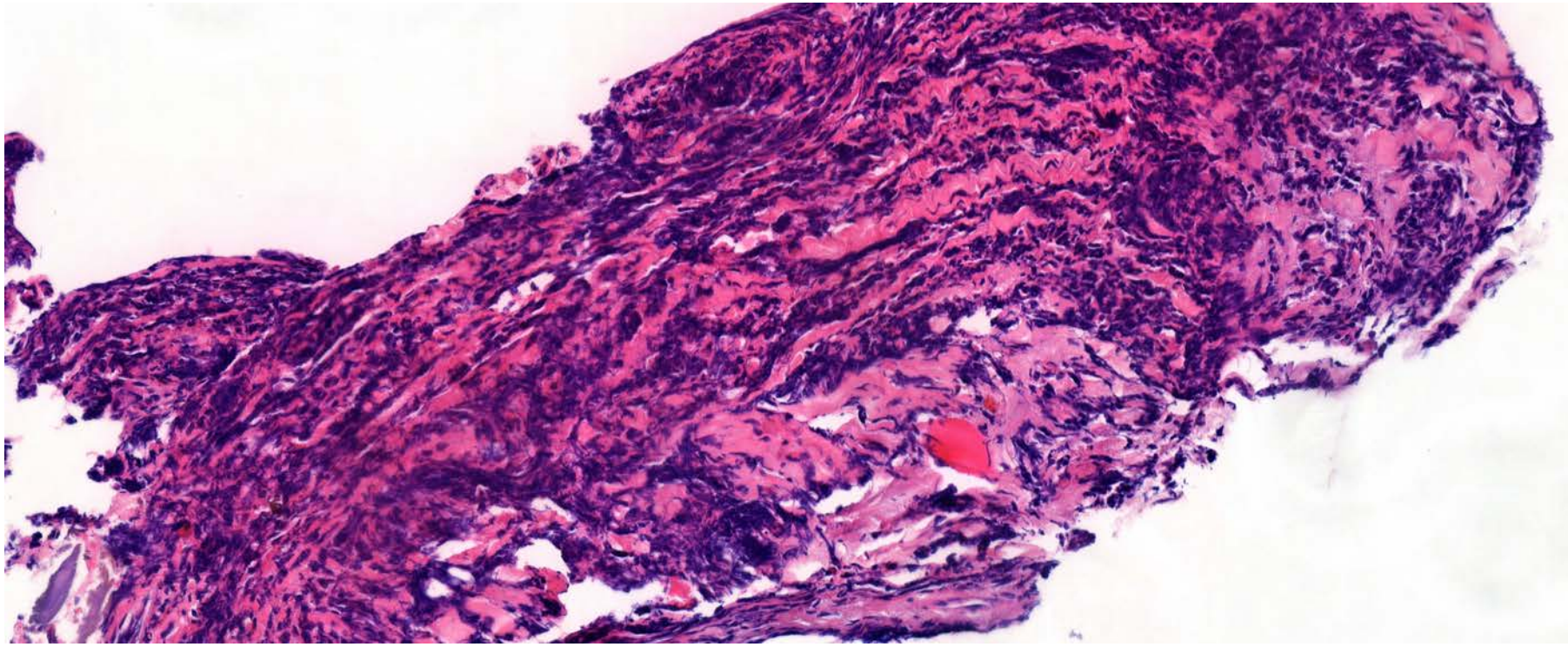
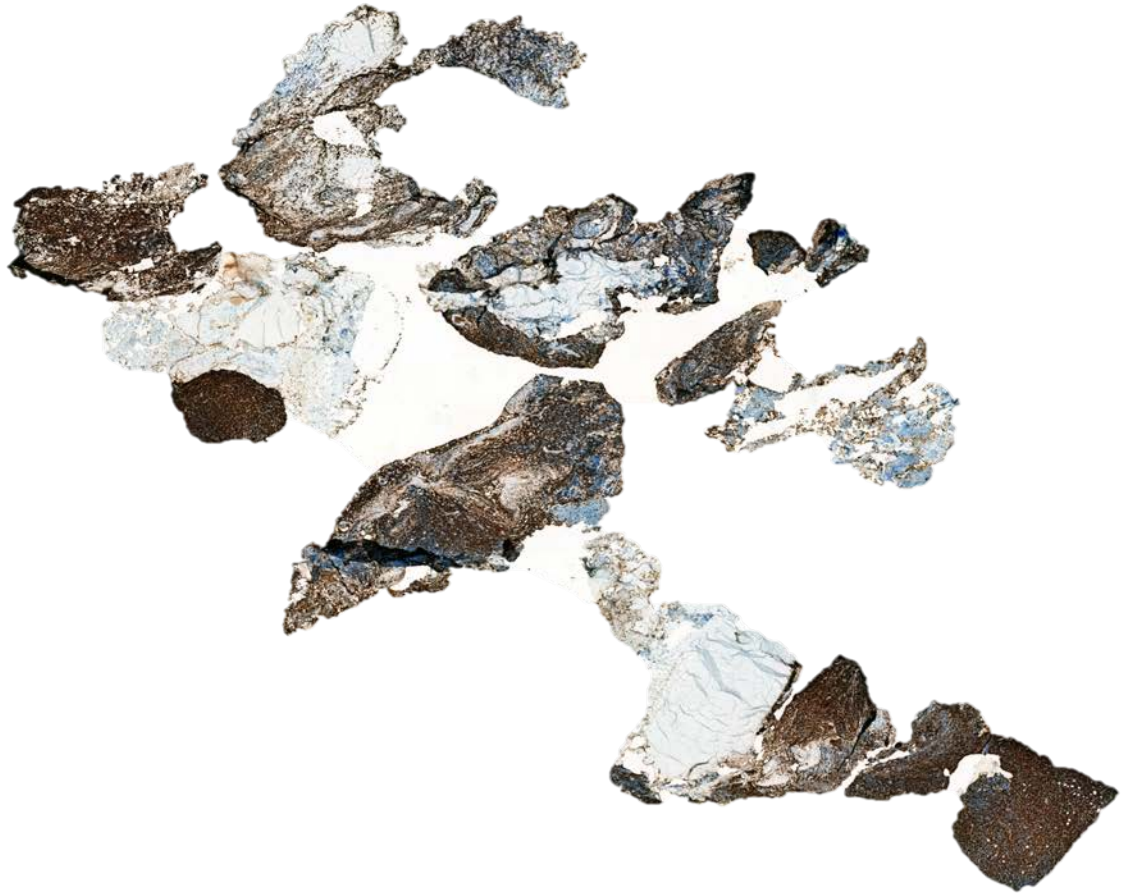
- **All of the above**



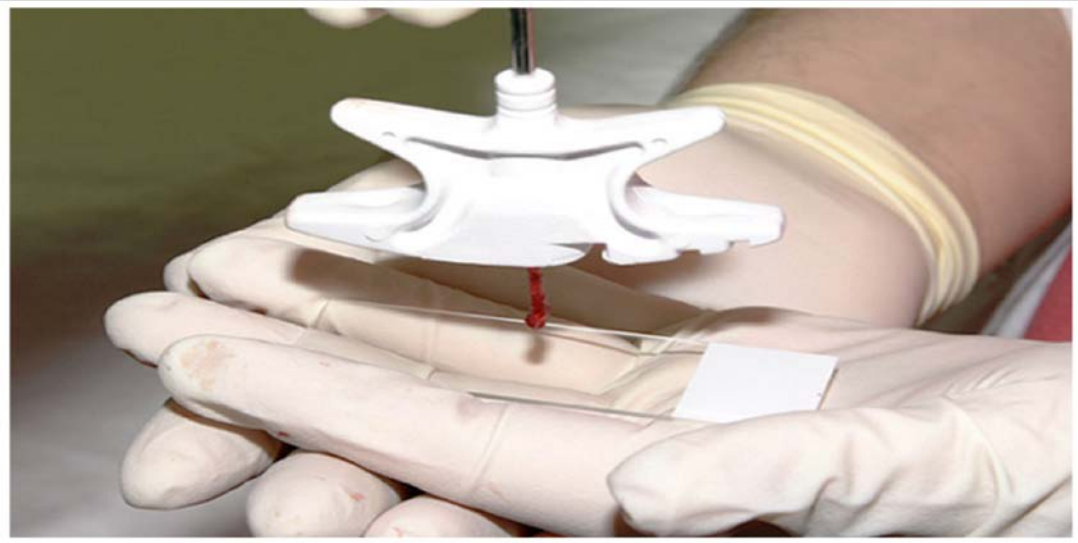
CD34+, CD117+, MPO+, Lyzozyme+

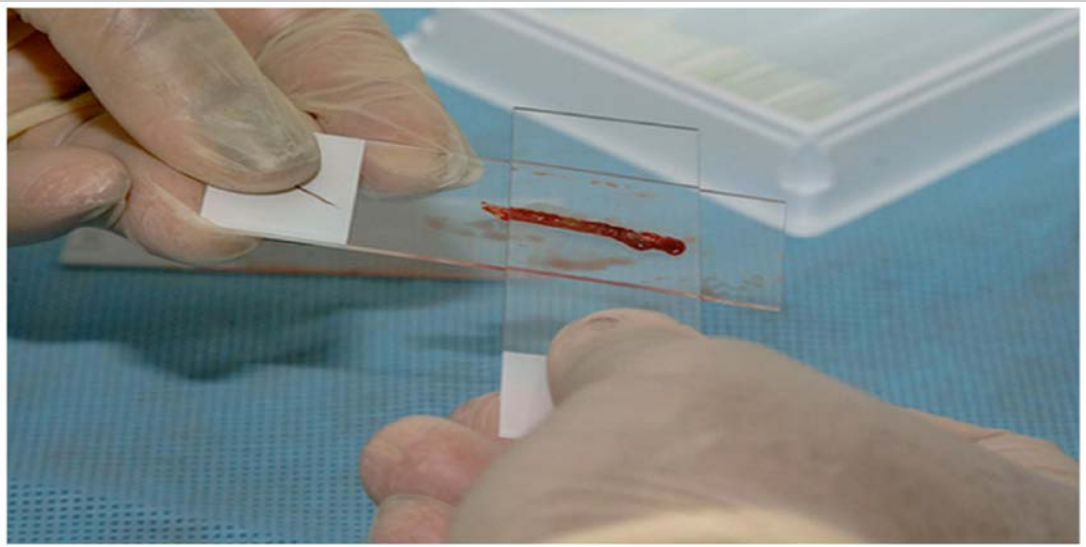
CD34+, CD117+, MPO+, Lyzozyme+



Acute myeloid Leukaemia

DRY TAPE : Touch imprints of the core biopsy

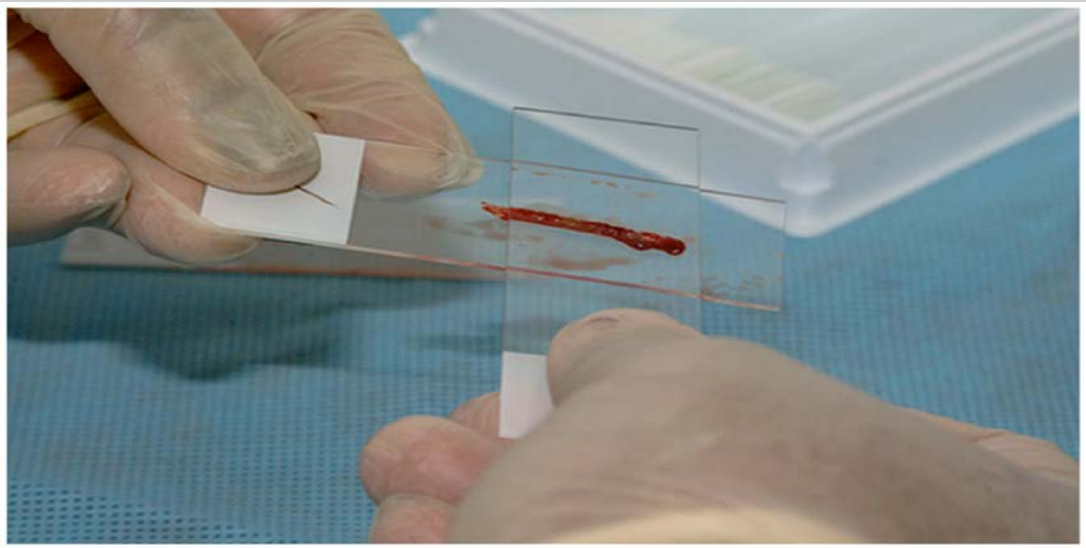




Riley et al., JCLA, 2004

Touch imprints of the core biopsy

Be gentle, be delicate !
Do NOT CRUSH !



Riley et al., JCLA, 2004

Acute Myeloid Leukaemia

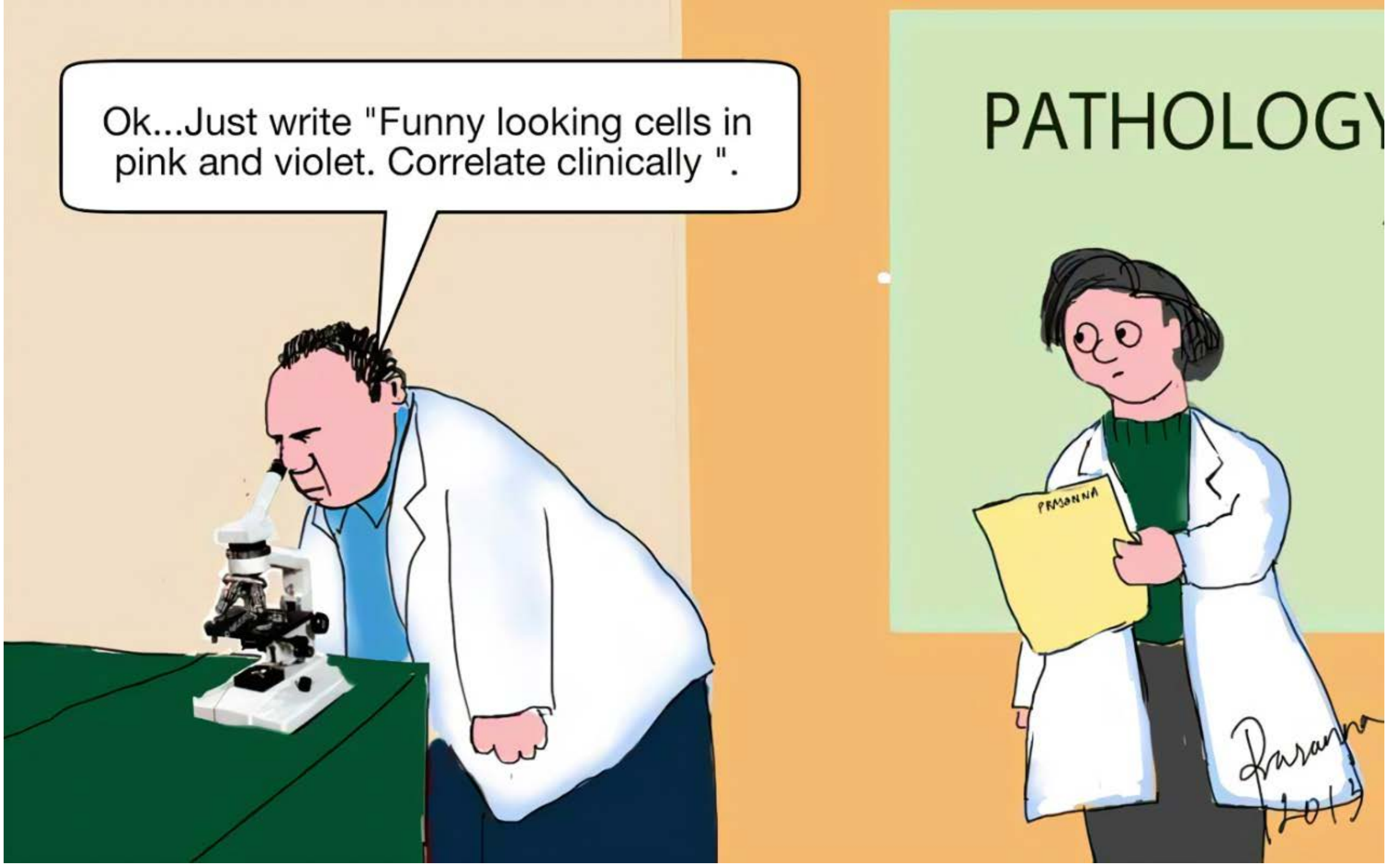
- **Heterogeneous group of diseases**
- Clonal proliferations of immature, non-lymphoid, bone marrow–derived cells
- Most often involve the bone marrow and peripheral blood
- May present in extramedullary tissues
- Aggressive clinical course
- Diagnostic on the basis of a minimum blast cell count in bone marrow (>20% or >10%)
- Several specific AML types are defined without regard to blast cell count
  - Acute myeloid leukemia with t(8;21)(q22;q22.1); RUNX1-RUNX1T1
  - Acute myeloid leukemia with inv(16)(p13.1q22) or t(16;16)(p13.1;q22); CBFB-MYH11
  - Acute promyelocytic leukemia with PML-RARA

Still a role for morphology in the diagnosis of AML?

- **Dry tapes !**
- Blasts (in all cases) should be counted the old fashion way, not based on flow cytometry !
- May allow establishment of a quick diagnosis
  - Especially important in the diagnosis of acute promyelocytic leukemia so therapy can be started
- Exclude relevant differential diagnoses
- Clues to the diagnoses of AML with recurrent genetic anomalies can be obtained by evaluating morphology

CONCLUSION

- Integration of clinical, morphologic, immunophenotypic, genetic, and other biologic features is mandatory to define specific disease entities
- The relative contribution of each feature varies, depending on the case
- Make your cytologists/pathologists/geneticians good ! by providing them relevant clinical informations and optimal samples.


Ok...Just write "Funny looking cells in pink and violet. Correlate clinically ".
PATHOLOGY
PARSONNA

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