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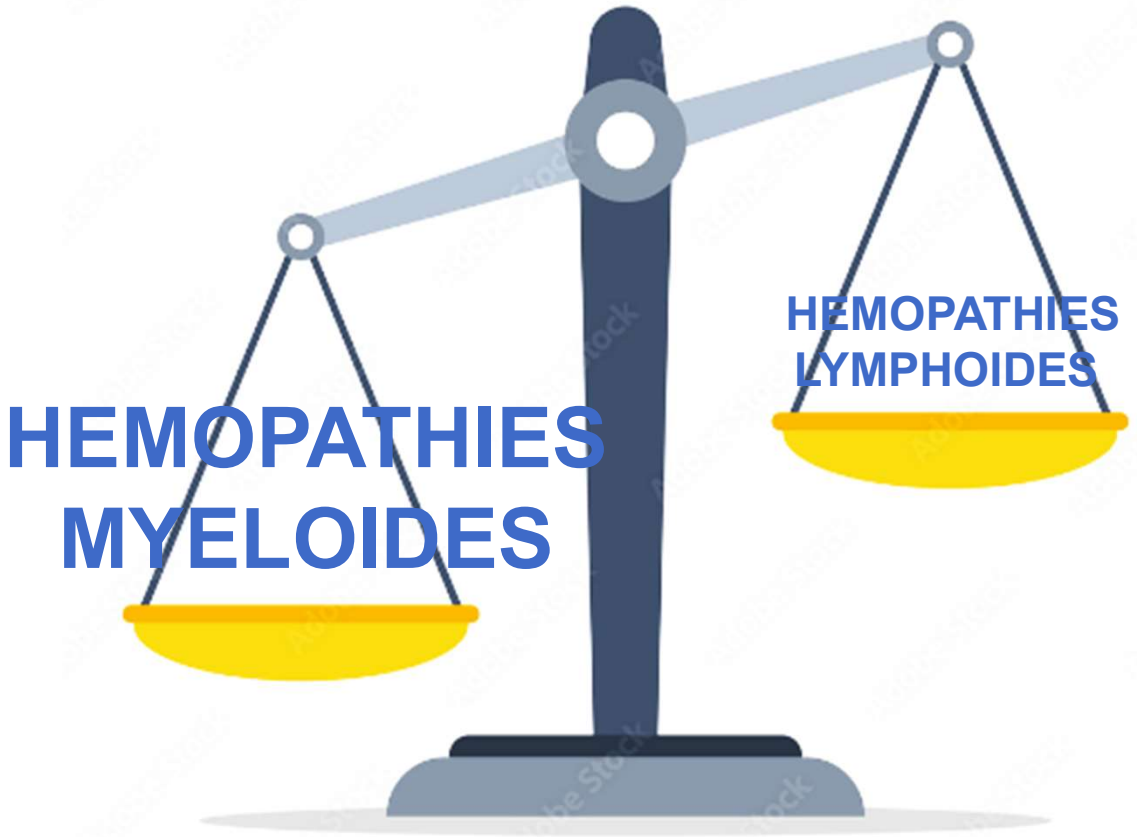
JFBM
6ÈMES JOURNÉES FRANCOPHONES DE BIOLOGIE MÉDICALE

Apport du **NGS** dans les **hémopathies malignes :**

Comment prescrire ?

Apport du NGS et … prescriptions en routine



OMS 2022 – Hémopathies myéloïdes

Leukemia www.nature.com/leu

REVIEW ARTICLE OPEN

Check for updates

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✉]

The upcoming 5th edition of the World Health Organization (WHO) Classification of Haematolymphoid Tumours is part of an effort to hierarchically catalogue human cancers arising in various organ systems within a single relational database. This paper summarizes the new WHO classification scheme for myeloid and histiocytic/dendritic neoplasms and provides an overview of the principles and rationale underpinning changes from the prior edition. The definition and diagnosis of disease types continues to be based on multiple clinicopathologic parameters, but with refinement of diagnostic criteria and emphasis on therapeutically and/or prognostically actionable biomarkers. While a genetic basis for defining diseases is sought where possible, the classification strives to keep practical worldwide applicability in perspective. The result is an enhanced, contemporary, evidence-based classification of myeloid and histiocytic/dendritic neoplasms, rooted in molecular biology and an organizational structure that permits future scalability as new discoveries continue to inexorably inform future editions.

Leukemia (2022) 36:1703–1719; https://doi.org/10.1038/s41375-022-01613-1

NMP

Table 1. Myeloproliferative neoplasms.

Chronic myeloid leukaemia
Polycythaemia vera
Essential thrombocythaemia
Primary myelofibrosis
Chronic neutrophilic leukaemia
Chronic eosinophilic leukaemia
Juvenile myelomonocytic leukaemia
Myeloproliferative neoplasm, not otherwise specified

SMD

Table 3. Classification and defining features of myelodysplastic neoplasms (MDS).

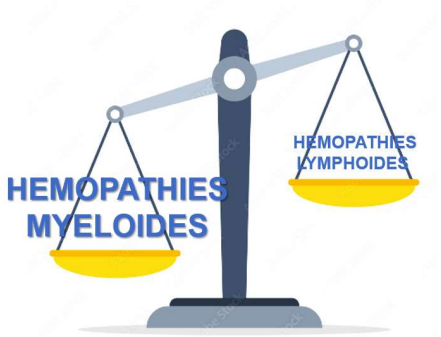
	Blasts	Cytogenetics	Mutations
MDS with defining genetic abnormalities			
MDS with low blasts and isolated 5q deletion (MDS-5q)	<5% BM and <2% PB	5q deletion alone, or with 1 other abnormality other than monosomy 7 or 7q deletion	
MDS with low blasts and *SF3B1* mutation[a] (MDS-*SF3B1*)		Absence of 5q 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[b] (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		

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

Formes frontières

Table 5. Myelodysplastic/myeloproliferative neoplasms.

Chronic myelomonocytic leukaemia
Myelodysplastic/myeloproliferative neoplasm with neutrophilia
Myelodysplastic/myeloproliferative neoplasm with *SF3B1* mutation and thrombocytosis
Myelodysplastic/myeloproliferative neoplasm, not otherwise specified



OMS 2022 – Hémopathies lymphoïdes

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Leukemia

REVIEW ARTICLE OPEN

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LYMPHOMA

The 5th edition of the World Health Organization Classification of Haematolymphoid Tumours: Lymphoid Neoplasms

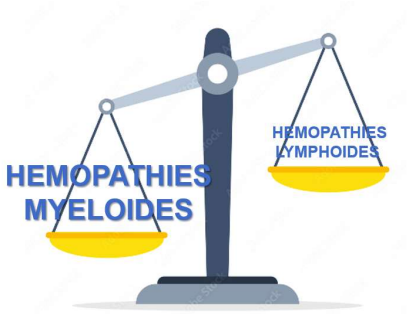
Rita Alaggio[1], Catalina Amador[2], Ioannis Anagnostopoulos[3], Ayoma D. Attygalle[4], Iguaracyra Barreto de Oliveira Araujo[5], Emilio Berti[6], Govind Bhagat[7], Anita Maria Borges[8], Daniel Boyer[9], Mariarita Calaminici[10], Amy Chadburn[11], John K. C. Chan[12], Wah Cheuk[12], Wee-Joo Chng[13], John K. Choi[14], Shih-Sung Chuang[15], Sarah E. Coupland[16], Magdalena Czader[17], Sandeep S. Dave[18], Daphne de Jong[19], Ming-Qing Du[20]✉, Kojo S. Elenitoba-Johnson[21], Judith Ferry[22]✉, Julia Geyer[11], Dita Gratzinger[23], Joan Guitart[24], Sumeet Gujral[25], Marian Harris[26], Christine J. Harrison[27], Sylvia Hartmann[28], Andreas Hochhaus[29], Patty M. Jansen[30], Kennosuke Karube[31], Werner Kempf[32], Joseph Khoury[33], Hiroshi Kimura[34], Wolfram Klapper[35], Alexandra E. Kovach[36], Shaji Kumar[37], Alexander J. Lazar[38], Stefano Lazzi[39], Lorenzo Leoncini[39], Nelson Leung[40], Vasiliki Leventaki[41], Xiao-Qiu Li[42], Megan S. Lim[21], Wei-Ping Liu[43], Abner Louissaint Jr.[22], Andrea Marcogliese[44], L. Jeffrey Medeiros[33], Michael Michal[45], Roberto N. Miranda[33], Christina Mitteldorf[46], Santiago Montes-Moreno[47], William Morice[48], Valentina Nardi[22], Kikkeri N. Naresh[49], Yasodha Natkunam[23], Siok-Bian Ng[50], Ilske Oschlies[35], German Ott[51]✉, Marie Parrens[52], Melissa Pulitzer[53], S. Vincent Rajkumar[54], Andrew C. Rawstron[55], Karen Rech[48], Andreas Rosenwald[3], Jonathan Said[56], Clémentine Sarkozy[57], Shahin Sayed[58], Caner Saygin[59], Anna Schuh[60], William Sewell[61], Reiner Siebert[62]✉, Aliyah R. Sohani[22], Reuben Tooze[63], Alexandra Traverse-Glehen[64], Francisco Vega[33], Beatrice Vergier[65], Ashutosh D. Wechalekar[66], Brent Wood[36], Luc Xerri[67] and Wenbin Xiao[53]

We herein present an overview of the upcoming 5th edition of the World Health Organization Classification of Haematolymphoid Tumours focussing on lymphoid neoplasms. Myeloid and histiocytic neoplasms will be presented in a separate accompanying article. Besides listing the entities of the classification, we highlight and explain changes from the revised 4th edition. These include reorganization of entities by a hierarchical system as is adopted throughout the 5th edition of the WHO classification of tumours of all organ systems, modification of nomenclature for some entities, revision of diagnostic criteria or subtypes, deletion of certain entities, and introduction of new entities, as well as inclusion of tumour-like lesions, mesenchymal lesions specific to lymph node and spleen, and germline predisposition syndromes associated with the lymphoid neoplasms.

Leukemia (2022) 36:1720–1748; https://doi.org/10.1038/s41375-022-01620-2

LLC et autres LMNH-B

Mature B-cell neoplasms	
Pre-neoplastic and neoplastic small lymphocytic proliferations	
Monoclonal B-cell lymphocytosis	(Same)
Chronic lymphocytic leukaemia/small lymphocytic lymphoma	(Same)
(Entity deleted)	B-cell prolymphocytic leukaemia
Splenic B-cell lymphomas and leukaemias	
Hairy cell leukaemia	(Same)
Splenic marginal zone lymphoma	(Same)
Splenic diffuse red pulp small B-cell lymphoma	(Same)
Splenic B-cell lymphoma/leukaemia with prominent nucleoli	*Not previously included* (encompassing hairy cell leukaemia variant and some cases of B-cell prolymphocytic leukaemia)
Lymphoplasmacytic lymphoma	
Lymphoplasmacytic lymphoma	(Same)
Marginal zone lymphoma	
Extranodal marginal zone lymphoma of mucosa-associated lymphoid tissue	(Same)
Primary cutaneous marginal zone lymphoma	*Not previously included* (originally included under "extranodal marginal zone lymphoma of mucosa-associated lymphoid tissue")
Nodal marginal zone lymphoma	(Same)
Paediatric marginal zone lymphoma	(Same)



NGS (Next Generation Sequencing) ou SHD (Séquençage Haut Débit)

NGS (Next Generation Sequencing) ou SHD (Séquençage Haut Débit)

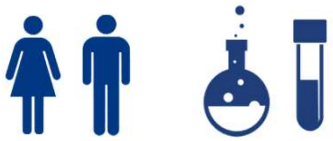
« Wet Lab »

EXTRACTION ADN 1 → PREPARATION LIBRAIRIES 2 → AMPLIFICATION CLONALE 3 → SEQUENCAGE 4

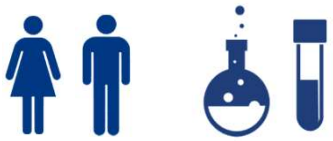
« Dry Lab »

APPEL DES BASES 1 → ALIGNEMENT 2 → DETECTION DES VARIANTS ET ANNOTATION 3 → INTERPRETATION CLINIQUE 4

Process technique NGS « Wet Lab »



Process technique NGS « Wet Lab »



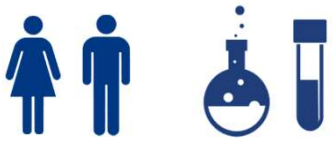
EXTRACTION ADN **1** → PREPARATION LIBRAIRIES **2** → AMPLIFICATION CLONALE **3** → SEQUENCAGE **4**

Sang

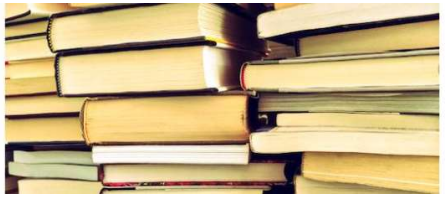
Moelle osseuse

EDTA

Process technique NGS « Wet Lab »



EXTRACTION ADN 1 → PREPARATION LIBRAIRIES 2 → AMPLIFICATION CLONALE 3 → SEQUENCAGE 4



	AVANTAGES	INCONVENIENTS
CAPTURE	Seuil de sensibilité 2% Homogénéité des reads	Qualité ADN
AMPLICON	ADN dégradé	Seuil de sensibilité 5%

Kit Kappa Roche / Kit Qiagen

2019 : Panel de gènes « myéloïdes » – capture – Sophia Genetics

Gene	Transcript	Exon rank
CEBPA	NM_004364	Full coding region
CSF3R	NM_000760	Full coding region
DNMT3A	NM_022552	Full coding region
ETV6	NM_001987	Full coding region
EZH2	NM_001203247	Full coding region
JAK2	NM_004972	Full coding region
RUNX1	NM_001754	Full coding region
TET2	NM_001127208	Full coding region
TP53	LRG_TP53 (LRG-specific mixed numbering)	Full coding region
ZRSR2	NM_005089	Full coding region
ABL1	NM_005157	4, 5, 6, 7, 8, 9
ASLX1	NM_015338	9, 11, 12, 14
BRAF	NM_004333	15
CALR	NM_004343	9
CBL	NM_005188	8, 9
FLT3	NM_004119	13, 14, 15, 20
HRAS	NM_176795	2, 3
IDH1	NM_005896	4
IDH2	NM_002168	4
KIT	NM_000222	2, 8, 9, 10, 11, 13, 17, 18
KRAS	NM_033360	2, 3
MPL	NM_005373	10
NPM1	NM_002520	10, 11
NRAS	NM_002524	2, 3
PTPN11	NM_002834	3, 7, 8, 9, 10, 11, 12, 13
SETBP1	NM_015559	4
SF3B1	NM_012433	10, 11, 12, 13, 14, 15, 16
SRSF2	NM_003016	1
U2AF1	NM_006758	2, 6
WT1	NM_024426	6, 7, 8, 9, 10

30 gènes rapportés dans les hémopathies myéloïdes
(NMP – LMMC – SMD – LAM)

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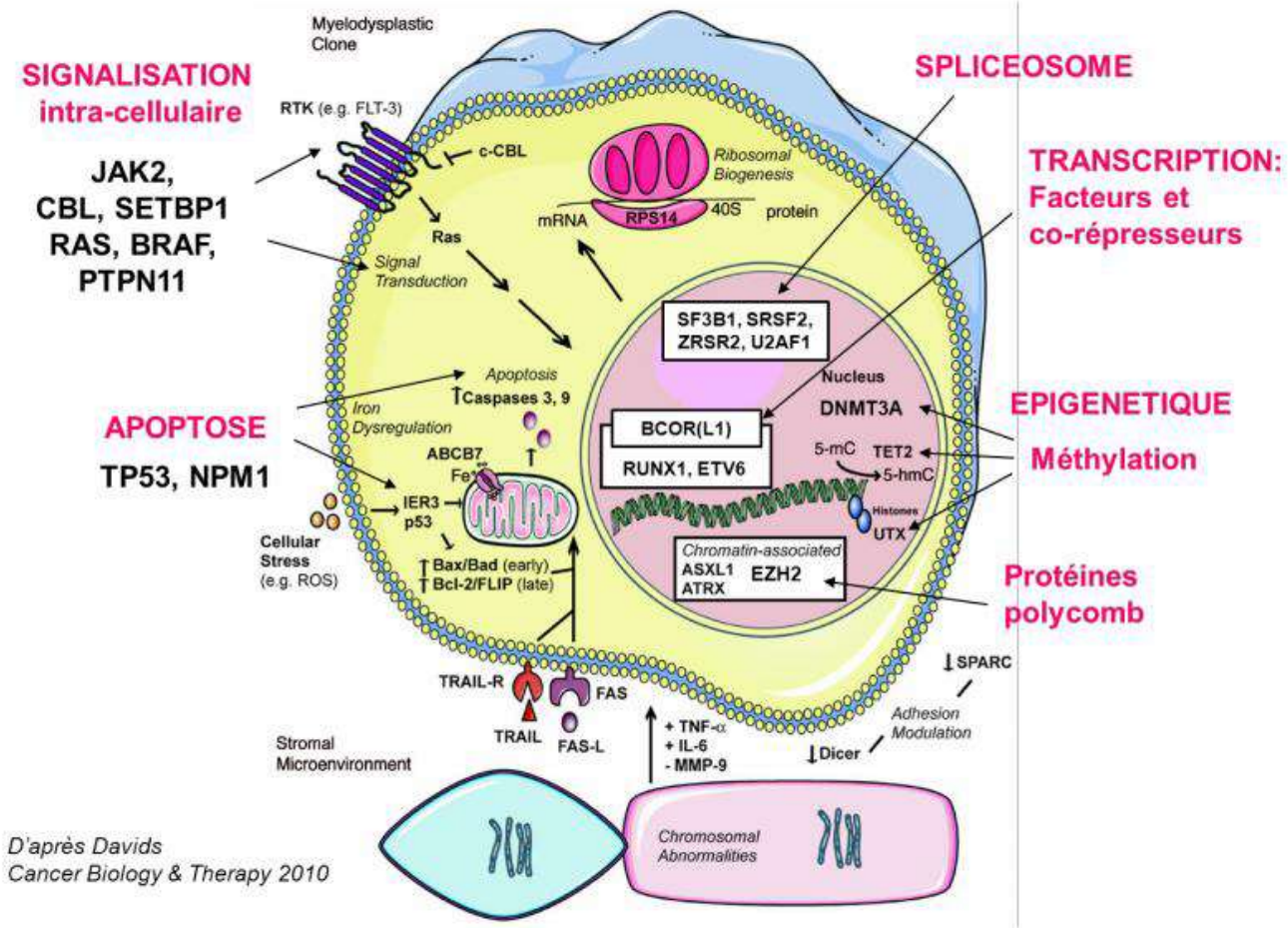

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IDH1	NM_005896	4
IDH2	NM_002168	4
KIT	NM_000222	2, 8, 9, 10, 11, 13, 17, 18
KRAS	NM_033360	2, 3
MPL	NM_005373	10
NPM1	NM_002520	10, 11
NRAS	NM_002524	2, 3
PTPN11	NM_002834	3, 7, 8, 9, 10, 11, 12, 13
SETBP1	NM_015559	4
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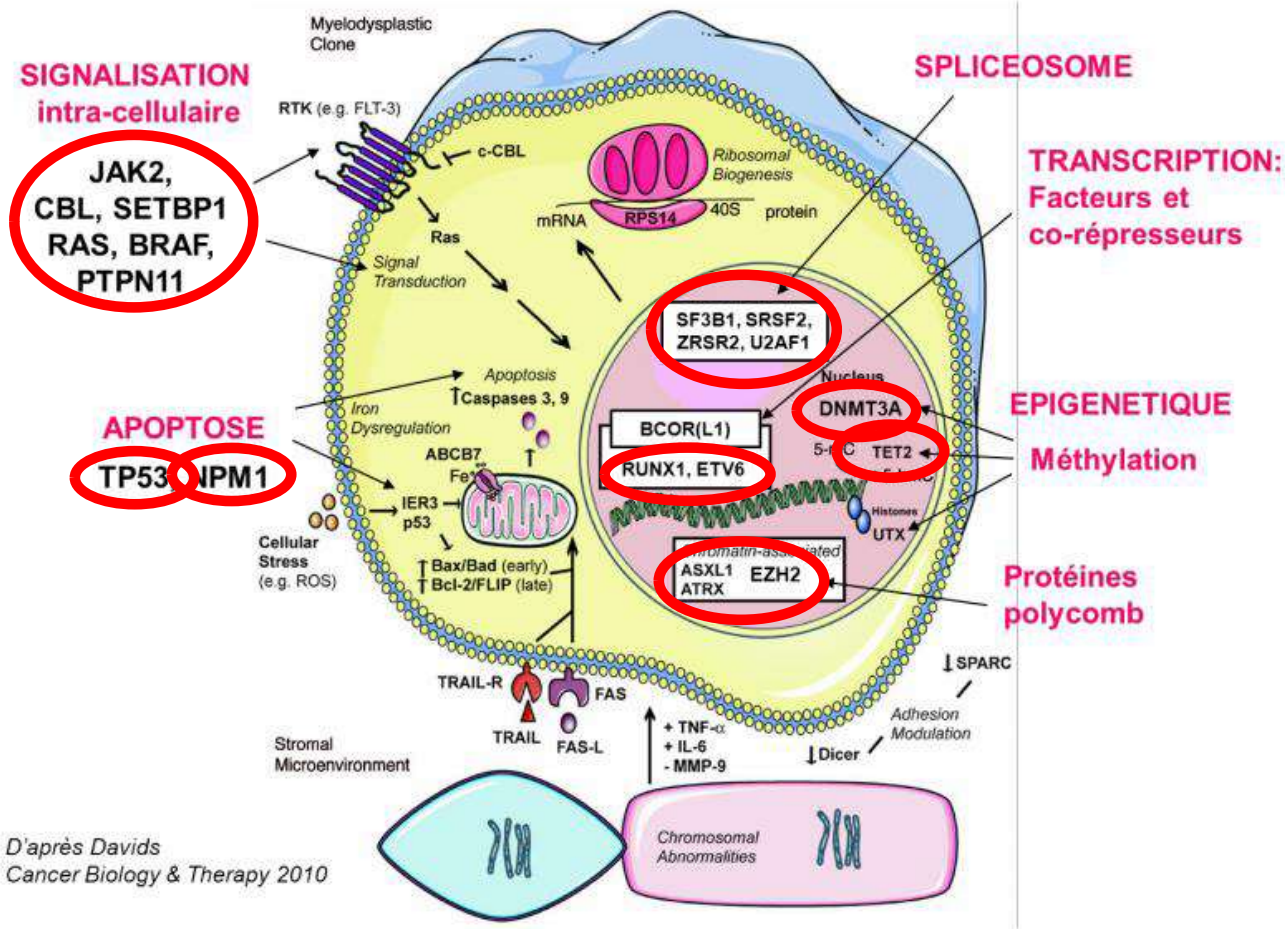

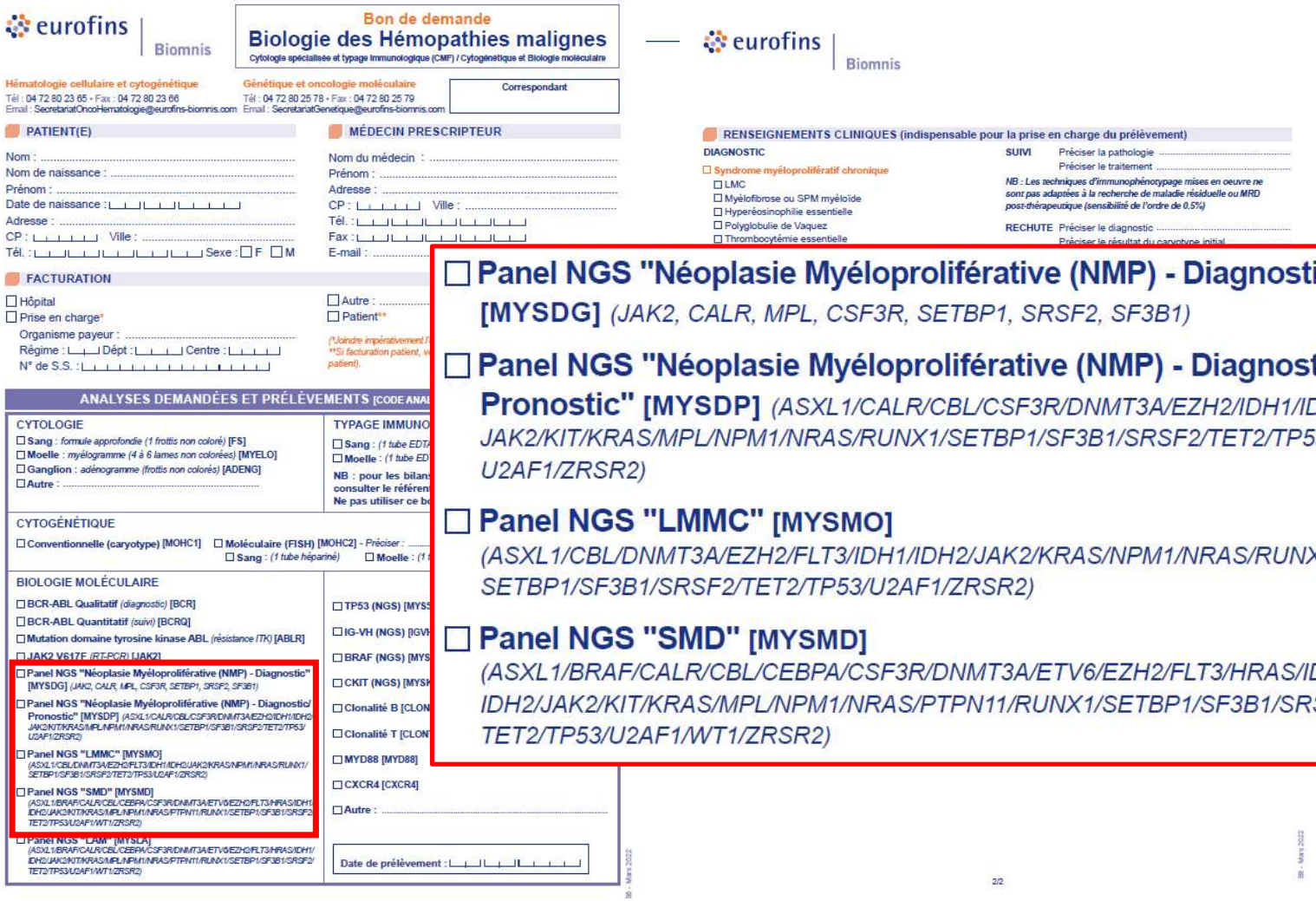

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Bon de demande

Biologie des Hémopathies malignes

Cytologie spécialisée et typage immunologique (CMF) / Cytogénétique et Biologie moléculaire

Hématologie cellulaire et cytogénétique
Tél : 04 72 80 23 65 • Fax : 04 72 80 23 66
Email : SecretariatOncoHematologie@eurofins-biomnis.com

Génétique et oncologie moléculaire
Tél : 04 72 80 25 78 • Fax : 04 72 80 25 79
Email : SecretariatGenetique@eurofins-biomnis.com

Correspondant

PATIENT(E)

Nom :
Nom de naissance :
Prénom :
Date de naissance :
Adresse :
CP : Ville :
Tél. : Sexe : ☐ F ☐ M

MÉDECIN PRESCRIPTEUR

Nom du médecin :
Prénom :
Adresse :
CP : Ville :
Tél. :
Fax :
E-mail :

FACTURATION

☐ Hôpital
☐ Prise en charge*
Organisme payeur :
Régime : Dépt : Centre :
N° de S.S. :
☐ Autre :
☐ Patient**

ANALYSES DEMANDÉES ET PRÉLÈVEMENTS [CODE ANAL

CYTOLOGIE

☐ Sang : *formule approfondie (1 frottis non coloré)* [FS]
☐ Moelle : *myélogramme (4 à 6 lames non colorées)* [MYELO]
☐ Ganglion : *adénogramme (frottis non colorés)* [ADENG]
☐ Autre :

TYPAGE IMMUNO

☐ Sang : *(1 tube EDT*
☐ Moelle : *(1 tube ED*

CYTOGÉNÉTIQUE

☐ Conventionnelle (caryotype) [MOHC1] ☐ Moléculaire (FISH) [MOHC2] - *Préciser :*
☐ Sang : *(1 tube hépariné)* ☐ Moelle : *(1*

BIOLOGIE MOLÉCULAIRE

☐ BCR-ABL Qualitatif *(diagnostic)* [BCR]
☐ BCR-ABL Quantitatif *(suivi)* [BCRQ]
☐ Mutation domaine tyrosine kinase ABL *(résistance ITK)* [ABLR]
☐ JAK2 V617F *(RT-PCR)* [JAK2]
☐ Panel NGS "Néoplasie Myéloprolifération (NMP) - Diagnostic" [MYSDG] *(JAK2, CALR, MPL, CSF3R, SETBP1, SRSF2, SF3B1)*
☐ Panel NGS "Néoplasie Myéloprolifération (NMP) - Diagnostic/ Pronostic" [MYSDP] *(ASXL1/CALR/CBL/CSF3R/DNMT3A/EZH2/IDH1/IDH2/ JAK2/KIT/KRAS/MPL/NPM1/NRAS/RUNX1/SETBP1/SF3B1/SRSF2/TET2/TP53/ U2AF1/ZRSR2)*
☐ Panel NGS "LMMC" [MYSMO] *(ASXL1/CBL/DNMT3A/EZH2/FLT3/IDH1/IDH2/JAK2/KRAS/NPM1/NRAS/RUNX1/ SETBP1/SF3B1/SRSF2/TET2/TP53/U2AF1/ZRSR2)*
☐ Panel NGS "SMD" [MYSMD] *(ASXL1/BRAF/CALR/CBL/CEBPA/CSF3R/DNMT3A/ETV6/EZH2/FLT3/HRAS/IDH1/ IDH2/JAK2/KIT/KRAS/MPL/NPM1/NRAS/PTPN11/RUNX1/SETBP1/SF3B1/SRSF2/ TET2/TP53/U2AF1/WT1/ZRSR2)*
☐ Panel NGS "LAM" [MYSLA] *(ASXL1/BRAF/CALR/CBL/CEBPA/CSF3R/DNMT3A/ETV6/EZH2/FLT3/HRAS/IDH1/ IDH2/JAK2/KIT/KRAS/MPL/NPM1/NRAS/PTPN11/RUNX1/SETBP1/SF3B1/SRSF2/ TET2/TP53/U2AF1/WT1/ZRSR2)*

☐ TP53 (NGS) [MYS
☐ IG-VH (NGS) [IGV
☐ BRAF (NGS) [MYS
☐ CKIT (NGS) [MYS
☐ Clonalité B [CLON
☐ Clonalité T [CLON
☐ MYD88 [MYD88]
☐ CXCR4 [CXCR4]
☐ Autre :

Date de prélèvement :

99 - Mars 2022

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RENSEIGNEMENTS CLINIQUES (indispensable pour la prise en charge du prélèvement)

DIAGNOSTIC

☐ Syndrome myéloprolifératif chronique
☐ LMC
☐ Myélofibrose ou SPM myéloïde
☐ Hyperéosinophilie essentielle
☐ Polyglobulie de Vaquez
☐ Thrombocytémie essentielle

SUIVI Préciser la pathologie
Préciser le traitement

NB : Les techniques d'immunophénotypage mises en oeuvre ne sont pas adaptées à la recherche de maladie résiduelle ou MRD post-thérapeutique (sensibilité de l'ordre de 0.5%)

RECHUTE Préciser le diagnostic
Préciser le résultat du caryotype initial

2/2

99 - Mars 2022

☐ **Panel NGS "Néoplasie Myéloprolifération (NMP) - Diagnostic" [MYSDG]** *(JAK2, CALR, MPL, CSF3R, SETBP1, SRSF2, SF3B1)*

☐ **Panel NGS "Néoplasie Myéloprolifération (NMP) - Diagnostic/ Pronostic" [MYSDP]** *(ASXL1/CALR/CBL/CSF3R/DNMT3A/EZH2/IDH1/IDH2/ JAK2/KIT/KRAS/MPL/NPM1/NRAS/RUNX1/SETBP1/SF3B1/SRSF2/TET2/TP53/ U2AF1/ZRSR2)*

☐ **Panel NGS "LMMC" [MYSMO]** *(ASXL1/CBL/DNMT3A/EZH2/FLT3/IDH1/IDH2/JAK2/KRAS/NPM1/NRAS/RUNX1/ SETBP1/SF3B1/SRSF2/TET2/TP53/U2AF1/ZRSR2)*

☐ **Panel NGS "SMD" [MYSMD]** *(ASXL1/BRAF/CALR/CBL/CEBPA/CSF3R/DNMT3A/ETV6/EZH2/FLT3/HRAS/IDH1/ IDH2/JAK2/KIT/KRAS/MPL/NPM1/NRAS/PTPN11/RUNX1/SETBP1/SF3B1/SRSF2/ TET2/TP53/U2AF1/WT1/ZRSR2)*

Fin 2023

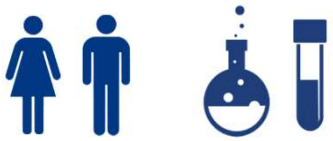
Nouveaux gènes dans les hémopathies myéloïdes

+

NGS « Lymphoïde »

85 gènes

Process technique NGS « Wet Lab »

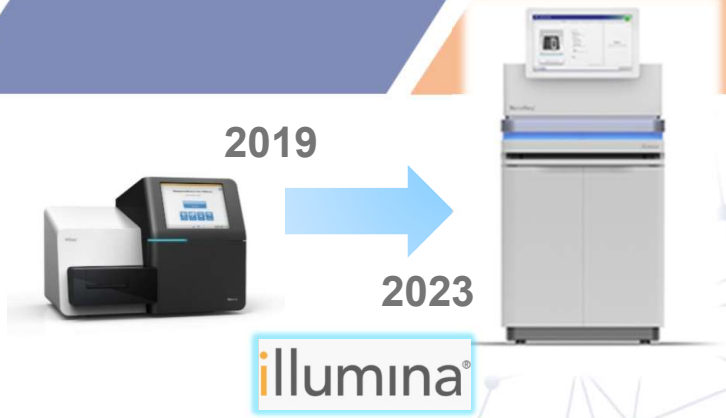


EXTRACTION ADN 1

PREPARATION LIBRAIRIES 2

AMPLIFICATION CLONALE 3

SEQUENCAGE 4



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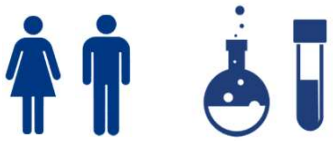


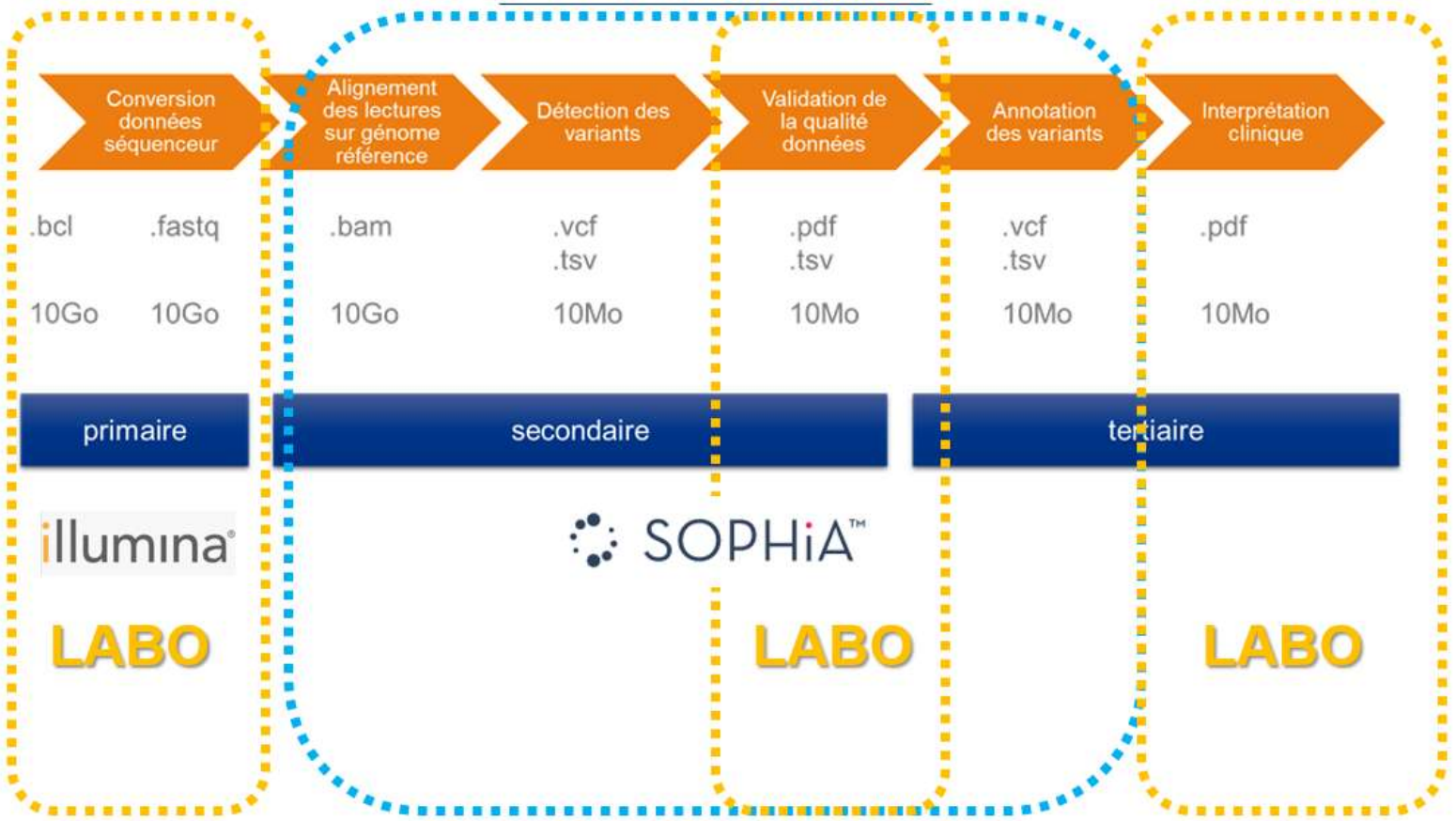
Process technique NGS « Dry Lab »

Process technique NGS « Dry Lab »

SOPHiA™

Outil bioinformatique *externalisé*

Process technique NGS « Dry Lab »



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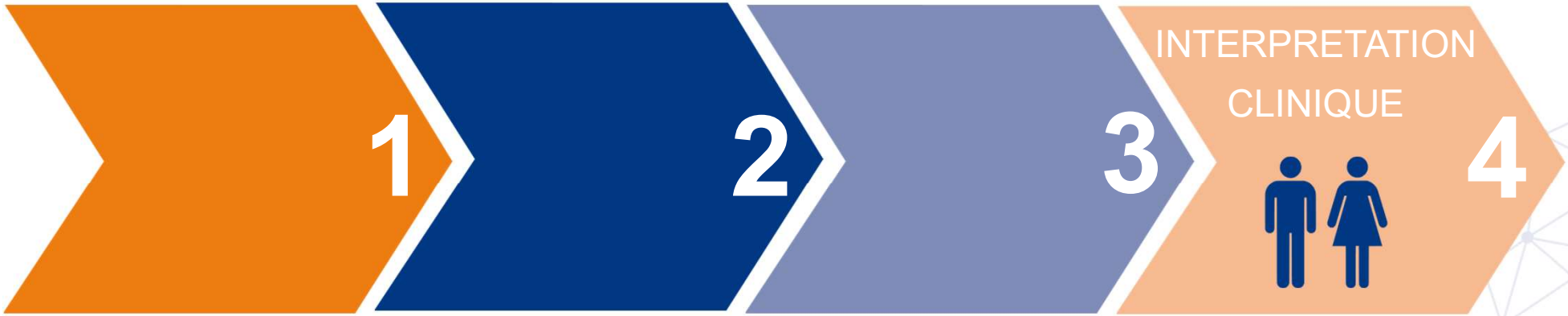

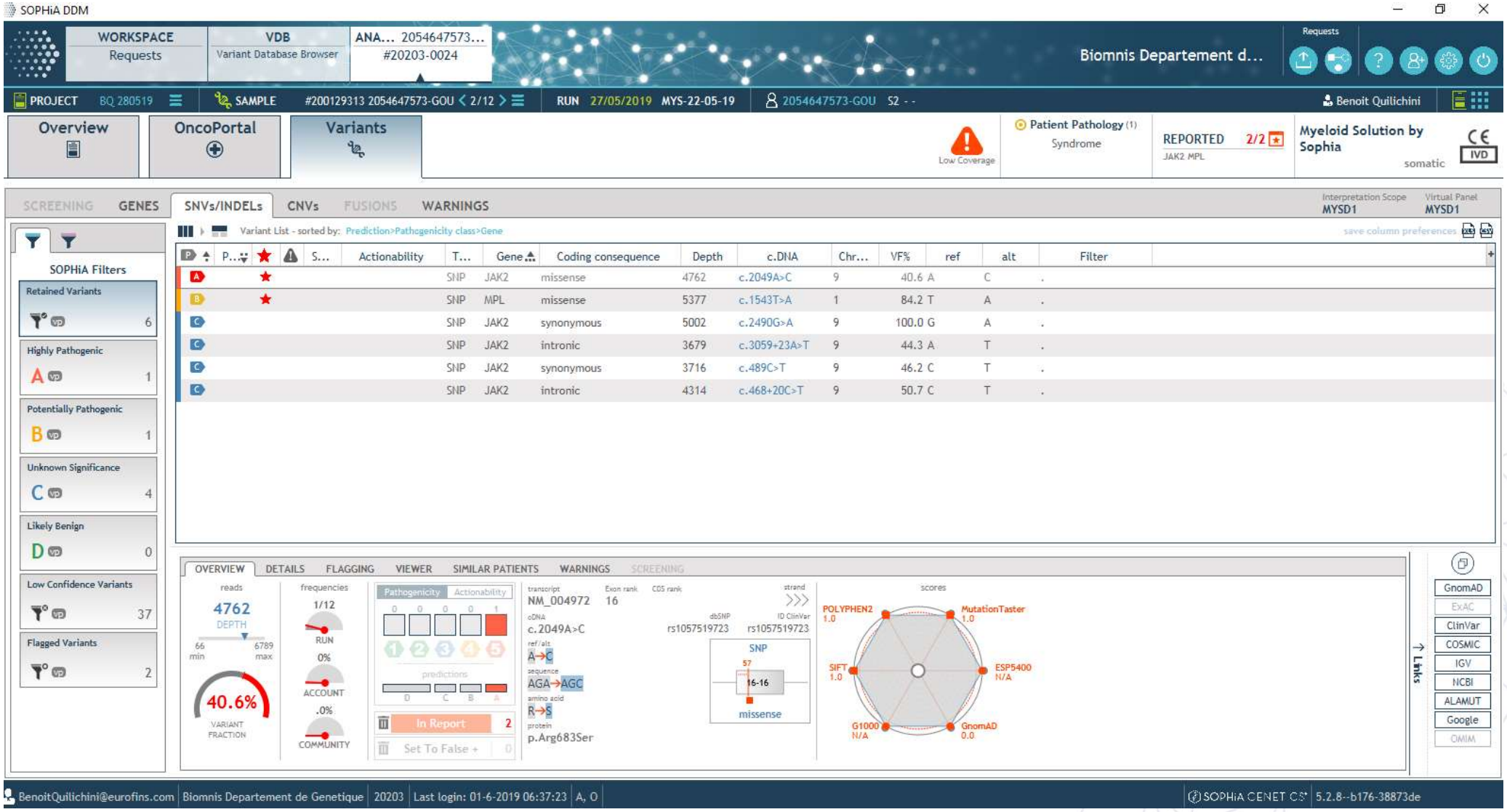
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RUN 27/05/2019 MYS-22-05-19
2054647573-GOU S2 - -
Benoit Quilichini
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Unknown Significance C 4
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Flagged Variants 2
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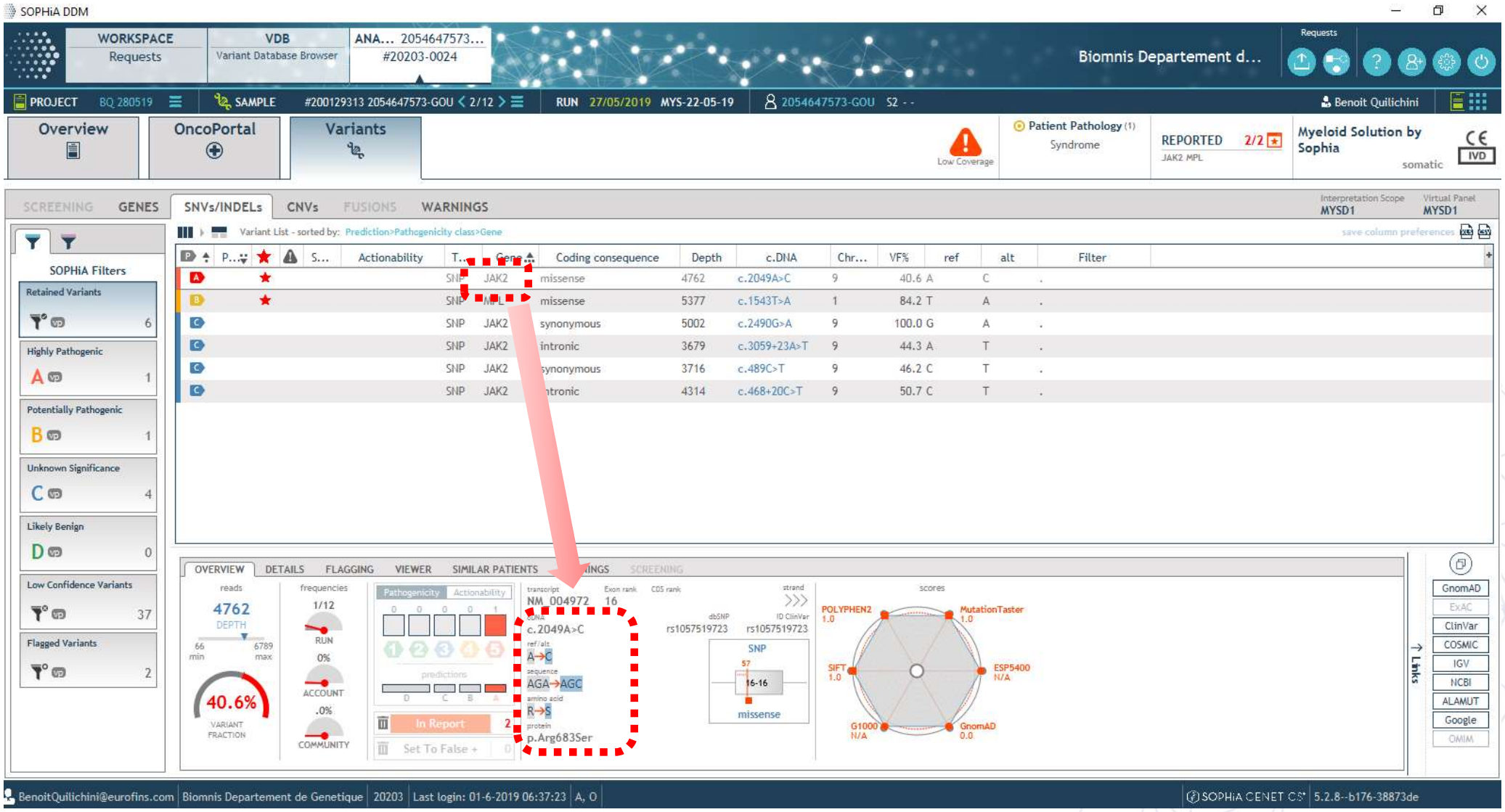
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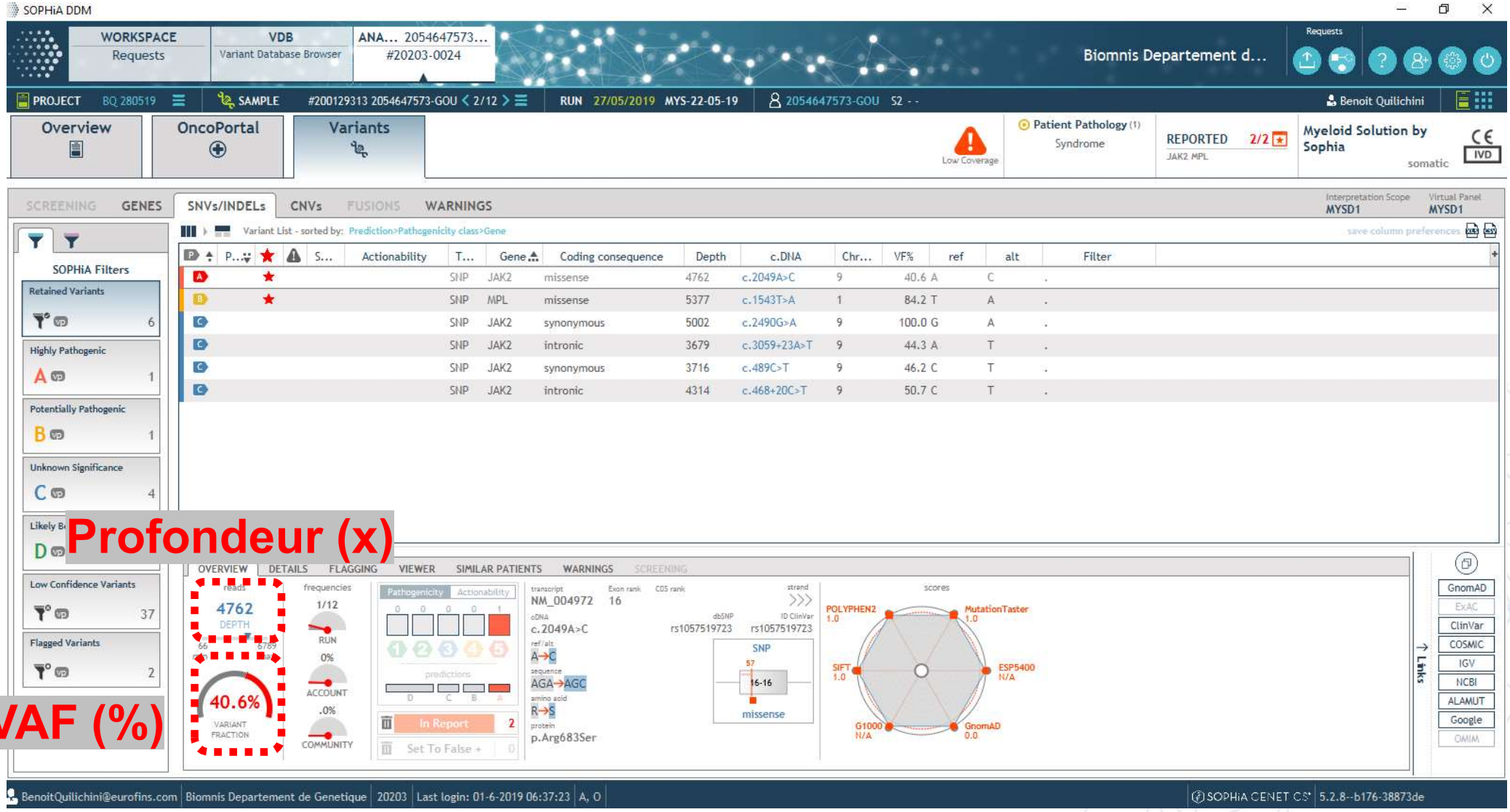
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Likely B
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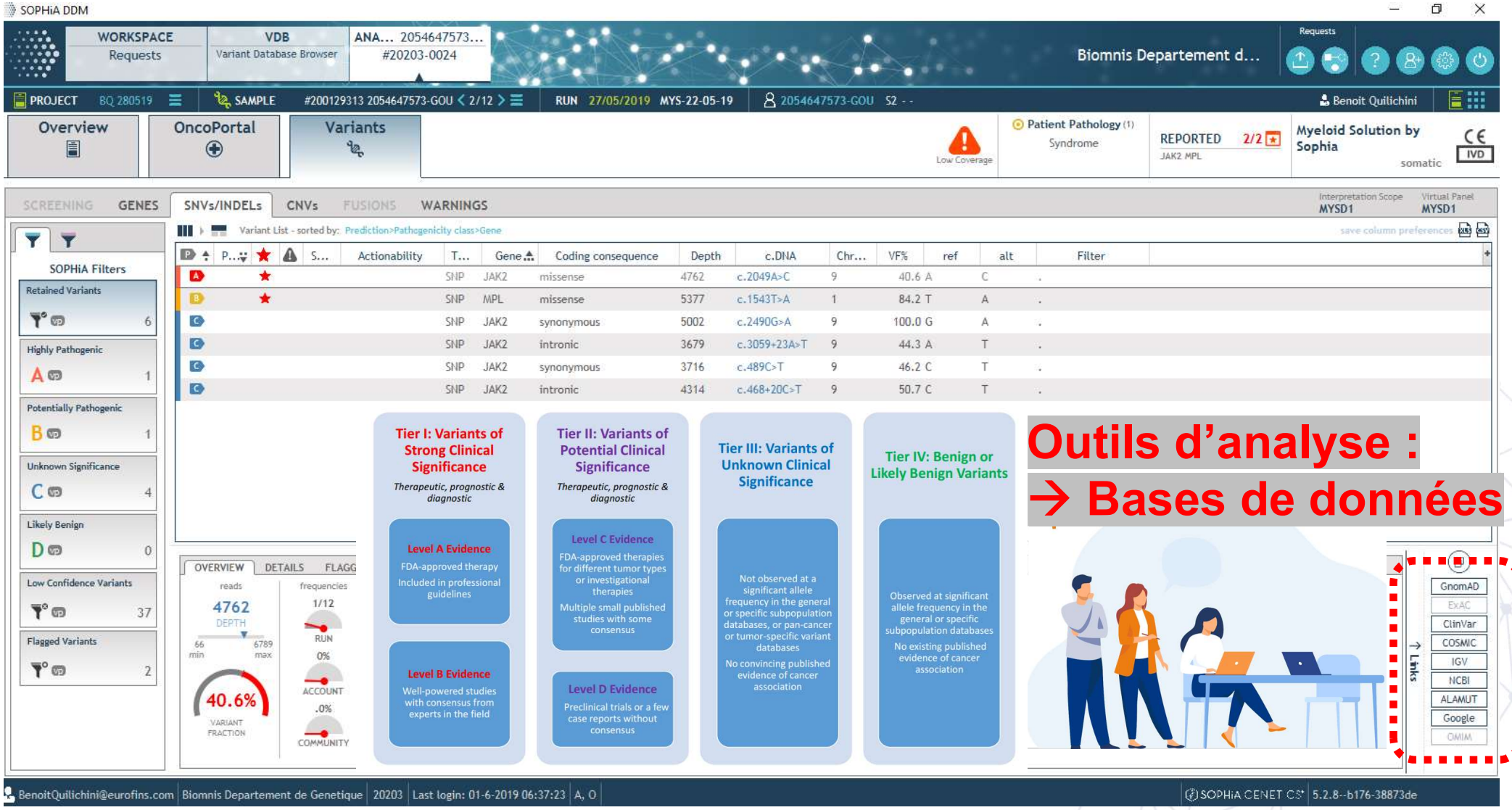
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DEPTH
66 min
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frequencies
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RUN
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COMMUNITY
Tier I: Variants of Strong Clinical Significance
Therapeutic, prognostic & diagnostic
Level A Evidence
FDA-approved therapy
Included in professional guidelines
Level B Evidence
Well-powered studies with consensus from experts in the field
Tier II: Variants of Potential Clinical Significance
Therapeutic, prognostic & diagnostic
Level C Evidence
FDA-approved therapies for different tumor types or investigational therapies
Multiple small published studies with some consensus
Level D Evidence
Preclinical trials or a few case reports without consensus
Tier III: Variants of Unknown Clinical Significance
Not observed at a significant allele frequency in the general or specific subpopulation databases, or pan-cancer or tumor-specific variant databases
No convincing published evidence of cancer association
Tier IV: Benign or Likely Benign Variants
Observed at significant allele frequency in the general or specific subpopulation databases
No existing published evidence of cancer association
Outils d'analyse :
→ Bases de données
Links
GnomAD
ExAC
ClinVar
COSMIC
IGV
NCBI
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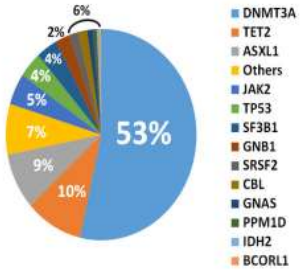
Validation d'un résultat

1. **Hot spot mutationnel dans les hémopathies malignes**
   - **Valeur diagnostique ?**
   - **Valeur pronostique ?**
   - Valeur théranostique
2. Variant rapporté dans les hémopathies mais pas un hot spot
   - Bases de données ?
   - Bibliographie ?
3. Variant non rapporté dans les hémopathies
   - Doit-on le rendre au clinicien ?
4. **CHIP** ? Variant issu d'une hématopoïèse clonale de signification indéterminée OMS 2022
   - Liée à l'âge
   - Etat « pré-leucémique » ?
5. Variant d'origine **constitutionnelle** ?
   - Valeur de VAF 50% / 100%
   - Vérification sur un autre tissu non hématopoïétique



Avantages du NGS

1. Exhaustivité d'analyse des gènes impliqués
   - Panel de gènes
     - Panel MYS = 30 gènes en 2019 → Panel 85 gènes en 2023
     - Nombre de gènes selon les recommandations en progression croissante +++

2. Sensibilité de détection
   - Capture : sensibilité 2% (Sanger 10 à 15%)

3. Rapidité d'analyse d'un grand nombre d'échantillons
   - Fonction de la flow cell utilisée
     - MiSeq : V2 ou V3 : 12 à 24 échantillons → subsampling à 36 échantillons
     - NovaSeq : 48 échantillons

NMP

Table 1. Myeloproliferative neoplasms.

Chronic myeloid leukaemia
Polycythaemia vera
Essential thrombocythaemia
Primary myelofibrosis
Chronic neutrophilic leukaemia
Chronic eosinophilic leukaemia
Juvenile myelomonocytic leukaemia
Myeloproliferative neoplasm, not otherwise specified

SMD

Table 3. Classification and defining features of myelodysplastic neoplasms (MDS).

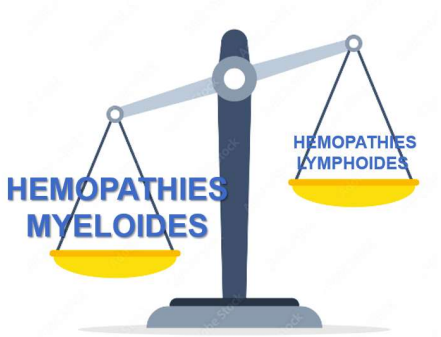
	Blasts	Cytogenetics	Mutations
MDS with defining genetic abnormalities			
MDS with low blasts and isolated 5q deletion (MDS-5q)	<5% BM and <2% PB	5q deletion alone, or with 1 other abnormality other than monosomy 7 or 7q deletion	
MDS with low blasts and *SF3B1* mutation[a] (MDS-*SF3B1*)		Absence of 5q 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[b] (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		

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

Formes frontières

Table 5. Myelodysplastic/myeloproliferative neoplasms.

Chronic myelomonocytic leukaemia
Myelodysplastic/myeloproliferative neoplasm with neutrophilia
Myelodysplastic/myeloproliferative neoplasm with *SF3B1* mutation and thrombocytosis
Myelodysplastic/myeloproliferative neoplasm, not otherwise specified



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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		

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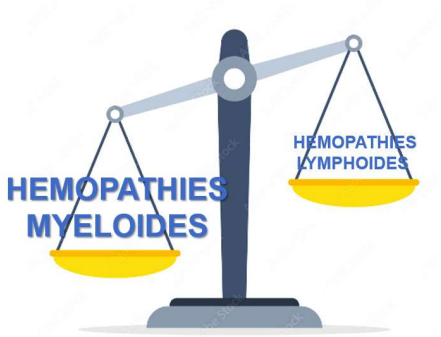
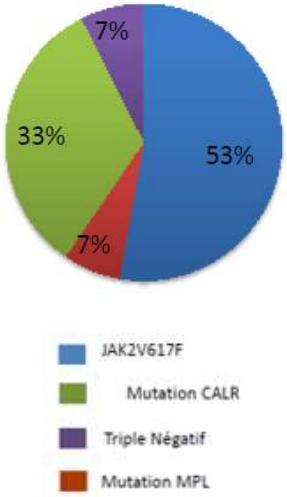


Apport du NGS dans la MF

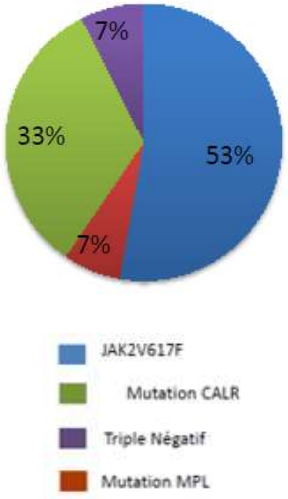
DIAGNOSTIC

Analyse du trio : **JAK2 « full exon » / CALR / MPL**
ET d'un panel de gènes « myéloïdes » en 1 seule étape
→ caractérisation des TN +++



Grinfeld et al N Engl J Med. 2018
Tefferi et al Am J Hematol. 2020
Coltro et al Blood. 2020

Apport du NGS dans la MF



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→ caractérisation des TN +++

PRONOSTIC

Mutations à valeur pronostique

/ risque de transformation en LA

SCORE MIPSS70+ ET GIPSS

CARYOTYPE

Grinfeld et al N Engl J Med. 2018
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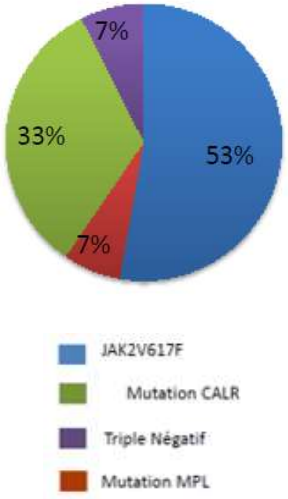
Coltro et al Blood. 2020

Apport du NGS dans la MF



DIAGNOSTIC

Analyse du trio : **JAK2 « full exon » / CALR / MPL**
ET d'un panel de gènes « myéloïdes » en 1 seule étape
→ caractérisation des TN +++

CHIP

PRONOSTIC

Mutations à valeur pronostique

/ risque de transformation en LA

SCORE MIPSS70+ ET GIPSS

CARYOTYPE

REPONSE AU TRTT

Mutations de résistance aux anti-JAK2

RAS
CBL

Grinfeld et al N Engl J Med. 2018
Tefferi et al Am J Hematol. 2020
Coltro et al Blood. 2020

Apport du NGS dans la MF

TABLE 5 New prognostic models in primary myelofibrosis (*see text for references*)

	MIPSS70 (3-tiered)		MIPSS70+ version 2.0 (5-tiered)		GIPSS (4-tiered)
	Genetic variables	Clinical variables	Genetic variables	Clinical variables	Genetic variables
	One HMR mutation (1 point)	Hemoglobin < 10 g/dL (1 point)	VHR karyotype (4 points)	Severe anemia (2 points)	VHR karyotype (2 points)
	≥ 2 HMR mutations (2 points)	Leukocytes > 25 × 10(9)/l (2 points)	Unfavorable karyotype (3 points)	Moderate anemia (1 point)	Unfavorable karyotype (1 point)
	Type 1/like CALR absent (1 point)	Platelets < 100 × 10(9)/L (2 points)	≥ 2 HMR mutations (3 points)	Circulating blasts ≥ 2% (1 point)	Type 1/like CALR absent (1 point)
		Circulating blasts ≥ 2% (1 point)	One HMR mutation (2 points)	Constitutional symptoms (2 points)	ASXL1mutation (1 point)
		Constitutional symptoms (1 point)	Type 1/like CALR absent (2 points)		SRSF2 mutation (1 point)
		Bone marrow fibrosis grade ≥ 2 (1 point)			U2AF1Q157 mutation (1 point)
Very low risk (median survival)			Zero points (not reached)		
Low risk (median survival)	0-1 points (not reached)		1–2 points (16.4 y)		Zero points (26.4 y)
Intermediate-1 risk (median survival)					One point (8 y)
Intermediate risk (median survival)	2–4 points (6.3 y)		3-4 points (7.7 y)		
Intermediate-2 risk (median survival)					Two points (4.2 y)
High risk (median survival)	≥5 points (3.1 y)		5–8 points (4.1 y)		≥3 points (2 y)
Very high risk (median survival)			≥9 points (1.8 y)		

ASXL1
SRSF2
EZH2
IDH1
IDH2

Note: Severe anemia, Hemoglobin <8 g/dL in women and < 9 g/dL in men; Moderate anemia, Hemoglobin 8-9.9 in women and 9-10.9 in men.
Abbreviations: GIPSS, genetically-inspired prognostic scoring system. *Survival quotes are for all age groups*; HMR, high molecular risk mutations include *ASXL1*, *SRSF2*, *EZH2*, *IDH1*, *IDH2* and, in addition, for GIPSS and MIPSS70+ version 2.0, *U2AF1Q157*; MIPSS70, mutation-enhanced international prognostic system for transplant-age patients (age ≤ 70 years); MIPSS70+ version 2.0, mutation and karyotype enhanced international prognostic system. *Survival quotes are for age ≤ 70 years*; VHR, very high risk karyotype.

Apport du NGS dans la MF

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	Type 1/like CALR absent (1 point)	Platelets < 100 × 10(9)/L (2 points)	≥ 2 HMR mutations (3 points)	Circulating blasts ≥ 2% (1 point)	Type 1/like CALR absent (1 point)
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Apport du NGS dans la MF

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ASXL1
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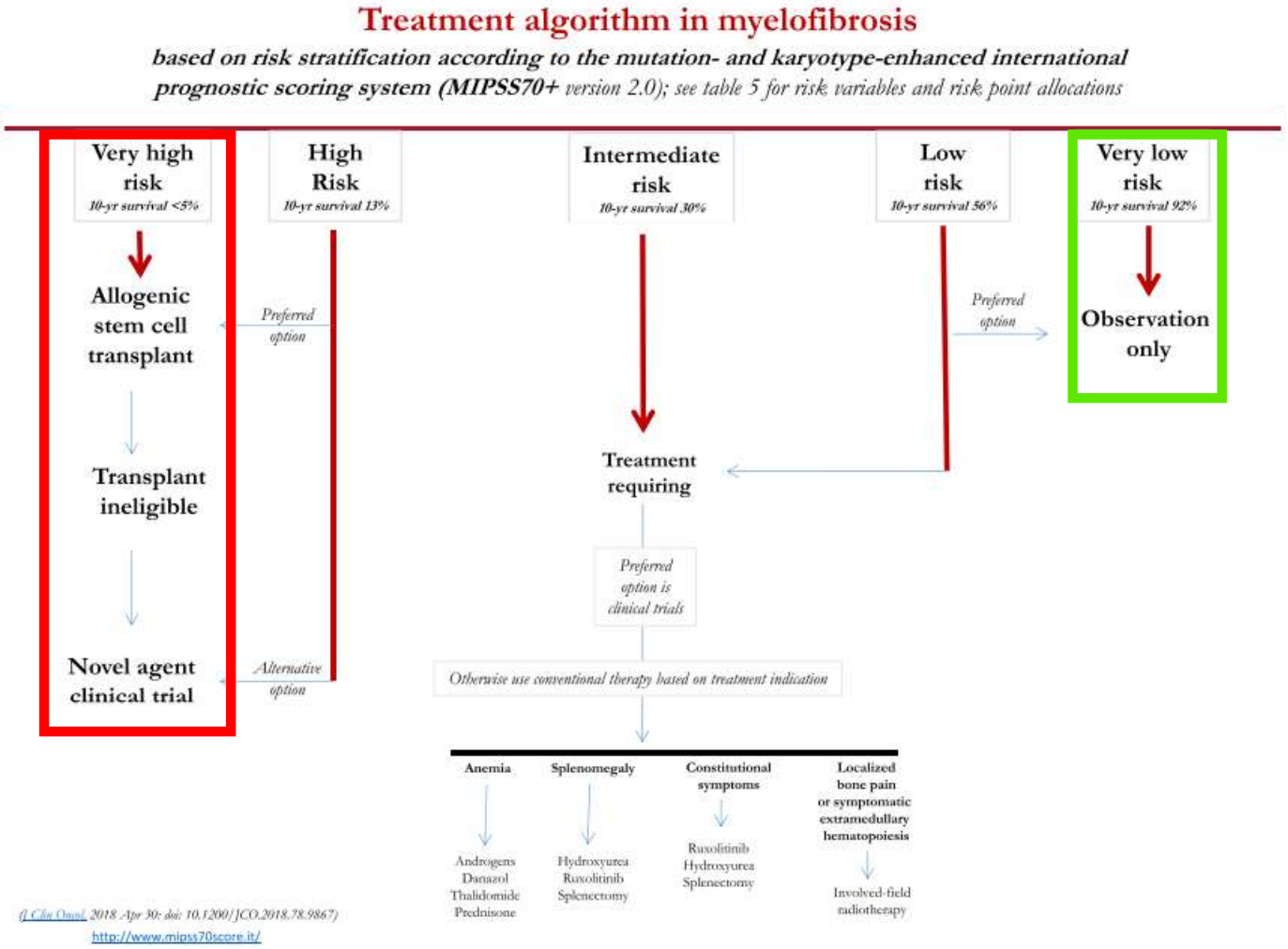
Apport du NGS dans la MF



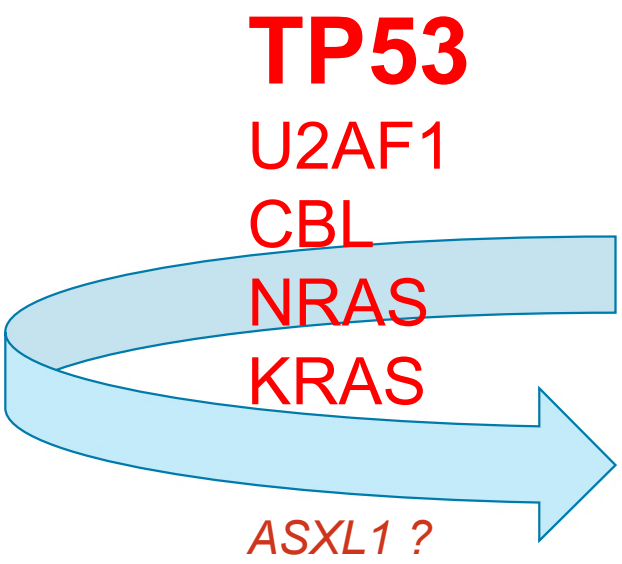
Et d'autres ?

REGULAR ARTICLE

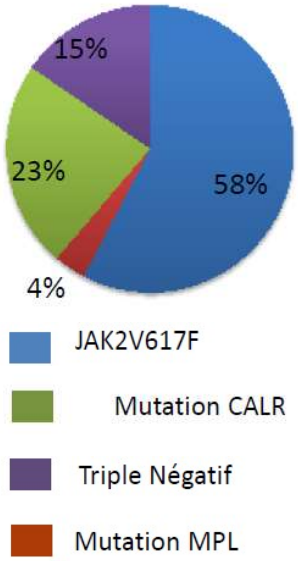
blood advances

Genomic analysis of primary and secondary myelofibrosis redefines the prognostic impact of *ASXL1* mutations: a FIM study

Damien Luque Paz,[1-3] Jérémie Riou,[4] Emmanuelle Verger,[5,6] Bruno Cassinat,[5,6] Aurélie Chauveau,[7] Jean-Christophe Ianotto,[8] Brigitte Dupriez,[9] Françoise Boyer,[10] Maxime Renard,[1-3] Olivier Mansier,[11,12] Anne Murati,[13] Jérôme Rey,[14] Gabriel Etienne,[15] Véronique Mansat-De Mas,[16] Suzanne Tavitian,[17] Olivier Nibourel,[18,19] Stéphane Girault,[20] Yannick Le Bris,[21,22] François Girodon,[23] Dana Ranta,[24] Jean-Claude Chomel,[25] Pascale Cony-Makhoul,[26] Pierre Sujobert,[27] Margot Robles,[28] Raouf Ben Abdelali,[29] Olivier Kosmider,[30] Laurane Cottin,[1-3] Lydia Roy,[31,32] Ivan Sloma,[33,34] Fabienne Vacheret,[35] Mathieu Wemeau,[36] Pascal Mossuz,[37] Borhane Slama,[38] Vincent Cussac,[39] Guillaume Denis,[40] Anouk Walter-Petrich,[41] Barbara Burroni,[42] Nathalie Jézéquel,[7] Stéphane Giraudier,[5,6] Eric Lippert,[7] Gérard Socié,[43] Jean-Jacques Kiladjian,[6,44] and Valérie Ugo,[1-3] on behalf of the French Intergroup of Myeloproliferative Neoplasms



Apport du NGS dans la Thrombocytémie essentielle



DIAGNOSTIC

Analyse du trio : **JAK2 « full exon » / CALR / MPL en 1 seule étape**

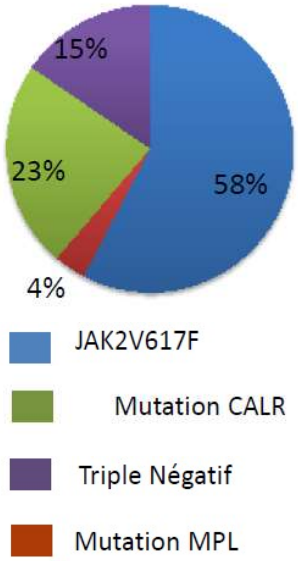
→ Rapidité pour le diagnostic d'une NMP
→ CALR : Sensibilité du NGS !
→ MPL : exhaustivité de l'exon 10 !

Exclure l'entité OMS : **SMD/NMP-SF3B1 muté et Thrombocytose** SF3B1

Autre mutation présente ? = preuve de **clonalité**

Grinfeld et al N Engl J Med. 2018
Tefferi et al Br J Haematol. 2020
Guglielmelli et al - BCJ 2021

Apport du NGS dans la Thrombocytémie essentielle



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PRONOSTIC

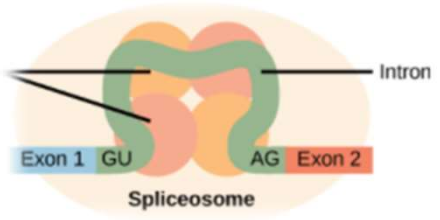
Mutations associées à JAK2/CALR/MPL avec valeur pronostique ?

- au diagnostic
- au suivi

/ risque de thrombose ? ASXL1 RUNX1 EZH2 +

/ si **évolution hématologique**

SF3B1 SRSF2 U2AF1 -

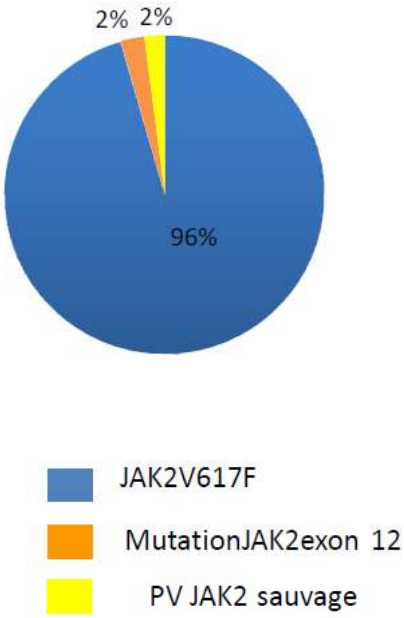


/ risque de transformation en MF / SMD / LA ← TP53

CARYOTYPE

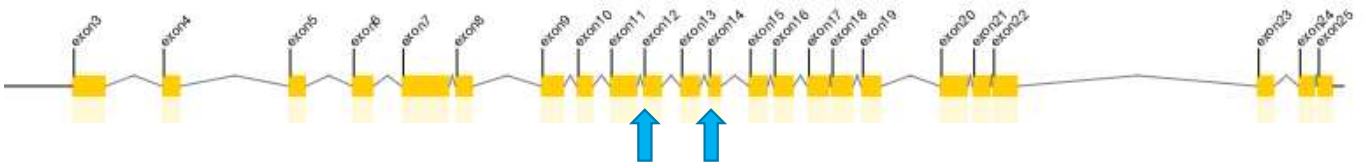
Grinfeld et al N Engl J Med. 2018
Tefferi et al Br J Haematol. 2020
Guglielmelli et al - BCJ 2021

Apport du NGS dans la maladie de Vaquez



DIAGNOSTIC

Analyse **JAK2** « **full exon** »



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CHIP

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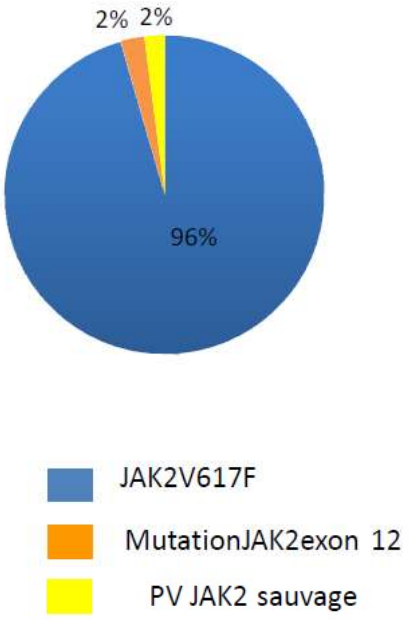
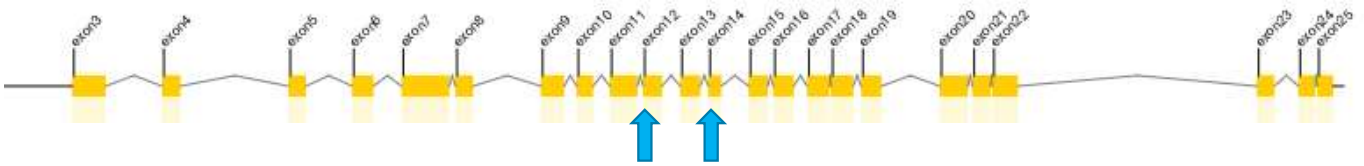


DIAGNOSTIC

Analyse **JAK2 « full exon »**



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CHIP

PRONOSTIC

Mutations associées à JAK2 avec valeur pronostique ?

- au diagnostic

- au suivi

/ risque de thrombose ?

/ si **évolution hématologique**

/ risque de transformation en **MF / LA**

SRSF2

CARYOTYPE

Grinfeld et al N Engl J Med. 2018
Tefferi et al Br J Haematol. 2020.

Et sur le plan **pronostique** pour la PV et la TE ?

OMS 2022 :

OMS 2022 : Intérêt pronostique du NGS dans les NMP

OMS 2022

While *JAK2*, *CALR*, and *MPL* mutations are considered driver events, mutations in other genes – particularly *TET2*, *ASXL1*, and *DNMT3A* – are found in over half of patients with MPN. Mutations affecting splicing regulators (*SRSF2, SF3B1, U2AF1, ZRSR2*) and other regulators of chromatin structure, epigenetic functions and cellular signaling (e.g., *EZH2, IDH1, IDH2, CBL, KRAS, NRAS, STAG2, TP53*) are less common. These additional mutations are more frequent in PMF and advanced disease compared to PV and ET, and some are known to correlate with a poorer prognostic risk (e.g., *EZH2, IDH1, IDH2, SRSF2, U2AF1*, and *ASXL1* mutations in PMF).

→ Aide **diagnostique** +++
→ Aide **pronostique** pour les **MFP** +++
→ Données à préciser pour l'impact **pronostique** d'anomalies moléculaires dans les PV / TE autres que JAK2/CALR/MPL

TET2
ASXL1
DNMT3A

SRSF2
SF3B1
U2AF1
ZRSR2
EZH2
IDH1
IDH2
CBL
KRAS
NRAS
STAG2
TP53

Scores pronostiques MIPSS-PV et MIPSS-TE

Table 3. Clinical-molecular prognostic scores in polycythemia vera and essential thrombocythemia.

Prognostic Score [Reference]	Clinical Variables (Points)	Molecular Variables (Points)	Risk Categories (Points)	Survival *
MIPSS-PV, Tefferi et al. [95]	Leukocyte count $\geq 15 \times 10^9$/L (1); thrombosis history (1); age > 67 years (2)	*SRSF2* mutation (3)	Low (0–1) Intermediate (2–3) High (4–7)	24 13.1 3.2
MIPSS-ET, Tefferi et al. [95]	Leukocyte count $\geq 11 \times 10^9$/L (1); age > 60 years (4); male sex (1)	*SRSF2*, *SF3B1*, *U2AF1*, and *TP53* mutation (2)	Low (0–1) Intermediate (2–5) High (6–8)	34.3 14.1 7.9

MIPSS, Mutation-Enhanced International Prognostic Scoring System; PV, polycythemia vera; ET, essential thrombocythemia. * Survival in years.

PV :
uniquement **SRSF2** !

TE :
4 gènes ! : SRSF2 – SF3B1 – U2AF1 – TP53

~~**ASXL1**~~

utilité du NGS !

Scores pronostiques MIPSS-PV et MIPSS-TE

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MIPSS, Mutation-Enhanced International Prognostic Scoring System; PV, polycythemia vera; ET, essential thrombocythemia. * Survival in years.

NMP

SMD

Table 1. Myeloproliferative neoplasms.

Chronic myeloid leukaemia
Polycythaemia vera
Essential thrombocythaemia
Primary myelofibrosis
Chronic neutrophilic leukaemia
Chronic eosinophilic leukaemia
Juvenile myelomonocytic leukaemia
Myeloproliferative neoplasm, not otherwise specified

Table 3. Classification and defining features of myelodysplastic neoplasms (MDS).

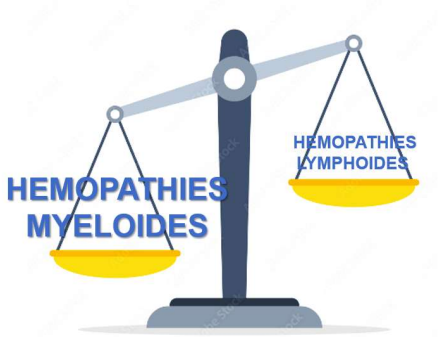
	Blasts	Cytogenetics	Mutations
MDS with defining genetic abnormalities			
MDS with low blasts and isolated 5q deletion (MDS-5q)	<5% BM and <2% PB	5q deletion alone, or with 1 other abnormality other than monosomy 7 or 7q deletion	
MDS with low blasts and *SF3B1* mutation[a] (MDS-*SF3B1*)		Absence of 5q 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[b] (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		

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

Formes frontières

Table 5. Myelodysplastic/myeloproliferative neoplasms.

Chronic myelomonocytic leukaemia
Myelodysplastic/myeloproliferative neoplasm with neutrophilia
Myelodysplastic/myeloproliferative neoplasm with *SF3B1* mutation and thrombocytosis
Myelodysplastic/myeloproliferative neoplasm, not otherwise specified



LMMC – OMS 2022

Table 6. Diagnostic criteria of chronic myelomonocytic leukaemia.

Prerequisite criteria
1. Persistent absolute ($\geq 0.5 \times 10^9$/L) and relative (≥10%) peripheral blood monocytosis.
2. Blasts constitute <20% of the cells in the peripheral blood and bone marrow.[a]
3. Not meeting diagnostic criteria of chronic myeloid leukaemia or other myeloproliferative neoplasms.[b]
4. Not meeting diagnostic criteria of myeloid/lymphoid neoplasms with tyrosine kinase fusions.[c]
Supporting criteria
1. Dysplasia involving ≥1 myeloid lineages.[d]
2. Acquired clonal cytogenetic or molecular abnormality.
3. Abnormal partitioning of peripheral blood monocyte subsets.[e]
Requirements for diagnosis
- Pre-requisite criteria must be present in all cases.
- If monocytosis is $\geq 1 \times 10^9$/L: one or more supporting criteria must be met.
- If monocytosis is ≥0.5 and $<1 \times 10^9$/L: supporting criteria 1 and 2 must be met.
Subtyping criteria
- Myelodysplastic CMML (MD-CMML): WBC $< 13 \times 10^9$/L
- Myeloproliferative CMML (MP-CMML): WBC $\geq 13 \times 10^9$/L
Subgrouping criteria (based on percentage of blasts and promonocytes)
CMML-1: <5% in peripheral blood and <10% in bone marrow
CMML-2: 5–19% in peripheral blood and 10-19% in bone marrow

Caryotype + NGS !!!

~~LMMC-0~~

Apport du NGS dans les LMMC

SANG MOELLE

DIAGNOSTIC

Panel gènes :

→ **TET2 – SRSF2** = aide diagnostique

Gene	Frequency, %		
TET2	29–61	*SF3B1*	6–10
ASXL1	32–44	*ZRSR2*	4–8
DNMT3A	2–12	*CBL*	8–22
EZH2	5–13	*KRAS*	7–16
IDH1[a]	1–2	*NRAS*	4–22
IDH2[a]	6–7	*NF1*	6–7
BCOR	6–7	*JAK2*	1–10
SRSF2	29–52	*RUNX1*	8–23
U2AF1	4–10	*SETBP1*	4–18
		NPM1[b]	1–3
		FLT3[a,b]	1–3

Paloma et al BJH 2019
Itzykson et al. Hemasphere 2018 (reco EHA et ELN)
Elena et al. Blood 2016

Apport du NGS dans les LMMC

SANG MOELLE

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CHIP

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BCOR	6–7
SRSF2	29–52
U2AF1	4–10
SF3B1	6–10
ZRSR2	4–8
CBL	8–22
KRAS	7–16
NRAS	4–22
NF1	6–7
JAK2	1–10
RUNX1	8–23
SETBP1	4–18
NPM1[b]	1–3
FLT3[a,b]	1–3

PRONOSTIC

Mutations à valeur pronostique
/ risque de transformation en LA → allogreffe
ASXL1 / NRAS / RUNX1 / SETBP1 (CPPS-mol)

NPM1 / FLT3 → diagnostic à rediscuter (LMMC-2 / LAM-M4)

CARYOTYPE

Paloma et al BJH 2019
Itzykson et al. Hemasphere 2018 (reco EHA et ELN)
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Apport du NGS dans les LMMC

SANG MOELLE

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U2AF1	4–10	*SETBP1*	4–18
		NPM1[b]	1–3
		FLT3[a,b]	1–3

PRONOSTIC

Mutations à valeur pronostique
/ risque de transformation en LA → allogreffe

ASXL1 / NRAS / RUNX1 / SETBP1 (CPPS-mol)

NPM1 / FLT3 → diagnostic à rediscuter (LMMC-2 / LAM-M4)

CARYOTYPE

CIBLES THERA-NOSTIQUES

IDH1 / IDH2 / FLT3 → rares !!! mais cibles thérapeutiques potentielles

Paloma et al BJH 2019
Itzykson et al. Hemasphere 2018 (reco EHA et ELN)
Elena et al. Blood 2016

https://qxmd.com/calculate/calculator_609/cmml-cpss-mol

Calculator | About | References

CMML CPSS-Mol

Estimate risk of progression to AML in those with CMML using molecular genetics data

Questions

1.	Cytogenetics	High (trisomy 8, complex, a...
2.	ASXL1	Mutated
3.	NRAS	Mutated
4.	RUNX1	Unmutated
5.	SETBP1	Unmutated
6.	BM blasts	≥5%
7.	WBC	≥13 x 10⁹/L
8.	Transfusions	Yes

About

The CPSS-Mol is a new CMML-specific prognostic scoring system (CPSS) that incorporates molecular genetic data resulting in a 4-level integrated clinical/pathological/genetic risk stratification tool. This tool was derived from a cohort of European patients, 93% of whom possessed 1 of 38 somatic mutations. Based on multivariable Cox regression analyses, cytogenetic abnormalities and mutations in RUNX1, NRAS, SETBP1, and ASXL1 were independently associated with overall survival (OS). The CPSS-Mol fully retained its ability to risk stratify survival in an independent validation cohort of CMML patients.

References

Elena C, Galli A, Such E, et al.
Integrating clinical features and genetic lesions in the risk assessment of patients with chronic myelomonocytic leukemia.

Results

Save | Copy Results

Genetic Risk Score

4

Genetic Risk Group

High

CPSS-Mol Score

7

Prognosis

High risk CPSS-Mol risk group: Individuals with this risk profile have a 48% cumulative incidence of AML at 48-months and a median overall survival of 18 months.

NMP

SMD

Table 1. Myeloproliferative neoplasms.

Chronic myeloid leukaemia
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Essential thrombocythaemia
Primary myelofibrosis
Chronic neutrophilic leukaemia
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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[b] (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		

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

Formes frontières

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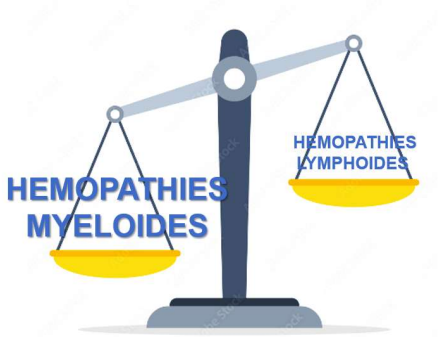
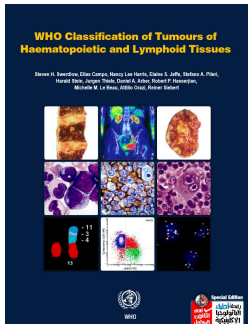
Chronic myelomonocytic leukaemia
Myelodysplastic/myeloproliferative neoplasm with neutrophilia
Myelodysplastic/myeloproliferative neoplasm with *SF3B1* mutation and thrombocytosis
Myelodysplastic/myeloproliferative neoplasm, not otherwise specified



LMC atypique BCR-ABL1 neg – OMS 2017



- Peripheral blood leukocytosis ≥ 13 x 10^9/L, due to increased numbers of neutrophils and their precursors (i.e. promyelocytes, myelocytes and metamyelocytes), with neutrophil precursors constituting ≥ 10% of the leukocytes
- Dysgranulopoiesis, which may include abnormal chromatin clumping
- No or minimal absolute basophilia; basophils constitute < 2% of the peripheral blood leukocytes
- No or minimal absolute monocytosis; monocytes constitute < 10% of the peripheral blood leukocytes
- Hypercellular bone marrow with granulocytic proliferation and granulocytic dysplasia, with or without dysplasia in the erythroid and megakaryocytic lineages
- < 20% blasts in the blood and bone marrow
- No evidence of PDGFRA, PDGFRB or FGFR1 rearrangement, or of PCM1-JAK2
- WHO criteria for BCR-ABL1-positive chronic myeloid leukaemia, primary myelofibrosis, polycythaemia vera, or essential thrombocythaemia[a] are not met

Panel NGS NMP-DG

[a] Myeloproliferative neoplasms (MPNs), in particular those in accelerated phase and/or in post-polycythaemia vera or post-essential thrombocythaemia myelofibrosis, if neutrophilic, may simulate aCML. A history of MPN, the presence of MPN features in the bone marrow, and/or MPN-associated mutations (in JAK2, CALR or MPL) tend to exclude the diagnosis of aCML; conversely, the diagnosis is supported by the presence of SETBP1 and/or ETNK1 mutations. CSF3R mutation is uncommon and, if detected, should prompt careful morphological review to exclude an alternative diagnosis of chronic neutrophilic leukaemia or another myeloid neoplasm.

~~LMC atypique BCR-ABL1 neg – OMS 2017~~

SMD/NMP avec neutrophilie – OMS 2022

- Peripheral blood leukocytosis ≥ 13 x 10^9/L, due to increased numbers of neutrophils and their precursors (i.e. promyelocytes, myelocytes and metamyelocytes), with neutrophil precursors constituting ≥ 10% of the leukocytes
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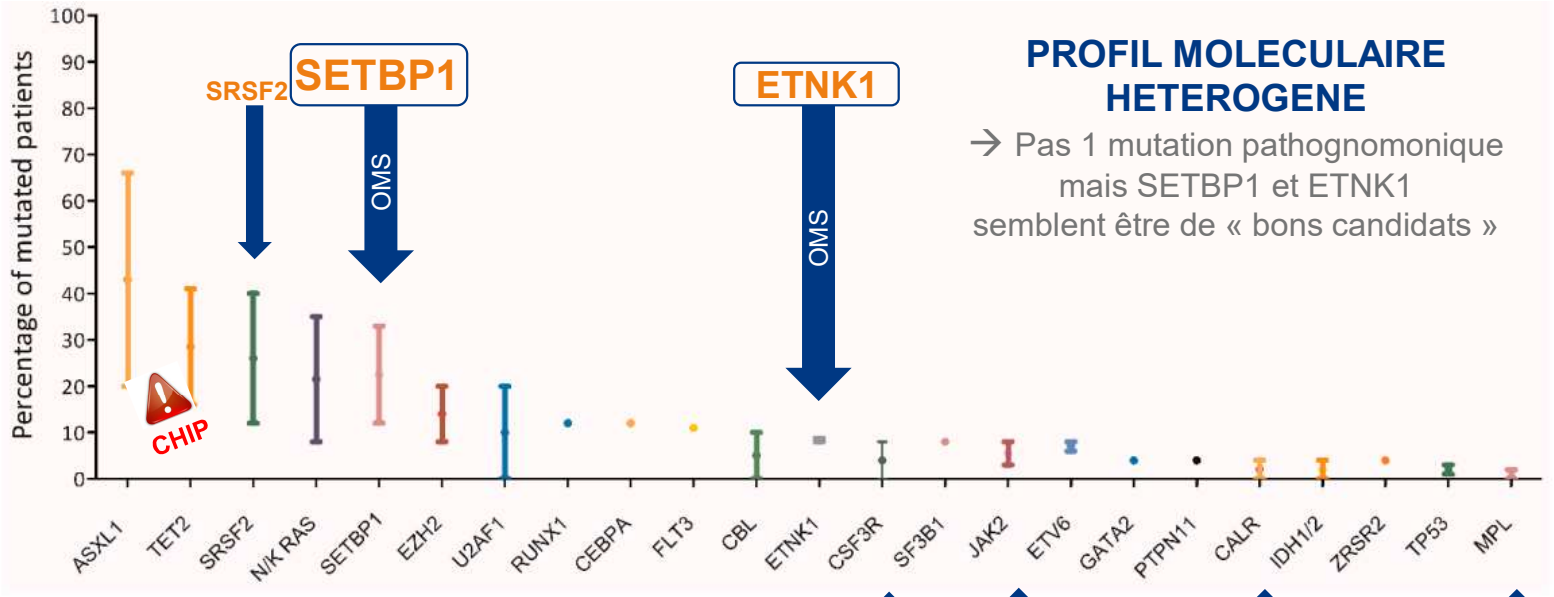
Panel NGS NMP-DG

Apport du NGS dans les SMD/NMP avec neutrophilie

DIAGNOSTIC d'exclusion

~~BCR::ABL~~



PROFIL MOLECULAIRE HETEROGENE

→ Pas 1 mutation pathognomonique mais SETBP1 et ETNK1 semblent être de « bons candidats »

PRONOSTIC –

Et surtout si SETBP1 muté : pronostic péjoratif

NMP

SMD

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Formes frontières

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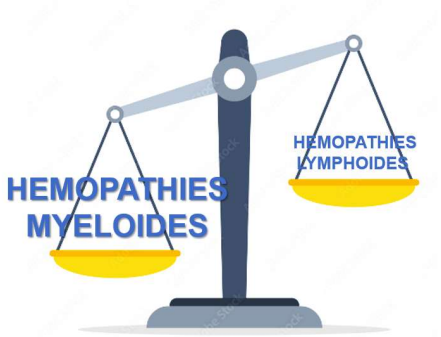
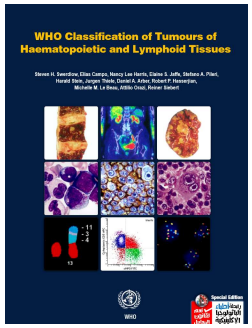


SMD/NMP avec SC et thrombocytose



- Anaemia associated with erythroid-lineage dysplasia, with or without multilineage dysplasia; ≥15% ring sideroblasts[a], < 1% blasts in the peripheral blood and < 5% blasts in the bone marrow
- Persistent thrombocytosis, with platelet count ≥ 450 x 10^9/L
- SF3B1 mutation or, in the absence of SF3B1 mutation, no history of recent cytotoxic or growth factor therapy that could explain the myelodysplastic/myeloproliferative features[b]
- No BCR-ABL1 fusion; no rearrangement of PDGFRA, PDGFRB or FGFR1; no PCM1-JAK2 and no t(3;3)(q21.3;q26.2), inv(3)(q21.3q26.2), or del(5q)[c]
- No history of myeloproliferative neoplasm, myelodysplastic syndrome (except myelodysplastic syndrome with ring sideroblasts), or other myelodysplastic/myeloproliferative neoplasm

Panel NGS NMP-DG

[a] ≥15% ring sideroblasts is a required criterion even if SF3B1 mutation is detected.
[b] The diagnosis of myelodysplastic/myeloproliferative neoplasm with ring sideroblasts and thrombocytosis is strongly supported by the presence of SF3B1 mutation together with a JAK2 V617F, CALR or MPL mutation.
[c] In a case that otherwise meets the diagnostic criteria for myelodysplastic syndrome with isolated del(5q).

Panel NGS NMP-DG

~~SMD/NMP avec SC et thrombocytose~~

SMD/NMP avec mutation SF3B1 et thrombocytose – OMS 2022

(si SF3B1 wt et SC > 15% la même dénomination antérieure est possible)

- Anaemia associated with erythroid-lineage dysplasia, with or without multilineage dysplasia; ≥15% ring sideroblasts[a], < 1% blasts in the peripheral blood and < 5% blasts in the bone marrow
- Persistent thrombocytosis, with platelet count ≥ 450 x 10^9/L
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Panel NGS NMP-DG

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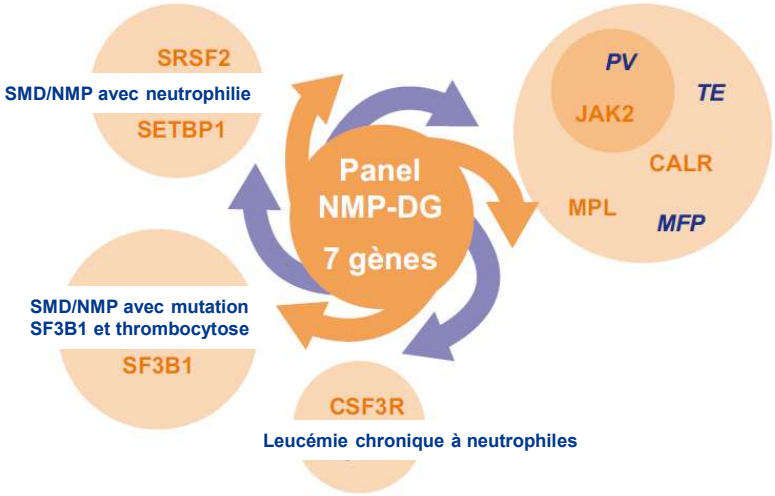
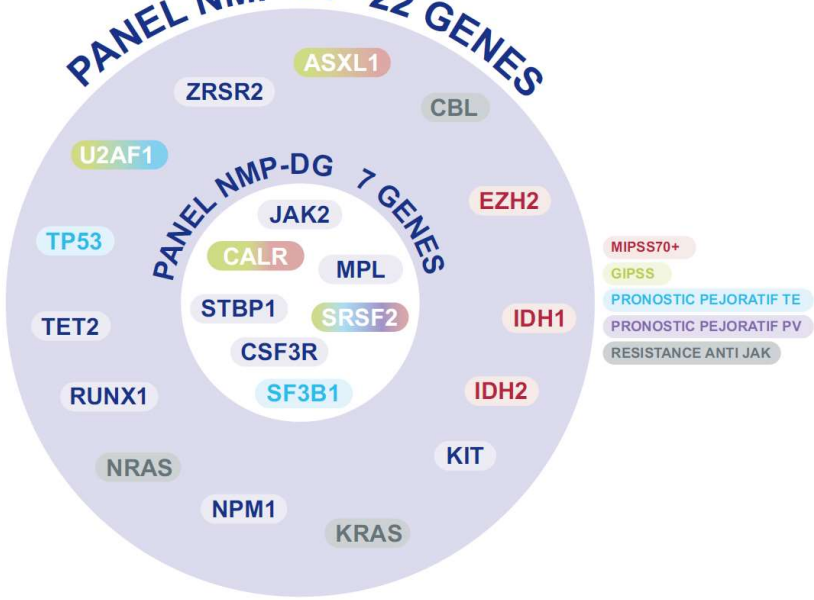
Panel NGS NMP-DG

PCR → NGS ciblé → NGS ciblé → ?

JAK2
CALR
MPL



DIAGNOSTIC



PRONOSTIC

NMP

SMD

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MDS, hypoplastic[b] (MDS-h)			
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Formes frontières

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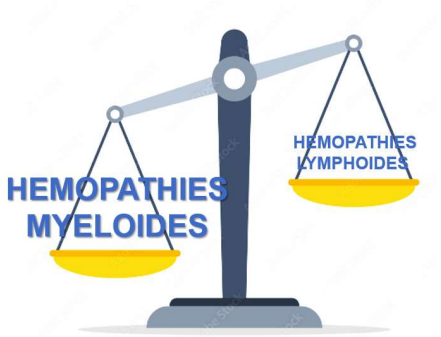
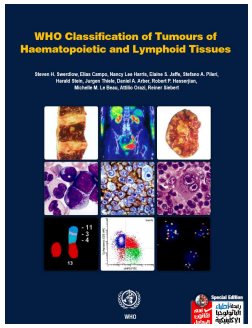


SMD



Entity name	Number of dysplastic lineages	Number of cytopenias[a]	Ring sideroblasts as percentage of marrow erythroid elements	Bone marrow (BM) and peripheral blood (PB) blasts	Cytogenetics by conventional karyotype analysis
MDS-SLD	1	1–2	< 15% / < 5% [b]	BM < 5%, PB < 1%, no Auer rods	Any, unless fulfils all criteria for MDS with isolated del(5q)
MDS-MLD	2–3	1–3	< 15% / < 5% [b]	BM < 5%, PB < 1%, no Auer rods	Any, unless fulfils all criteria for MDS with isolated del(5q)
MDS-RS					
MDS-RS-SLD	1	1–2	≥ 15% / ≥ 5% [b]	BM < 5%, PB < 1%, no Auer rods	Any, unless fulfils all criteria for MDS with isolated del(5q)
MDS-RS-MLD	2–3	1–3	≥ 15% / ≥ 5% [b]	BM < 5%, PB < 1%, no Auer rods	Any, unless fulfils all criteria for MDS with isolated del(5q)
MDS with isolated del(5q)	1–3	1–2	None or any	BM < 5%, PB < 1%, no Auer rods	del(5q) alone or with 1 additional abnormality, except loss of chromosome 7 or del(7q)
MDS-EB					
MDS-EB-1	1–3	1-3	None or any	BM 5-9% or PB 2–4%, BM < 10% and PB < 5%, no Auer rods	Any
MDS-EB-2	1–3	1–3	None or any	BM 10-19% or PB 5-19% or Auer rods, BM and PB < 20%	Any
MDS-U					
with 1% blood blasts	1–3	1–3	None or any	BM < 5%, PB = 1% [c], no Auer rods	Any
with SLD and pancytopenia	1	3	None or any	BM < 5%, PB < 1%, no Auer rods	Any
based on defining cytogenetic abnormality	0	1–3	< 15% [d]	BM < 5%, PB < 1%, no Auer rods	MDS-defining abnormality [e]

MDS-EB, MDS with excess blasts; MDS-MLD, MDS with multilineage dysplasia; MDS-RS, MDS with ring sideroblasts; MDS-RS-MLD, MDS with ring sideroblasts and multilineage dysplasia; MDS-RS-SLD, MDS with ring sideroblasts and single lineage dysplasia; MDS-SLD, MDS with single lineage dysplasia; MDS-U, MDS, unclassifiable; SLD, single lineage dysplasia.

[a] Cytopenias defined as haemoglobin concentration < 10 g/dL, platelet count < 100 x 10^9/L and absolute neutrophil count < 1.8 x 10^9/L, although MDS can present with mild anaemia or thrombocytopenia above these levels; PB monocytes must be < 1 x 10^9/L.
[b] If SF3B1 mutation is present.
[c] 1% PB blasts must be recorded on ≥ 2 separate occasions.
[d] Cases with ≥ 15% ring sideroblasts by definition have significant erythroid dysplasia and are classified as MDS-RS-SLD.

Classification OMS 2017

SF3B1 « évoqué »

?

Panel NGS

SMD et OMS 2022

Classification OMS **2022**

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SF3B1
TP53

?

Panel NGS

SMD et OMS 2022

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MDS with low blasts and *SF3B1* mutation[a] (MDS-*SF3B1*)		Absence of 5q deletion, monosomy 7, or complex karyotype	*SF3B1*
MDS with biallelic *TP53* inactivation (MDS-bi*TP53*) / MDS/AML	<20% BM and PB	Usually complex	Two or more *TP53* mutations, or 1 mutation with evidence of *TP53* copy number loss or cnLOH
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VAF ?

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Les scores actuellement utilisés dans les SMD …

IPSS - 1997

	Prognostic score value				
	0	**0.5**	**1**	**1.5**	**2**
Prognostic category					
Cytogenetics	Good	Intermediate	Poor		
BM blasts, %	≤ 5	5–10		11–20	21–30
Cytopenia[a]	0–1	2–3			

Cytogenetic group	**Characteristics**
Good	Normal, −Y, del(5q), del(20q)
Intermediate	All other karyotypic abnormalities
Poor	Complex (≥ 3 abnormalities) or chromosome 7 abnormalities

MOELLE

Greenberg et al, Blood 1997
Greeberg et al, Blood 2012

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MOELLE

IPSS-R - 2012

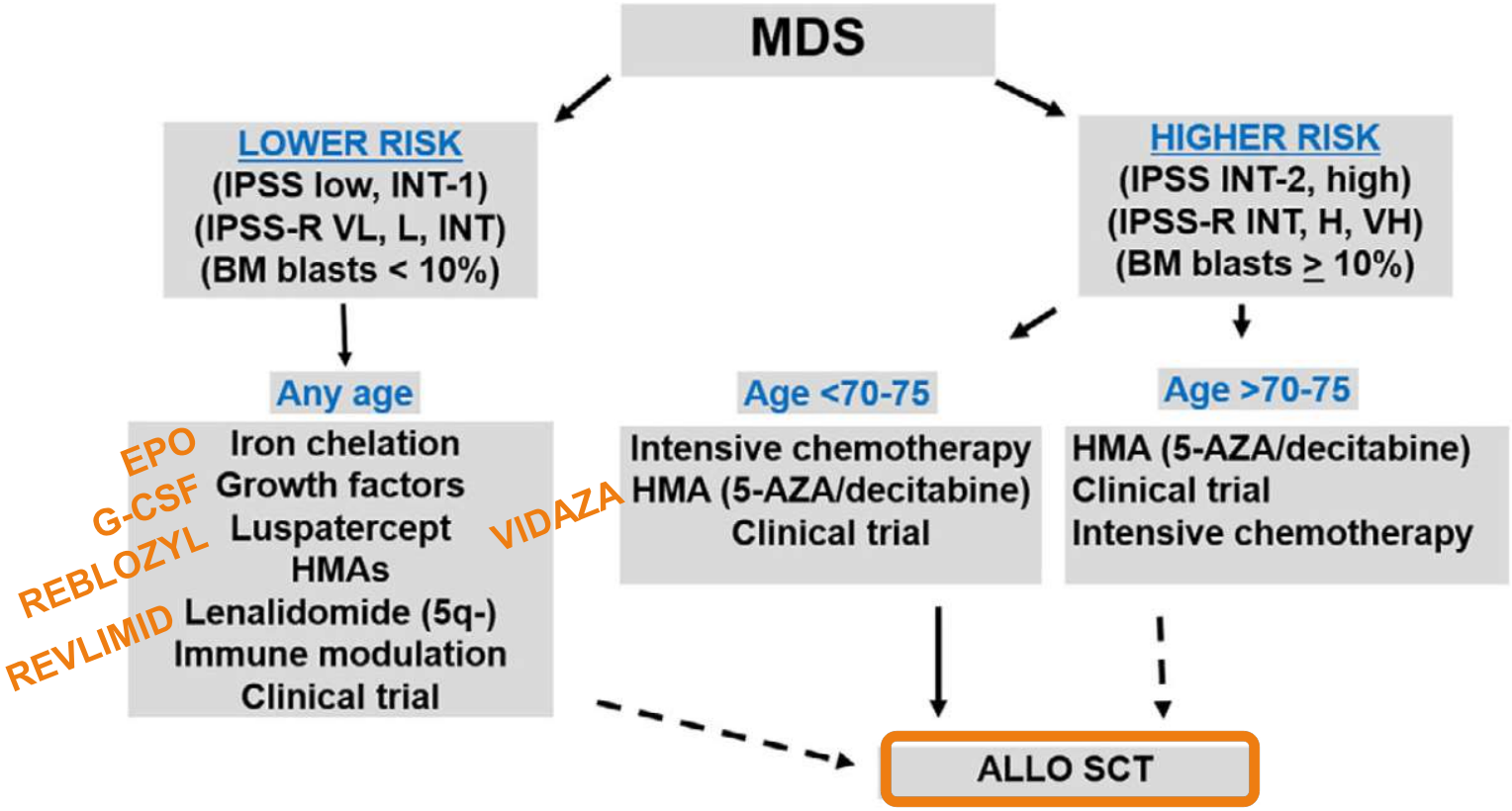
	Prognostic score value						
	0	0.5	1	1.5	2	3	4
Prognostic category							
Cytogenetics	Very good		Good		Intermediate	Poor	Very poor
BM blasts, %	≤ 2		> 2 to < 5		5–10	> 10	
Hgb, g/dL	≥ 10		8 to < 10	< 8			
Platelets, x 10^9/L	≥ 100	50 to < 100	< 50				
ANC, x 10^9/L	≥ 0.8	< 0.8					

Cytogenetic group	Characteristics
Very good	−Y, del(11q)
Good	Normal, del(5q), del(12p), del(20q), del(5q) + 1 additional abnormality
Intermediate	del(7q), +8, +19, i(17q), other abnormalities not in other groups
Poor	−7, inv(3)/t(3q), −7/del(7q) + 1 additional abnormality, complex (3 abnormalities)
Very poor	Complex (> 3 abnormalities)

MOELLE

Greenberg et al, Blood 1997
Greeberg et al, Blood 2012

…pour choisir un traitement adapté !



Garcia-Manero et al, AJH 2020

Apport du NGS dans les SMD

SANG

MOELLE

DIAGNOSTIC

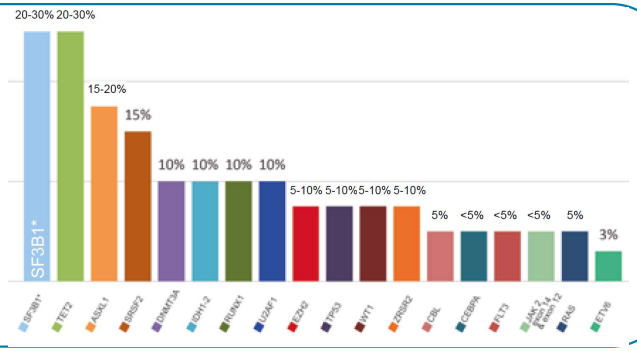
Panel 29 gènes :

→ **SF3B1 = SMD – SC**

CHIP

→ **Clonalité moléculaire** = aide diagnostique +++

Si cytologie médullaire non contributive / caryotype normal (40/50%) !



Apport du NGS dans les SMD

SANG MOELLE

DIAGNOSTIC

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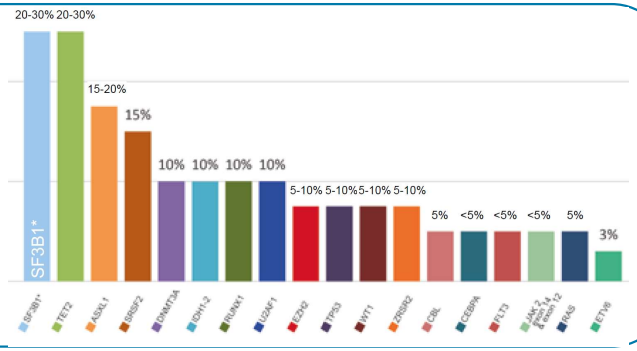

PRONOSTIC

Mutations à valeur pronostique

→ **Favorable : SF3B1** (SC ?)

→ **Défavorable : TP53, EZH2, ETV6, RUNX1, ASXL1, CBL, DNMT3A, IDH1/2 …** / risque de transformation en LAM

→ ***Score moléculaire ?***

CARYOTYPE

Apport du NGS dans les SMD

SANG MOELLE

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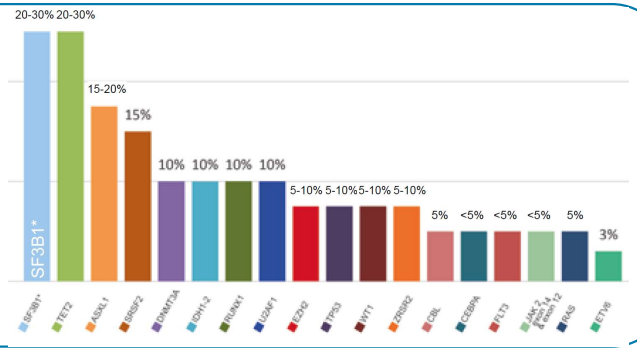

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→ ***Score moléculaire ?***

CARYOTYPE

REPONSE AU TRTT ET CIBLES THERA-NOSTIQUES

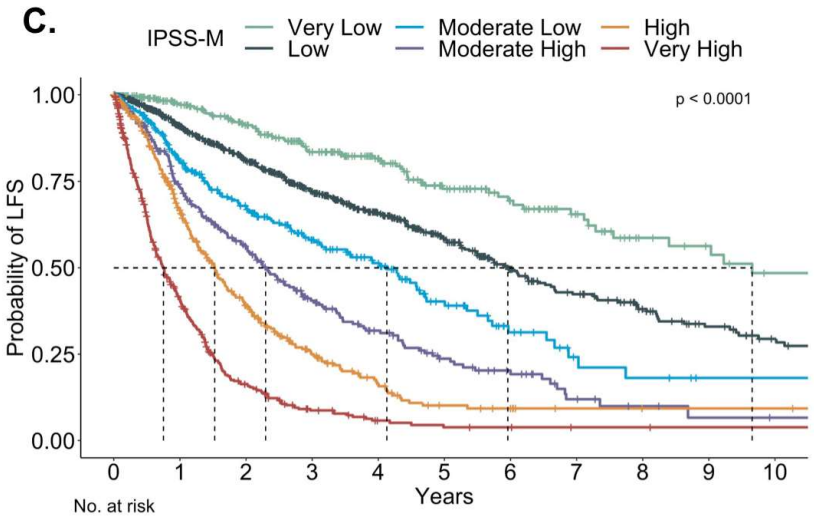
Résistance au lénalidomide pour les Sd 5q- : TP53
Cibles thérapeutiques potentielles : IDH1 / IDH2 / FLT3 → rares
Hypométhylants : TET2 + / ASXL1 –

Score moléculaire dans les SMD

SMD et score IPSS-M

2957 patients
Score IPSS-M :
- Hb
- Plaquettes
- % de blastes
- IPSS-R cytogénétique

16 gènes principaux
TP53, MLL, FLT3, SF3B1, NPM1, NRAS, ETV6, IDH2, CBL, EZH2, U2AF1, SRSF2, DNMT3A, ASXL1, KRAS
15 gènes « secondaires »
BCOR, BCORL1, CEBPA, ETNK1, GATA2, GNB1, IDH1, NF1, PHF6, PPMD1, PRPF8, PTPN11, SETBP1, STAG2, WT1



Bernard et al, ASH Abstract 2021

SMD et score IPSS-M

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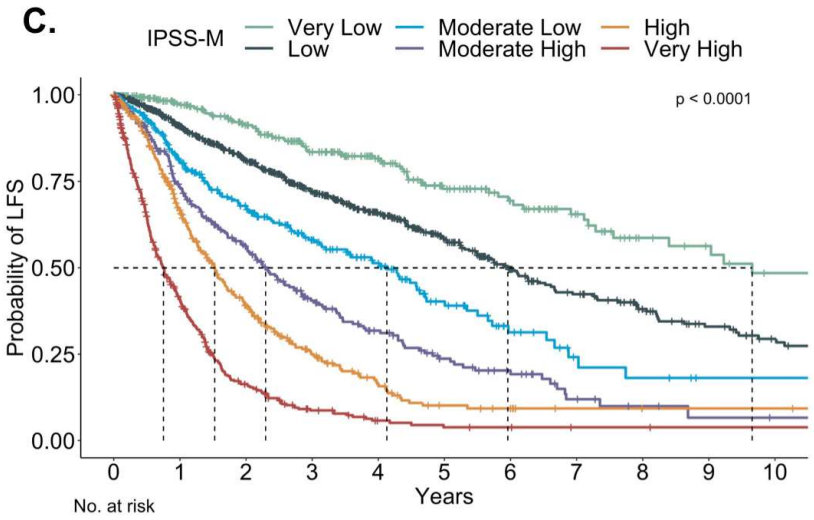
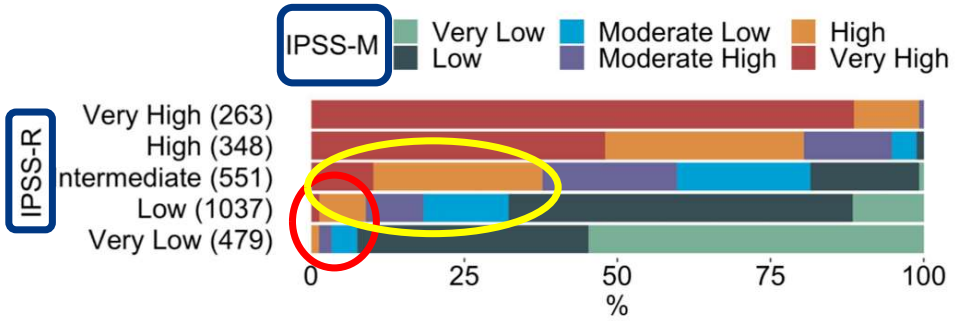


Comparaison IPSS-R et IPSS-M :



46% des patients changent de groupe pronostique en intégrant les données moléculaires :
→ 74% basculent en risque plus élevé
→ 26% basculent en risque moins élevé
→ 6% des faibles risques basculent en haut risque !

Bernard et al, ASH Abstract 2021

NEJM Evidence

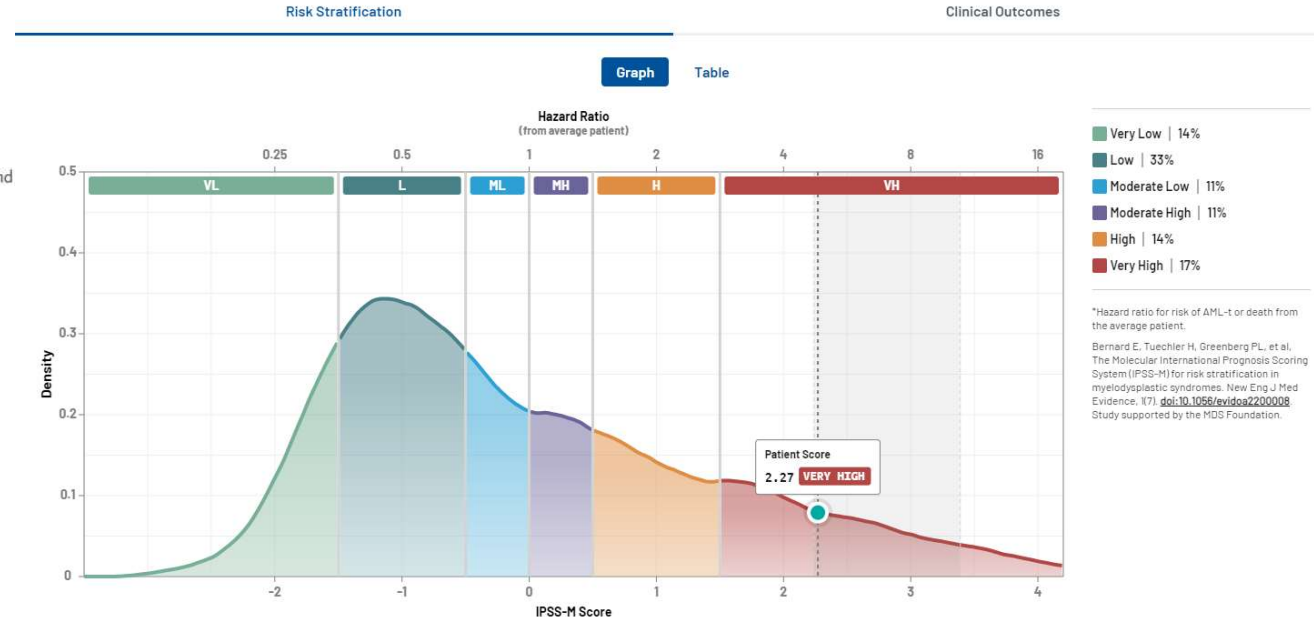
Published June 12, 2022
NEJM Evid 2022; 1 (7)
DOI: 10.1056/EVIDoa2200008

https://mds-risk-model.com/

ORIGINAL ARTICLE

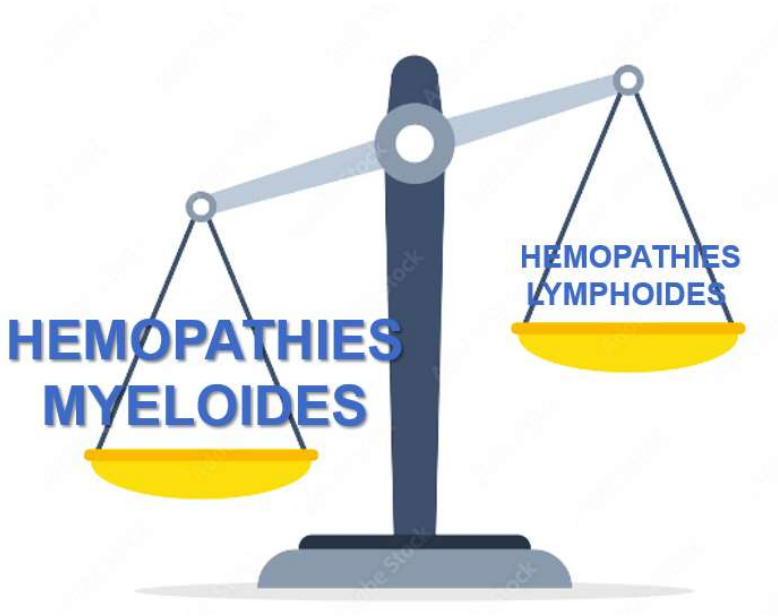
Molecular International Prognostic Scoring System for Myelodysplastic Syndromes

Elsa Bernard, Ph.D.,[1] Heinz Tuechler, Peter L. Greenberg, M.D.,[2] Robert P. Hasserjian, M.D.,[3] Juan E. Arango Ossa, M.S.,[1] Yasuhito Nannya, M.D., Ph.D.,[4,5] Sean M. Devlin, Ph.D.,[1] Maria Creignou, M.D.,[6] Philippe Pinel, M.S.,[1] Lily Monnier, M.S.,[1] Gunes Gundem, Ph.D.,[1] Juan S. Medina-Martinez, M.S.,[1] Dylan Domenico, B.S.,[1] Martin Jädersten, M.D., Ph.D.,[6] Ulrich Germing, M.D.,[7] Guillermo Sanz, M.D., Ph.D.,[8,9,10] Arjan A. van de Loosdrecht, M.D., Ph.D.,[11] Olivier Kosmider, M.D., Ph.D.,[12] Matilde Y. Follo, Ph.D.,[13] Felicitas Thol, M.D.,[14] Lurdes Zamora, Ph.D.,[15] Ronald F. Pinheiro, Ph.D.,[16] Andrea Pellagatti, Ph.D.,[17] Harold K. Elias, M.D.,[18] Detlef Haase, M.D., Ph.D.,[19] Christina Ganster, Ph.D.,[19] Lionel Ades, M.D., Ph.D.,[20] Magnus Tobiasson, M.D., Ph.D.,[6] Laura Palomo, Ph.D.,[21] Matteo Giovanni Della Porta, M.D.,[22] Akifumi Takaori-Kondo, M.D., Ph.D.,[23] Takayuki Ishikawa, M.D., Ph.D.,[24] Shigeru Chiba, M.D., Ph.D.,[25] Senji Kasahara, M.D., Ph.D.,[26] Yasushi Miyazaki, M.D., Ph.D.,[27] Agnes Viale, Ph.D.,[28] Kety Huberman, B.S.,[28] Pierre Fenaux, M.D., Ph.D.,[20] Monika Belickova, Ph.D.,[29] Michael R. Savona, M.D.,[30] Virginia M. Klimek, M.D.,[18] Fabio P. S. Santos, M.D., Ph.D.,[31] Jacqueline Boultwood, Ph.D.,[17] Ioannis Kotsianidis, M.D., Ph.D.,[32] Valeria Santini, M.D.,[33] Francesc Solé, Ph.D.,[21] Uwe Platzbecker, M.D.,[34] Michael Heuser, M.D.,[14] Peter Valent, M.D.,[35,36] Kazuma Ohyashiki, M.D., Ph.D.,[37] Carlo Finelli, M.D.,[38] Maria Teresa Voso, M.D.,[39] Lee-Yung Shih, M.S.,[40] Michaela Fontenay, M.D., Ph.D.,[12] Joop H. Jansen, Ph.D.,[41] José Cervera, M.D., Ph.D.,[42] Norbert Gattermann, M.D.,[7] Benjamin L. Ebert, M.D., Ph.D.,[43] Rafael Bejar, M.D., Ph.D.,[44] Luca Malcovati, M.D.,[45] Mario Cazzola, M.D.,[45] Seishi Ogawa, M.D., Ph.D.,[4,46,47] Eva Hellström-Lindberg, M.D., Ph.D.,[6] and Elli Papaemmanuil, Ph.D.[1]



LLC et autres LMNH-B

Mature B-cell neoplasms	
Pre-neoplastic and neoplastic small lymphocytic proliferations	
Monoclonal B-cell lymphocytosis	(Same)
Chronic lymphocytic leukaemia/small lymphocytic lymphoma	(Same)
(Entity deleted)	B-cell prolymphocytic leukaemia
Splenic B-cell lymphomas and leukaemias	
Hairy cell leukaemia	(Same)
Splenic marginal zone lymphoma	(Same)
Splenic diffuse red pulp small B-cell lymphoma	(Same)
Splenic B-cell lymphoma/leukaemia with prominent nucleoli	*Not previously included* (encompassing hairy cell leukaemia variant and some cases of B-cell prolymphocytic leukaemia)
Lymphoplasmacytic lymphoma	
Lymphoplasmacytic lymphoma	(Same)
Marginal zone lymphoma	
Extranodal marginal zone lymphoma of mucosa-associated lymphoid tissue	(Same)
Primary cutaneous marginal zone lymphoma	*Not previously included* (originally included under "extranodal marginal zone lymphoma of mucosa-associated lymphoid tissue")
Nodal marginal zone lymphoma	(Same)
Paediatric marginal zone lymphoma	(Same)



NGS et LMNH

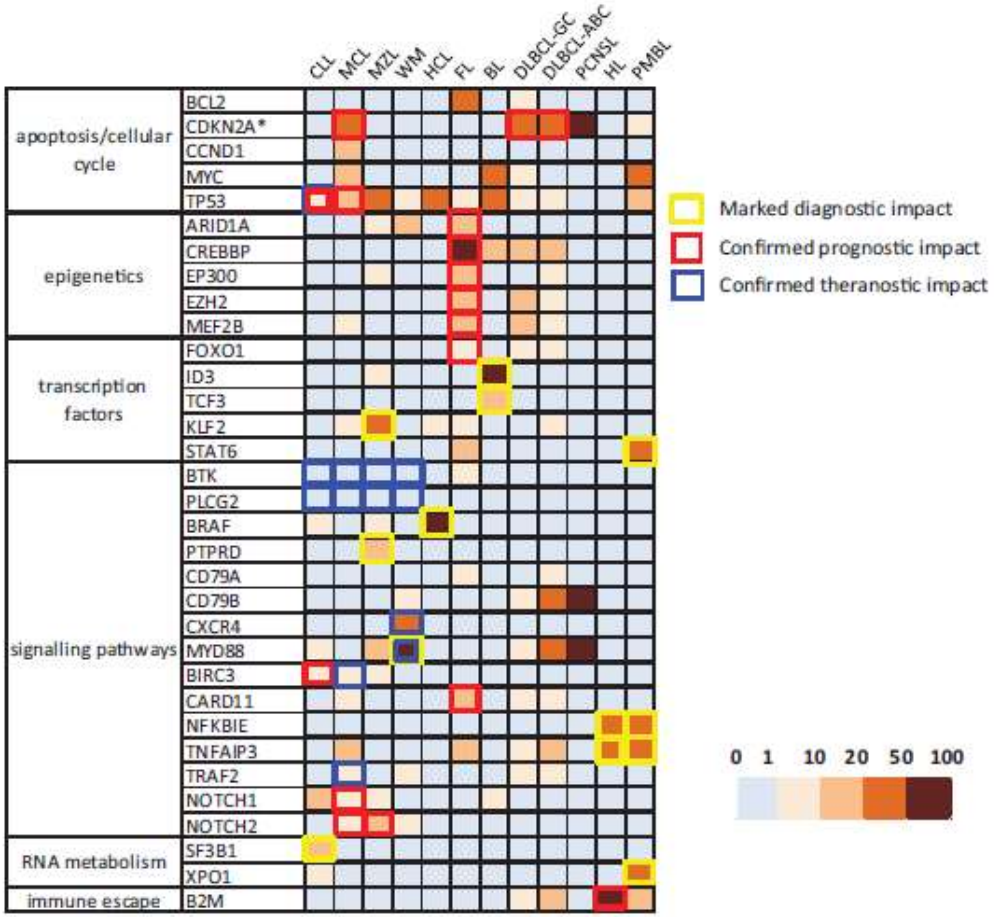


Figure 1. A heatmap representation of the prevalence of gene alterations in mature B lymphoid malignancies from the LYSA/GBMHM consensus panel. The * symbol for CDKN2A underlines that this locus is altered by deletions (and not mutations). The borders of the squares are colored when the alteration has a clear clinical impact in a particular lymphoma subtype (diagnostic in yellow, prognostic in red, and theranostic in blue). ABC = activated B cell, BL = Burkitt lymphoma, CLL = chronic lymphocytic leukemia, DLBLCL = diffuse large B cell lymphoma, FL = follicular lymphoma, GC = germinal center, HCL = hairy cell lymphoma, HL = Hodgkin lymphoma, MCL = mantle cell lymphoma, MZL = marginal zone lymphoma, PCNSL = primary central nervous system lymphoma, PMBCL = primary mediastinal B cell lymphoma, WM = Waldenström macroglobulinemia.

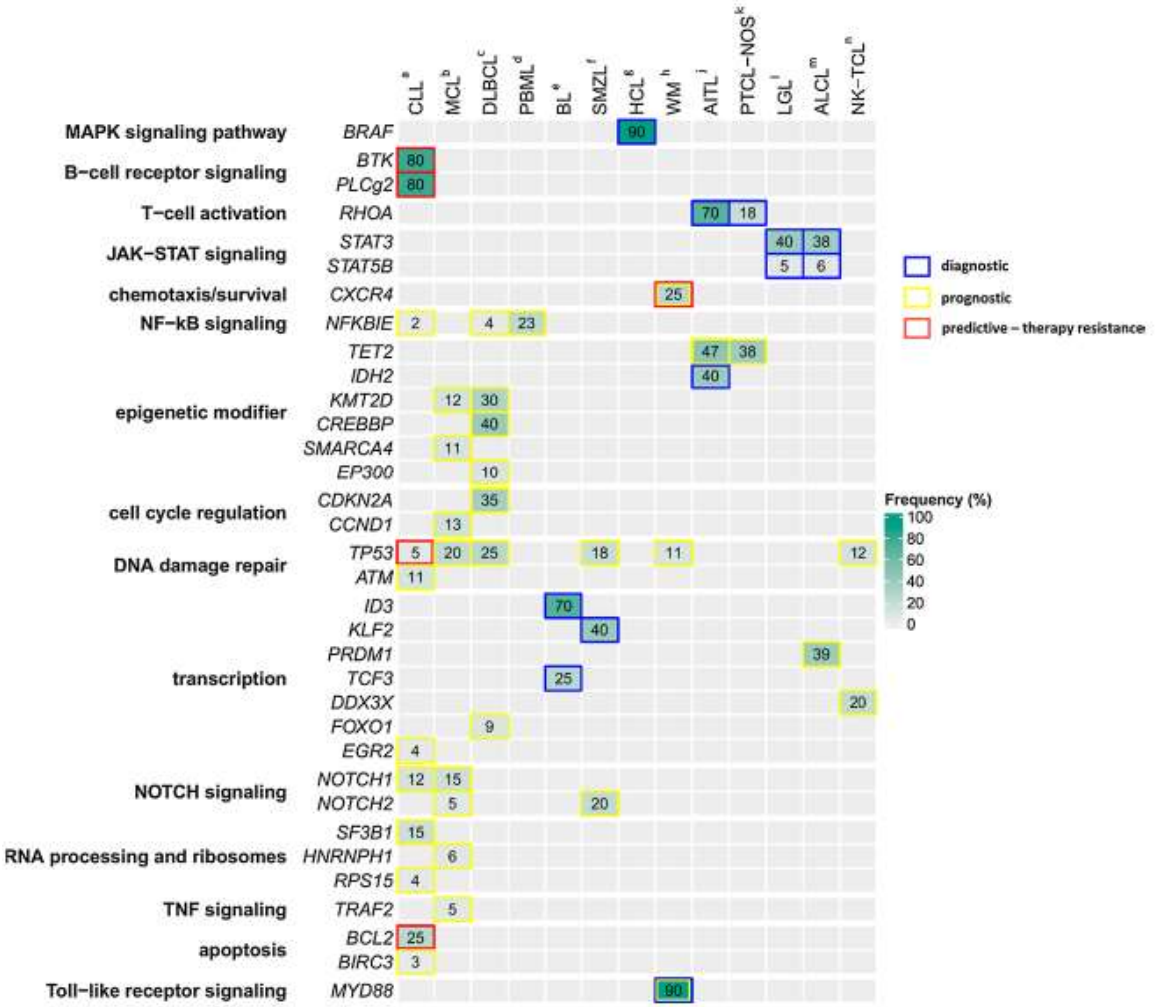
AIDE

DIAGNOSTIQUE

PRONOSTIQUE

THERANOSTIQUE

NGS et LMNH



AIDE

DIAGNOSTIQUE

PRONOSTIQUE

THERANOSTIQUE

Apport du NGS dans la LLC

DIAG

PRONOSTIC

DEFAVORABLE

- NOTCH1 (10%)
- SF3B1
- **TP53**
- XPO1
- NFKBIE
- POT1
- BIRC3
- EGR2
- ATM
- RPS15

TP53 / NOTCH1 / XPO1
TP53 /SF3B1

IGVH

Caryotype
NOTCH1 ⟺ +12
XPO1 ⟺ del(11q)
BIRC3 ⟺ del(11q)
MYD88 ⟺ del(13q)

Score pronostique : CLL-IPI

PAS DE VALEUR PRONOSTIQUE

MYD88

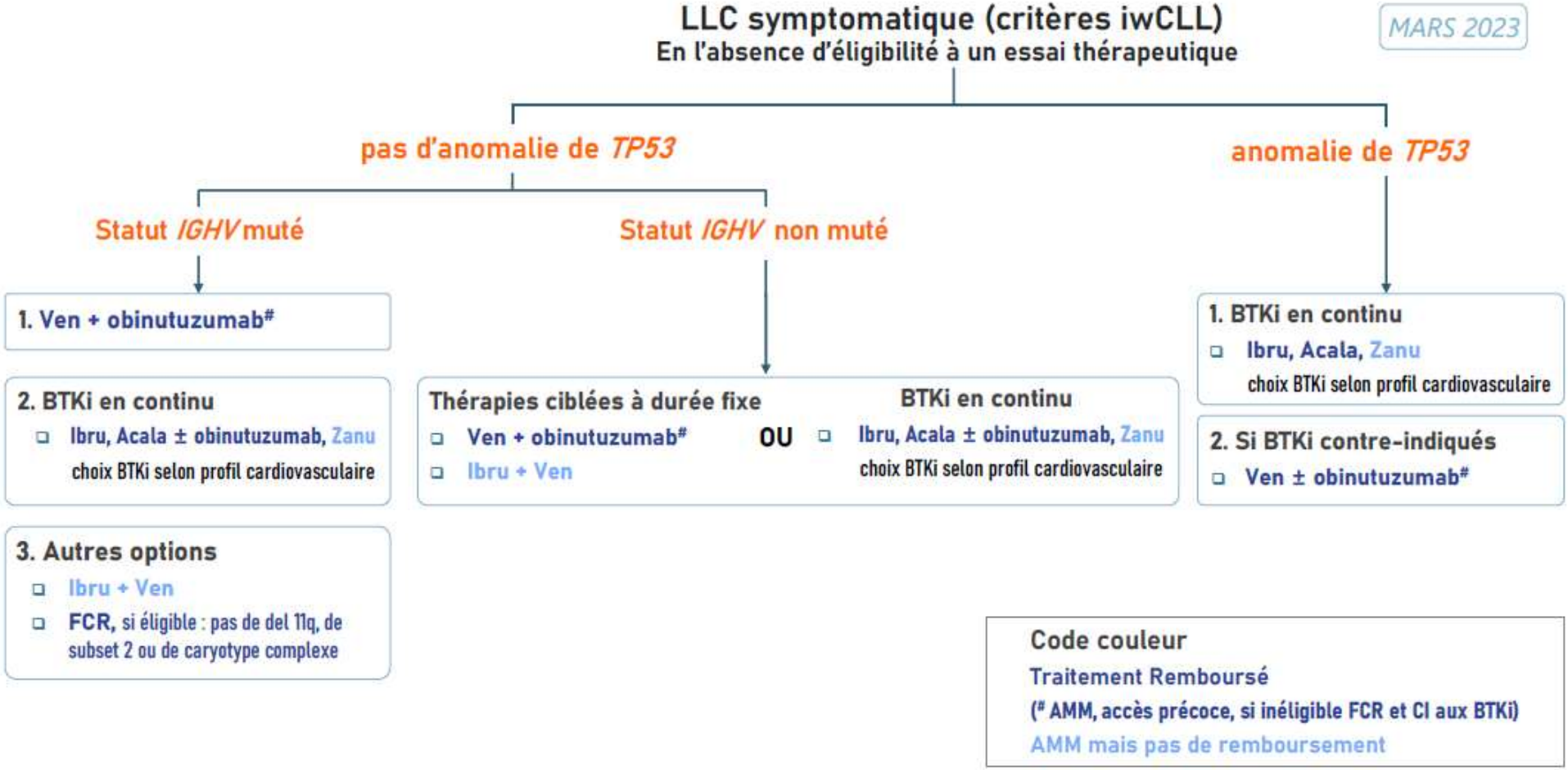
REPONSE AU TRTT

TP53 → BTKi (Ibru/Acala) ⟺ R → BTK / PLCG2

R Venetoclax : BCL2 / MCL1 / BAX / BRAF

Mansouri et al 2022
Hallek et al. 2021
Brieghel et a. 2020

LLC | **recommandations FILO** | algorithme de **traitement de 1re ligne**



Apport du NGS dans la maladie de Waldenström

DIAGNOSTIC

MYD88 (90%)

CXRC4 (40%)

ARID1A
CD79B
NOTCH2

PRONOSTIC

DEFAVORABLE

TP53

CXCR4

REPONSE AU TRTT

R → BTKi (Ibrutinib) CXCR4 / BTK / PLCG2

MYD88 S → Rituximab

Mansouri et al 2022
Heiman et al.2020

Prise en charge des NGS

GOUVERNEMENT
Liberté
Égalité
Fraternité

Evaluation de la HAS des actes inscrits au RIHN

(Référentiel des actes Innovants Hors Nomenclature)

- A l'exception du BCR-ABL (08/04/21 – B460), **aucun acte de biologie moléculaire somatique n'est inscrit à la NABM**
- **Pourtant ces actes sont DIAGNOSTIQUE / PRONOSTIQUE / THERANOSTIQUE**
- En pratique : utilisation du RIHN ou HN pour la cotation de ces actes

Prise en charge des NGS

GOUVERNEMENT
Liberté
Égalité
Fraternité

Evaluation de la HAS des actes inscrits au **RIHN**

(Référentiel des actes Innovants Hors Nomenclature)

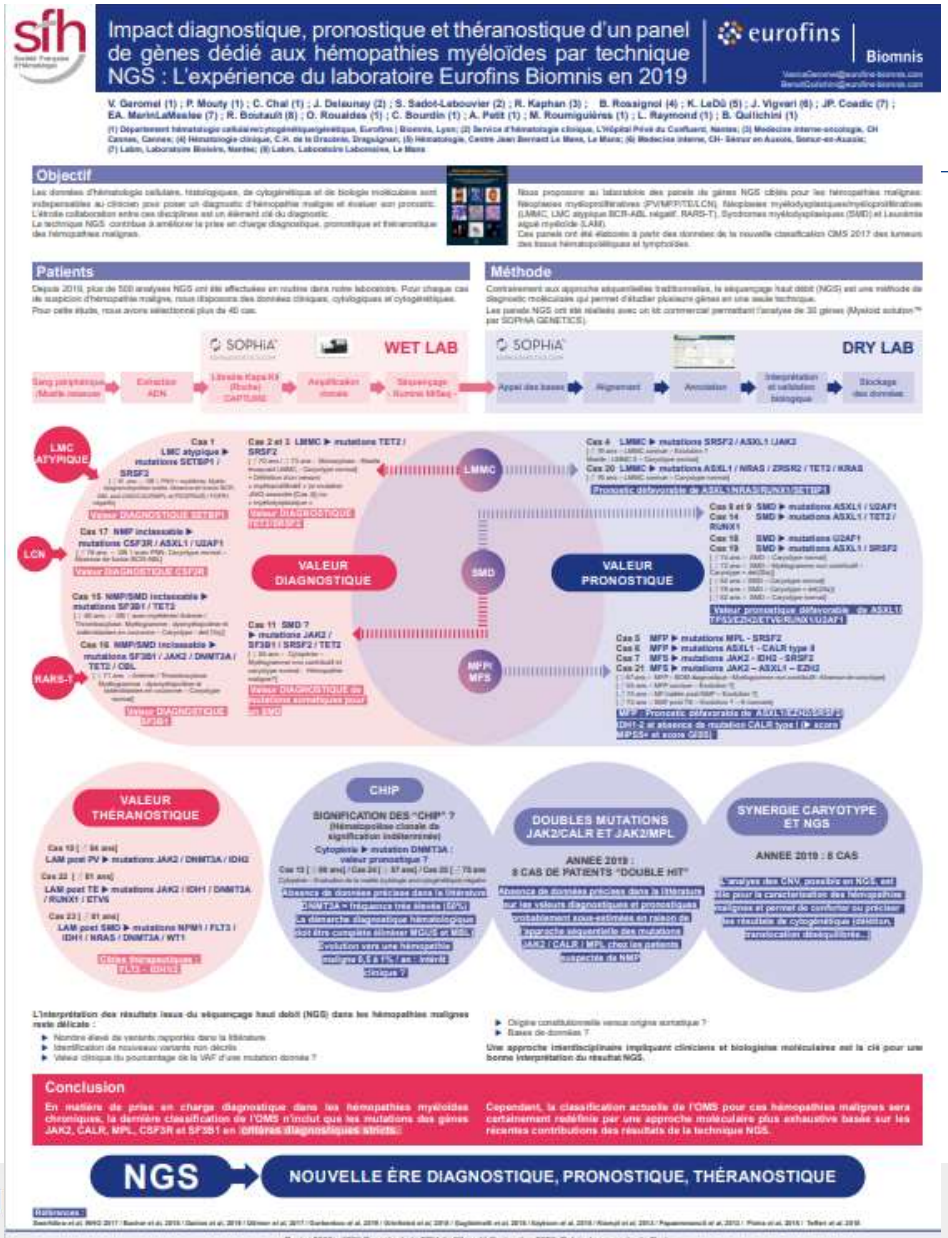
- A l'exception du BCR-ABL (08/04/21 - B460), **aucun acte de biologie moléculaire somatique n'est inscrit à la NABM**
- **Pourtant ces actes sont DIAGNOSTIQUE / PRONOSTIQUE / THERANOSTIQUE**
- En pratique : utilisation du RIHN ou HN pour la cotation de ces actes :

N452	Forfait séquençage haut débit (NGS) < 20 kb	882,90 €	Selon indications fixées par l'INCa et la DGOS
N453	Forfait séquençage haut débit (NGS) > 20 kb et < 100 kb	1 503,90 €	Selon indications fixées par l'INCa et la DGOS
N454	Forfait séquençage haut débit (NGS) > 100 kb et < 500 kb	2 205,90 €	Selon indications fixées par l'INCa et la DGOS
N455	Forfait mutationnel syndromes myéloprolifératifs	124,20 €	Autres mutations à impact diagnostique et/ou théranostique des syndromes myéloprolifératifs (forfait 2 à 5): CALR exon 9, MPL W515 , JAK2 exon 12, CSFR3 exons 14 à 17, SETBP1 exon 4. Par cible

Prise en charge des NGS

GOUVERNEMENT
Liberté
Égalité
Fraternité

Communications scientifiques


Impact diagnostique, pronostique et théranostique d'un panel de gènes dédié aux hémopathies myéloïdes par technique NGS : L'expérience du laboratoire Eurofins Biomnis en 2019
eurofins | Biomnis
Objectif
Patients
Méthode
WET LAB
DRY LAB
LMC ATYPIQUE
LCN
MARS-T
LMMC
SMD
MPN MFS
VALEUR DIAGNOSTIQUE
VALEUR PRONOSTIQUE
VALEUR THÉRANOSTIQUE
CHIP
DOUBLES MUTATIONS JAK2/CALR ET JAK2/MPL
SYNERGIE CARYOTYPE ET NGS
Conclusion
NGS
NOUVELLE ÈRE DIAGNOSTIQUE, PRONOSTIQUE, THÉRANOSTIQUE

Impact diagnostique, pronostique et théranostique d'un panel de gènes dédié aux hémopathies myéloïdes par technique NGS : L'expérience du laboratoire Eurofins Biomnis en 2019

V. Geromel (1) ; P. Mouty (1) ; C. Chal (1) ; J. Delaunay (2) ; S. Sadot-Leboucier (2) ; R. Kaphan (3) ; B. Rossignol (4) ; K. LeDû (5) ; J. Vigneri (6) ; JP. Coadic (7) ; EA. MerinLaMaalee (7) ; R. Boutault (8) ; O. Rouaides (1) ; C. Bourdin (1) ; A. Petit (1) ; M. Roumiguières (1) ; L. Raymond (1) ; B. Quilichini (1)

(1) Département hématologie cellulaire/cytogénétique/génétique, Eurofins | Biomnis, Lyon; (2) Service d'hématologie clinique, L'Hôpital Privé du Confluent, Nantes; (3) Médecine interne-oncologie, CH Cannes, Cannes; (4) Hématologie clinique, C.H. de la Drôménie, Dragulignan; (5) Hématologie, Centre Jean Bernard Le Mans, Le Mans; (6) Médecine interne, CH- Sénur en Auxois, Senur-en-Auxois; (7) Labm, Laboratoire Biomnis, Nantes; (8) Labm, Laboratoire Labomnis, Le Mans

Objectif

Les données d'hématologie cellulaire, histologiques, de cytogénétique et de biologie moléculaires sont indispensables au clinicien pour poser un diagnostic d'hémopathie maligne et évaluer son pronostic. L'étroite collaboration entre ces disciplines est un élément clé du diagnostic.
La technique NGS contribue à améliorer la prise en charge diagnostique, pronostique et théranostique des hémopathies malignes.

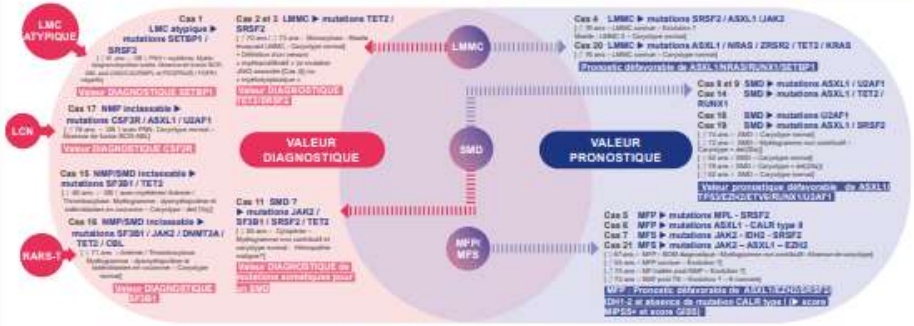
Nous proposons au laboratoire des panels de gènes NGS ciblés pour les hémopathies malignes: Néoplasies myéloprolifératives (PV/MPF/TE/LCN), Néoplasies myélodysplasiques/myéloprolifératives (LMMC, LMC atypique BCR-ABL négatif, RARS-T), Syndromes myélodysplasiques (SMD) et Leucémie aiguë myéloïde (LAM).
Ces panels ont été élaborés à partir des données de la nouvelle classification OMS 2017 des tumeurs des tissus hématopoïétiques et lymphoïdes.

Patients

Depuis 2019, plus de 500 analyses NGS ont été effectuées en routine dans notre laboratoire. Pour chaque cas de suspicion d'hémopathie maligne, nous disposons des données cliniques, cytologiques et cytogénétiques.
Pour cette étude, nous avons sélectionné plus de 40 cas.

Méthode

Contrairement aux approche séquentielles traditionnelles, le séquençage haut débit (NGS) est une méthode de diagnostic moléculaire qui permet d'étudier plusieurs gènes en une seule technique.
Les panels NGS ont été réalisés avec un kit commercial permettant l'analyse de 30 gènes (Myeloid solution™ par SOPHIA GENETICS).



L'interprétation des résultats issus du séquençage haut débit (NGS) dans les hémopathies malignes reste délicate :

- Nombre élevé de variants rapportés dans la littérature
- Identification de nouveaux variants non décrits
- Valeur clinique du pourcentage de la VAF d'une mutation donnée ?
- Origine constitutionnelle versus origine somatique ?
- Bases de données ?

Une approche interdisciplinaire impliquant cliniciens et biologistes moléculaires est la clé pour une bonne interprétation du résultat NGS.

Conclusion

En matière de prise en charge diagnostique dans les hémopathies myéloïdes chroniques, la dernière classification de l'OMS n'inclut que les mutations des gènes JAK2, CALR, MPL, CSF3R et SF3B1 en critères diagnostiques stricts.

Cependant, la classification actuelle de l'OMS pour ces hémopathies malignes sera certainement redéfinie par une approche moléculaire plus exhaustive basée sur les récentes contributions des résultats de la technique NGS.

NGS → NOUVELLE ÈRE DIAGNOSTIQUE, PRONOSTIQUE, THÉRANOSTIQUE

Références :
Swerdlow SH, Campo E, Harris NL, Jaffe ES, Pileri SA, Stein H, et al. WHO classification of tumours of haematopoietic and lymphoid tissues. 4th ed. Lyon: IARC Press, 2017 - Grinfeld, et al. NEJM 2018; 379:1416-30 - Tefferi et al. BJH 2020; 189(2): 291-302 - Tefferi et al. AJH 2021; 96: 145-162

Poster 0000 - 42ème Congrès de la SFH du 08 au 11 Septembre 2020, Palais des congrès de Paris

Etude d'une cohorte de cas double mutés JAK2/CALR et JAK2/MPL dans des NMP non LMC et revue de la littérature.

L. Bertannier(1), V. Geromel(1), A. Petit(1), C. Bourdin(1), V. Goutsloy(1), P. Mouty(1), B. Rossignol(2), E. Berthoux(3), C. Quinquenel(4), K. Grissa(5), C. Kennel(6), M. Roumiguière(7), L. Raymond(1), B. Quilichini(1)

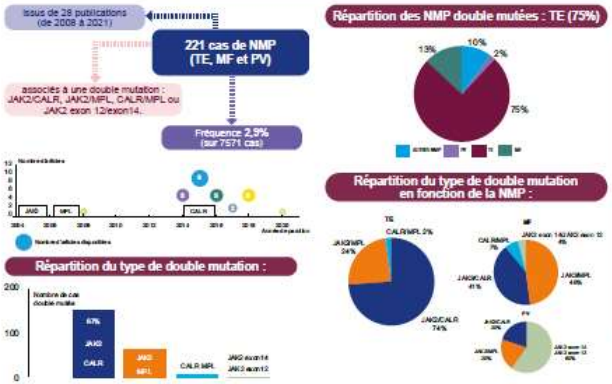
(1) Départements d'Hématologie cellulaire, de Cytogénétique et de Génétique, Eurofins Biomnis, Lyon - (2) Service d'Hématologie clinique, C.H. de la Drôménie, Dragulignan - (3) Service de médecine interne, C.H. Saint Joseph-Saint Luc, Lyon - (4) Service de médecine interne, C.H. d'Ardèche Nord, Annonay - (5) Service d'Hématologie clinique, C.H. William Morey, Chalon sur Saône

Introduction

L'analyse des mutations JAK2, CALR et MPL est la base moléculaire pour poser le diagnostic d'une NMP non LMC (OMS 2017). L'analyse séquentielle réalisée historiquement dans les laboratoires ne permet pas d'explorer la présence de cas double mutés JAK2/CALR, JAK2/MPL, CALR/MPL, JAK2 exon 12 – exon 14.

→ Le laboratoire Eurofins Biomnis propose depuis 2019 une approche globale par technique de séquençage haut débit (NGS) avec un panel de gènes à visée diagnostique dédié pour la détection simultanée des mutations JAK2, CALR et MPL.

Revue de la littérature



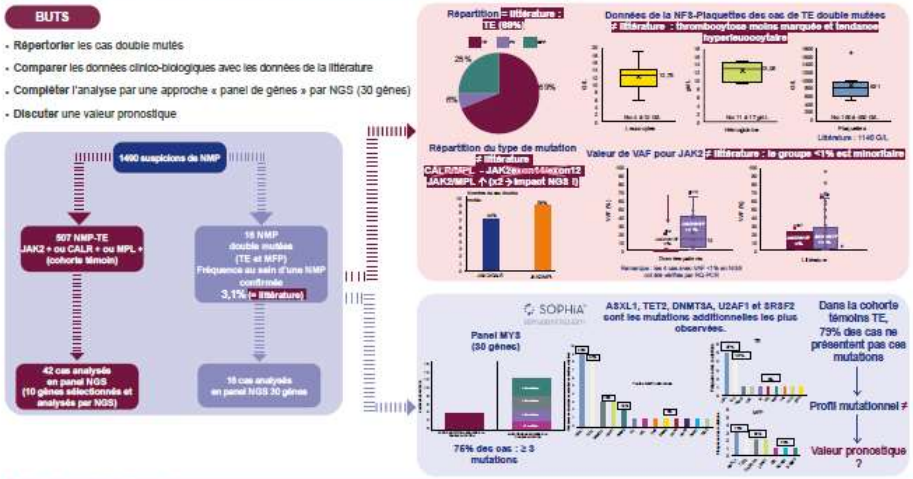
BIAIS D'ANALYSE OBSERVÉS DANS LA LITTÉRATURE :

- Hétérogénéité d'échantillonnage des cohortes étudiées
- Hétérogénéité des techniques utilisées pour la détection des mutations (RQ-PCR / HRM / Sanger / NGS ...) = seuil de détection différent
- Caractérisation parfois partielle du variant observé (hot spot ? / variant pathogène - probablement pathogène ? / variant non rapporté ? / variant d'origine constitutionnelle ?)
- Signification clinico-biologique d'une VAF < 1% (en particulier pour JAK2 : 47% des cas double mutés présentent une VAF JAK2 V617F <1%)
- Absence de données de suivi clinique pour évaluer l'impact pronostique
- Seulement 3 publications (2018) avec une analyse complémentaire d'un panel de gènes par NGS – absence de caractérisation moléculaire des cas double mutés

Etude au laboratoire Eurofins Biomnis

BUTS

- **Répertorier** les cas double mutés
- **Comparer** les données clinico-biologiques avec les données de la littérature
- **Compléter** l'analyse par une approche « panel de gènes » par NGS (30 gènes)
- **Discuter** une valeur pronostique



Conclusion / Perspectives

La notion de « NMP double mutée » est une réalité au sein des NMP. Il reste à en définir plus précisément sa valeur pronostique sur des séries plus importantes en proposant, par exemple, une étude moléculaire élargie en tenant compte des nouveaux marqueurs pronostiques dans la TE (SF3B1, SRSF2, U2AF1 et TP53).

Références :
Swerdlow SH, Campo E, Harris NL, Jaffe ES, Pileri SA, Stein H, et al. WHO classification of tumours of haematopoietic and lymphoid tissues. 4th ed. Lyon: IARC Press, 2017 - Grinfeld, et al. NEJM 2018; 379:1416-30 - Tefferi et al. BJH 2020; 189(2): 291-302 - Tefferi et al. AJH 2021; 96: 145-162

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Etude alternant en LP

Analyse par NGS de données moléculaires à visée pronostique dans la TE et la PV

Étude d'une cohorte de 89 patients

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