Update of WHO and Molecular Classifications in Myelodysplastic syndromes

Mario Cazzola, MD

Department of Hematology Oncology, Fondazione IRCCS Policlinico San Matteo, and Department of Molecular Medicine, University of Pavia, Pavia, Italy

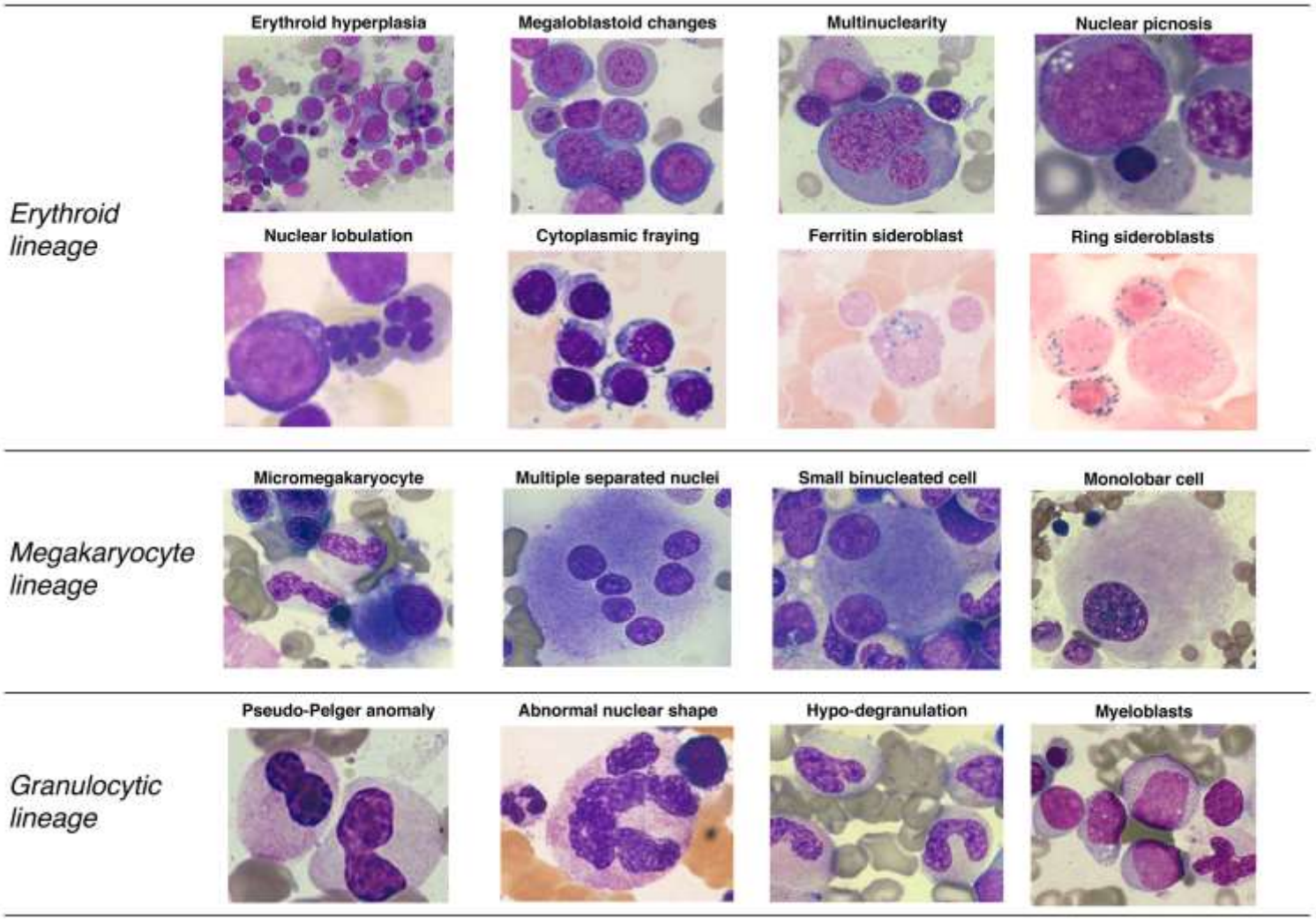
MDS Foundation Symposium

San Francisco, CA, December 5th, 2014

Mario Cazzola Disclosures

- PI in sponsored clinical trials: no personal financial relationship with pharmaceutical companies
- Research grants from non-profit organizations or governmental agencies exclusively
- Associate Editor for Blood

Pivotal role of morphology in diagnosis and prognostication of MDS



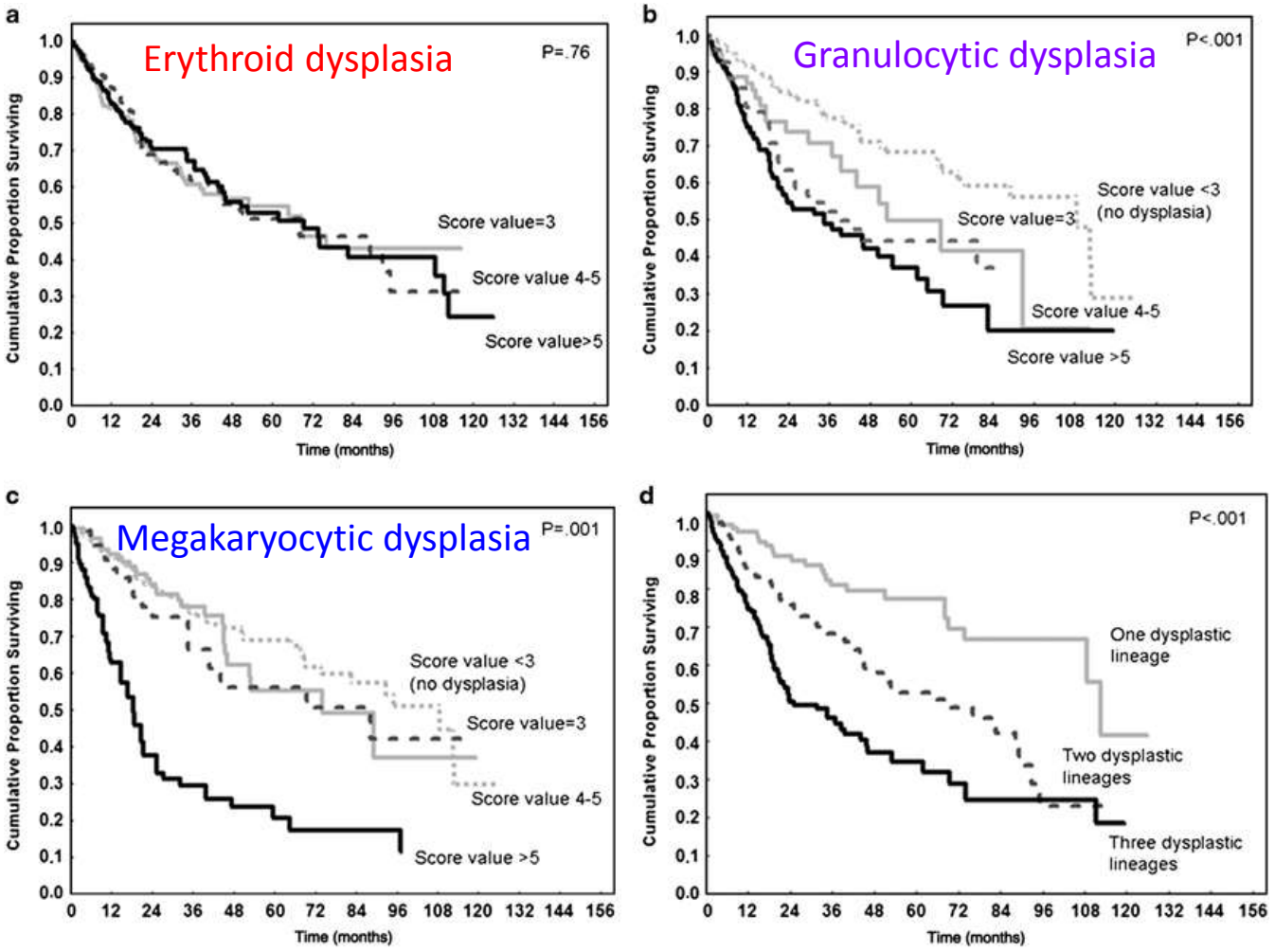
Cazzola et al. Blood. 2013 Dec 12;122(25):4021-34

WHO classification of MDS

- Refractory Cytopenia with Unilineage Dysplasia (RCUD) *(mainly refractory anemia)*
- Refractory Anemia with Ring Sideroblasts (RARS)
- Refractory Cytopenia with Multilineage Dysplasia (RCMD)
- Refractory Anemia with Excess Blasts (RAEB type I and II)
- Myelodysplastic Syndrome with Isolated del(5q)

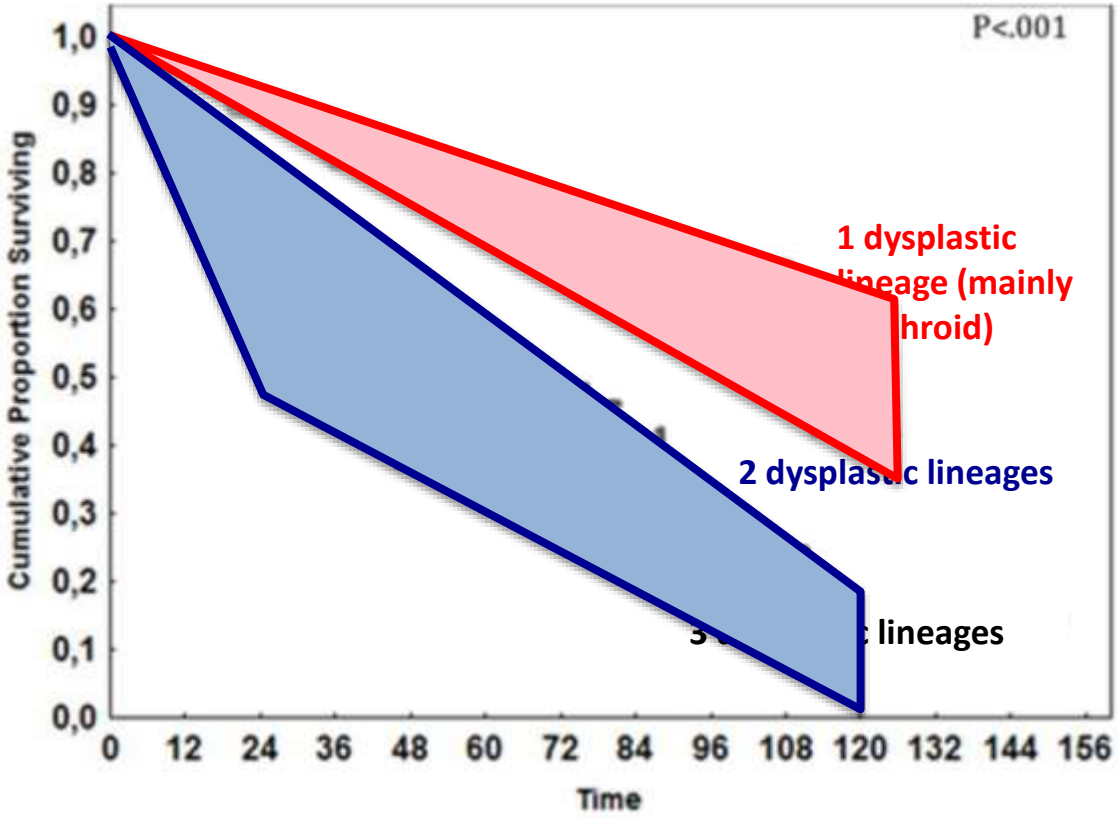
Swerdlow et al (Editors). IARC 2008

Overall survival according to erythroid, granulocytic and megakaryocytic morphological score value



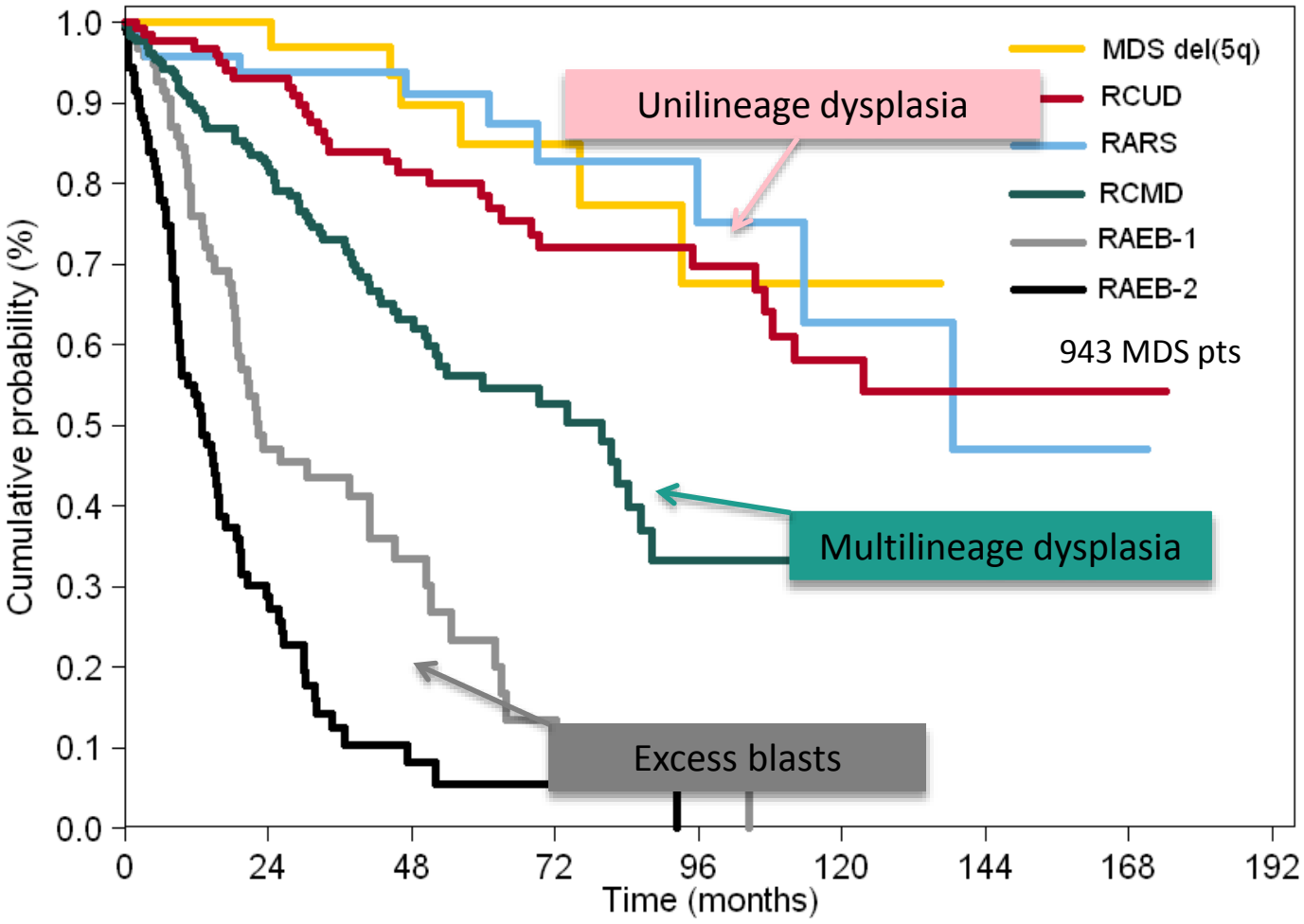
Della Porta et al. 2014 May 20. doi: 10.1038/leu.2014.161

Prognostic relevance of dysplasia: number of lineages involved



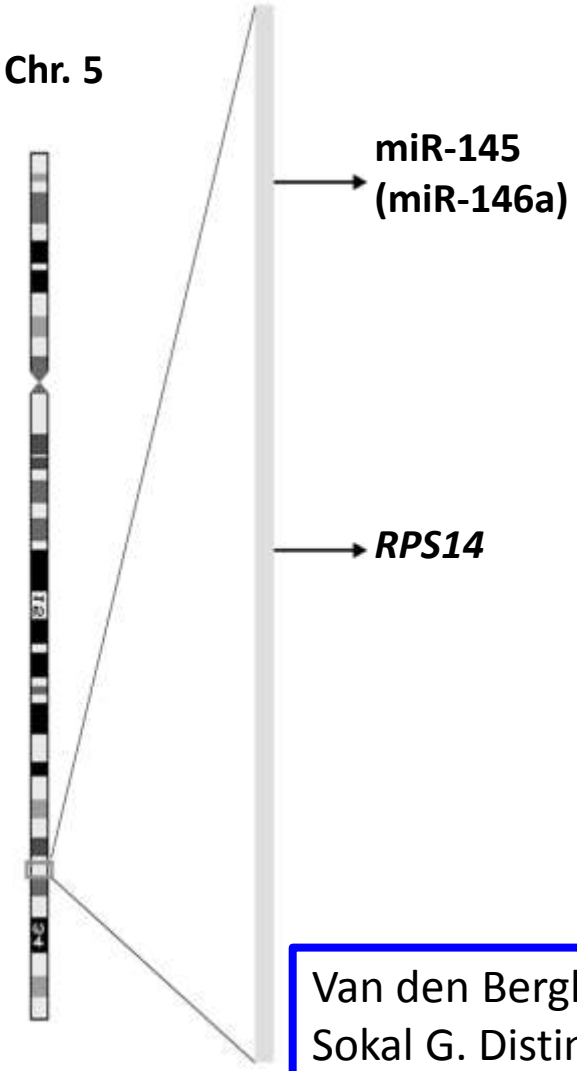
Della Porta et al. 2014 May 20. doi: 10.1038/leu.2014.161

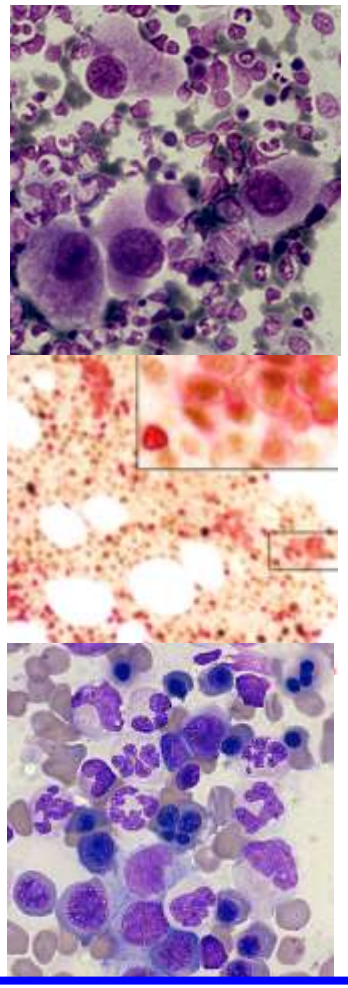
Outcome of MDS according to WHO classification



Cazzola. Haematologica. 2011 Mar;96(3):349-52.

MDS with isolate del(5q): distinct nosologic entity caused by haploinsufficiency of genes mapping on the deleted region





Loss of a micro RNA and thrombocytosis
Starczynowski et al. Nat Med. 2010 Jan;16(1):49-58.

Coordinate loss of a microRNA and protein-coding gene cooperate in the pathogenesis of 5q- syndrome
Kumar et al. Blood. 2011 Oct 27;118(17):4666-73

Activation of p53 and apoptosis of immature red cells
Barlow et al. Nat Med. 2010 Jan;16(1):59-66
Pellagatti et al. Blood. 2010 Apr 1;115(13):2721-3
Dutt et al. Blood. 2011 Mar 3;117(9):2567-76

Haploinsufficiency of *RPS14* phenocopies the disease in normal hematopoietic progenitor cells
Ebert et al. Nature. 2008 Jan 17;451(7176):335-9

Van den Berghe H, Cassiman JJ, David G, Fryns JP, Michaux JL, Sokal G. Distinct haematological disorder with deletion of long arm of no. 5 chromosome. Nature. 1974 Oct 4;251(5474):437-8.

Lenalidomide induces ubiquitination and degradation of CSNK1A1 in MDS with del(5q)

- Lenalidomide induces the ubiquitination and consequent degradation of CSNK1A1

- del(5q) cells have only one copy of *CSNK1A1*, so they are selectively depleted over wild-type cells

Fink et al. ASH 2014, abstract #4

MDS prognostic scoring systems

- **WPSS**
  - WHO classification (ring sideroblasts, multilineage dysplasia, excess blasts)
  - IPSS cytogenetics
  - severity of anemia (transfusion requirement)

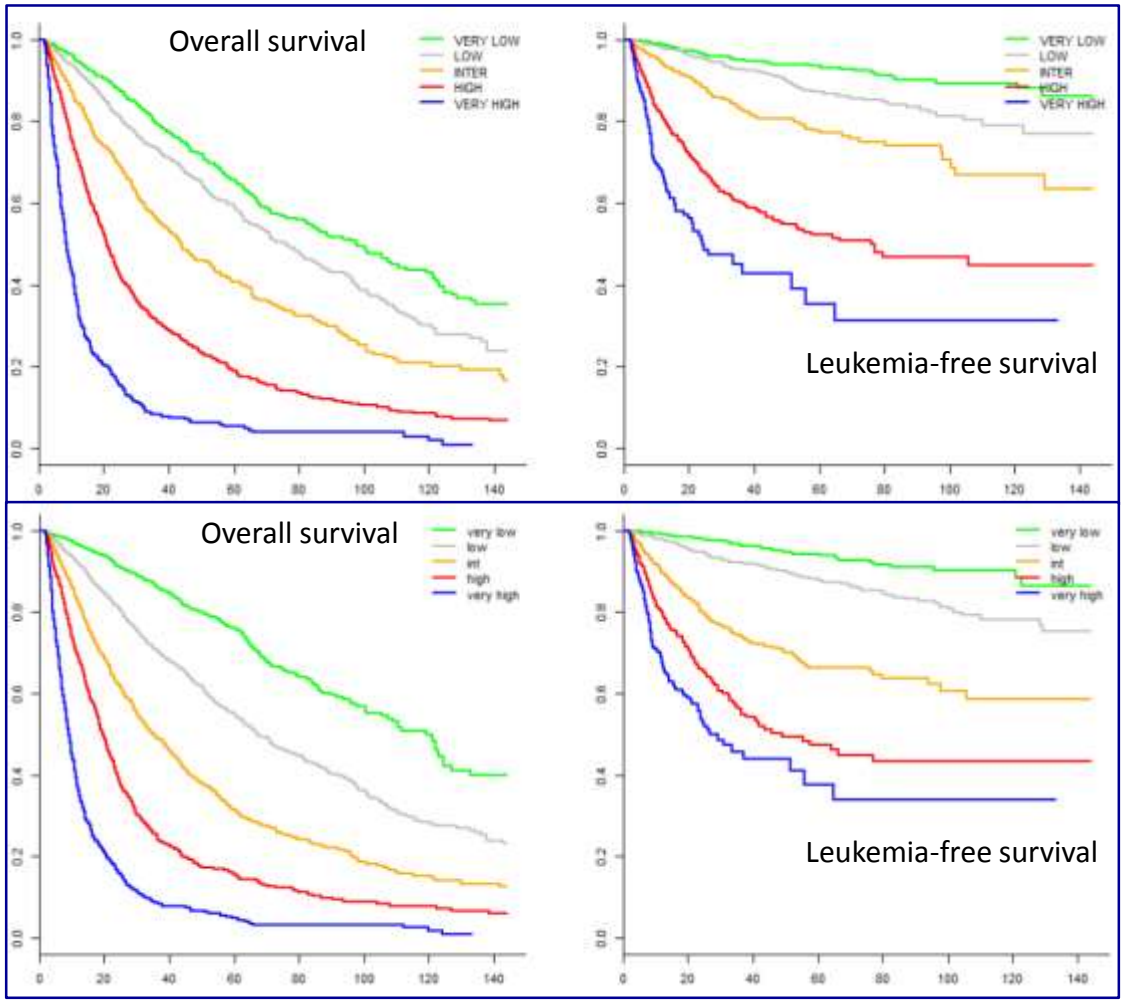
- **IPSS-R**
  - degree of cytopenia (Hb, ANC, PLT)
  - excess blasts (≤2%, 3-4%, 5-10%, >10%)
  - revised cytogenetics

A study of the International Working Group for Prognosis in Myelodysplasia (IWG-PM) on 5326 untreated MDS patients

WPSS

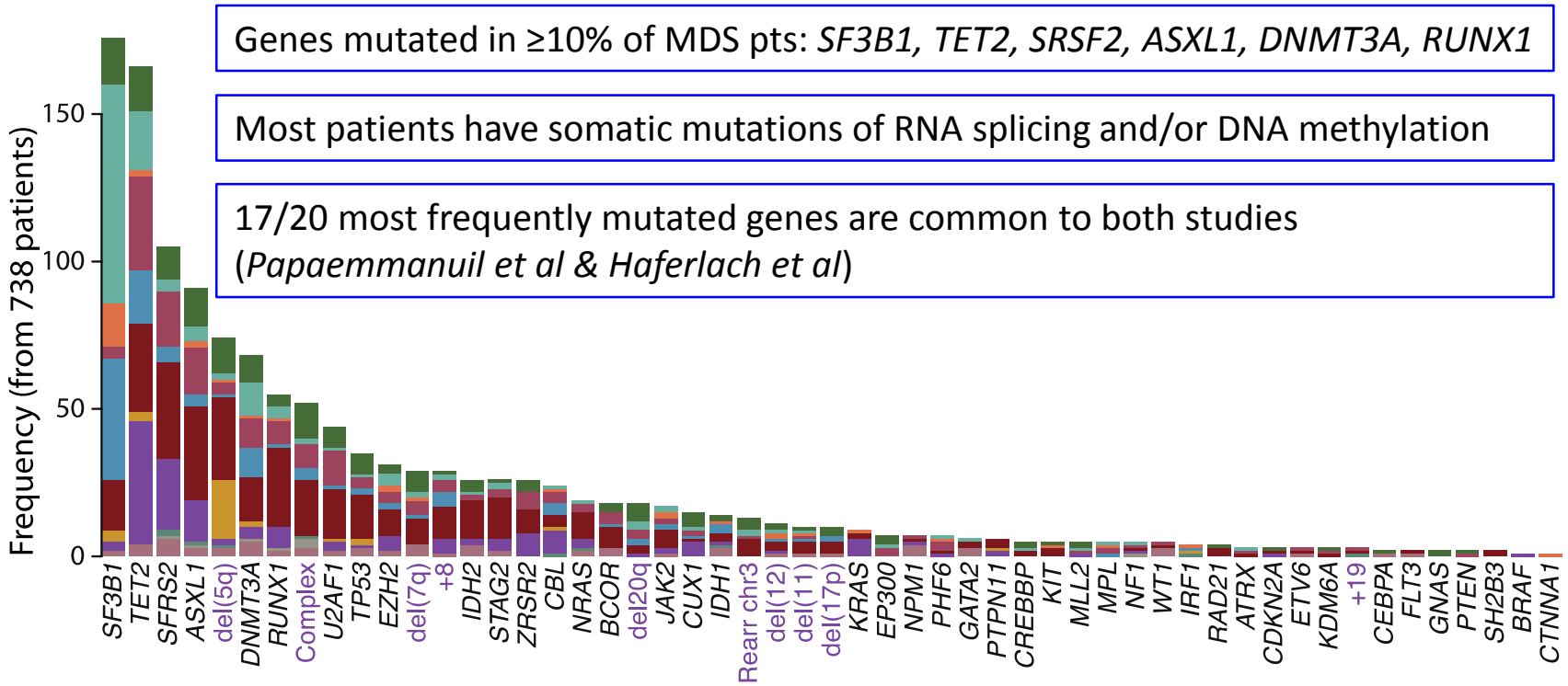
Kendall's tau 0.72 (*P*<.001)

IPSS-R



Della Porta et al. 2014, unpublished results

Somatic gene mutations in patients with MDS



Genes mutated in ≥10% of MDS pts: *SF3B1, TET2, SRSF2, ASXL1, DNMT3A, RUNX1*

Most patients have somatic mutations of RNA splicing and/or DNA methylation

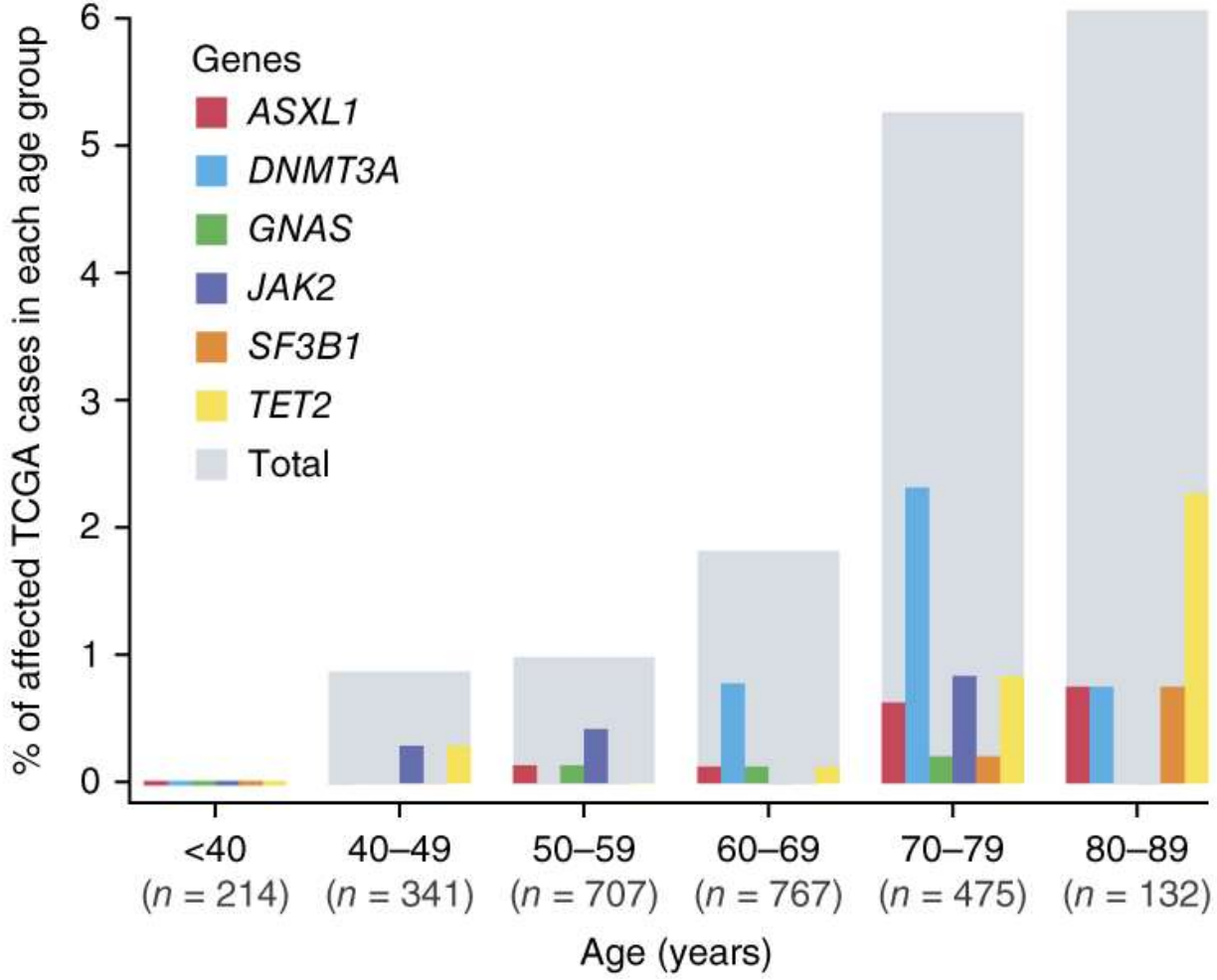
17/20 most frequently mutated genes are common to both studies (*Papaemmanuil et al & Haferlach et al*)

Papaemmanuil et al. Blood. 2013 Nov 21;122(22):3616-27

Haferlach et al. Leukemia. 2014 Feb;28(2):241-7

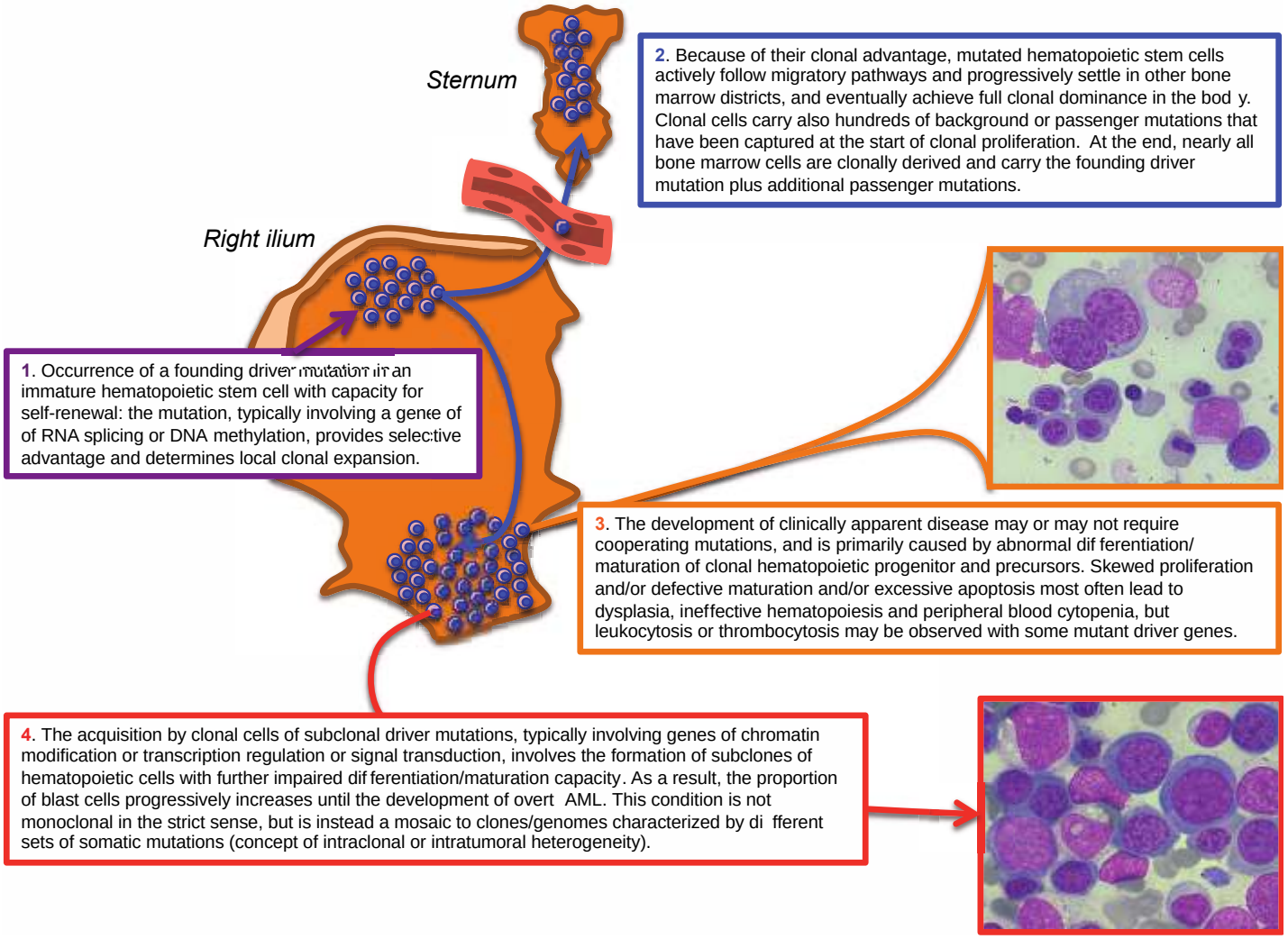
The blood cells of individuals with solid tumors contain mutations that may represent premalignant events that cause clonal hematopoietic expansion

The Cancer Genome Atlas (TCGA)



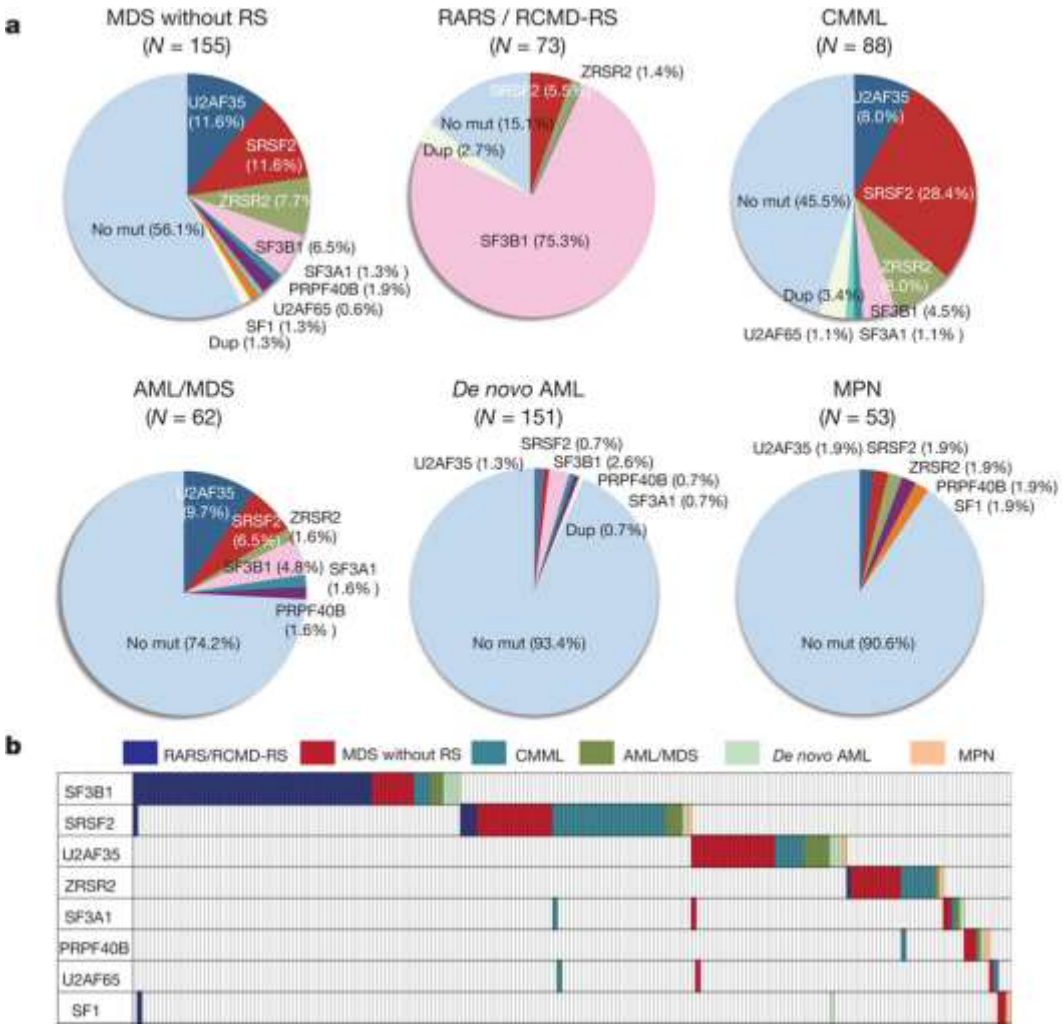
Xie et al. Nat Med. 2014 Oct 19. doi: 10.1038/nm.3733

Genetic basis of myelodysplastic syndromes



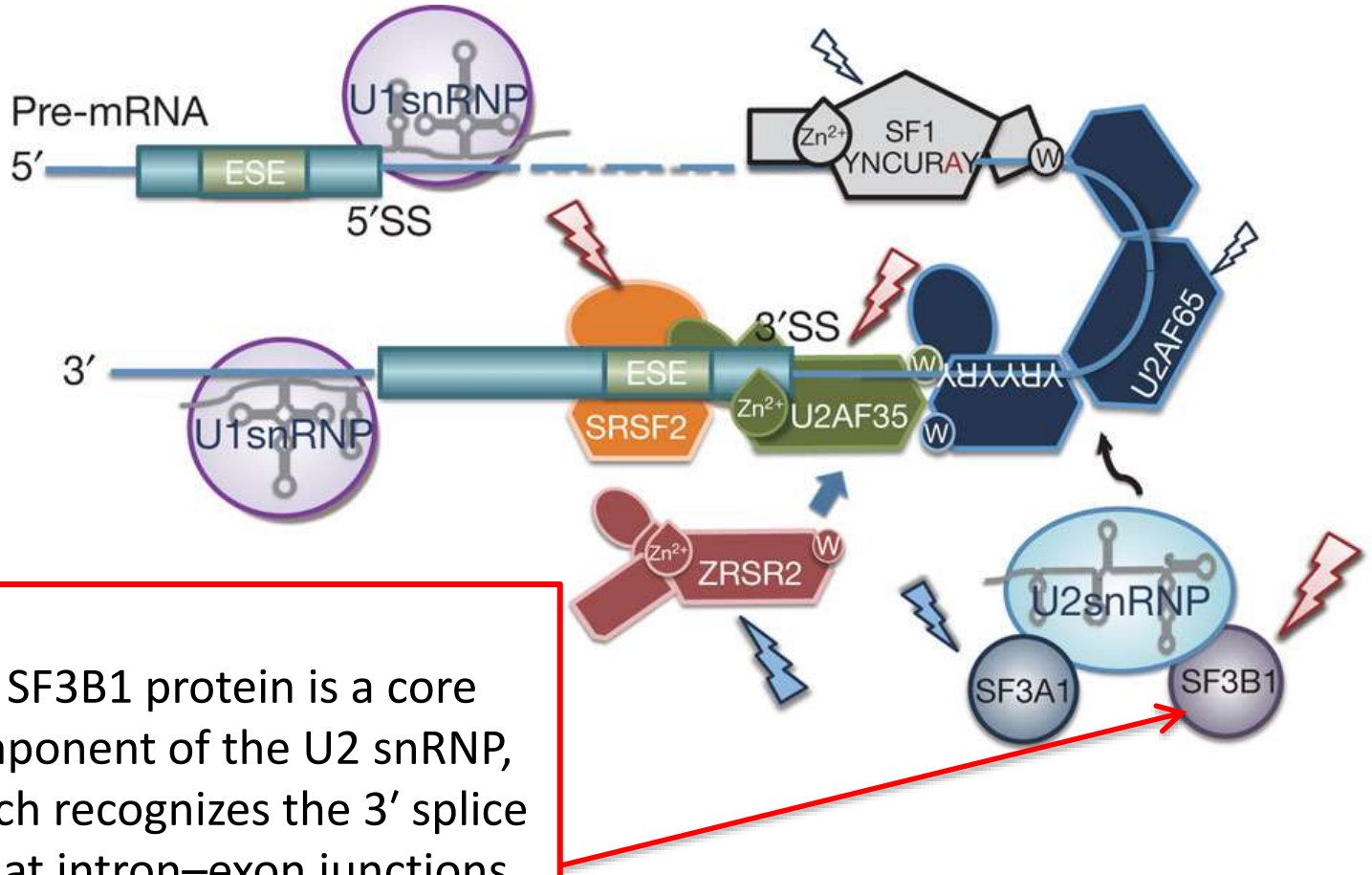
Cazzola et al. Blood. 2013 Dec 12;122(25):4021-34

Frequencies and distribution of spliceosome pathway gene mutations in myeloid neoplasms



Yoshida et al. Nature. 2011 Sep 11;478(7367):64-9

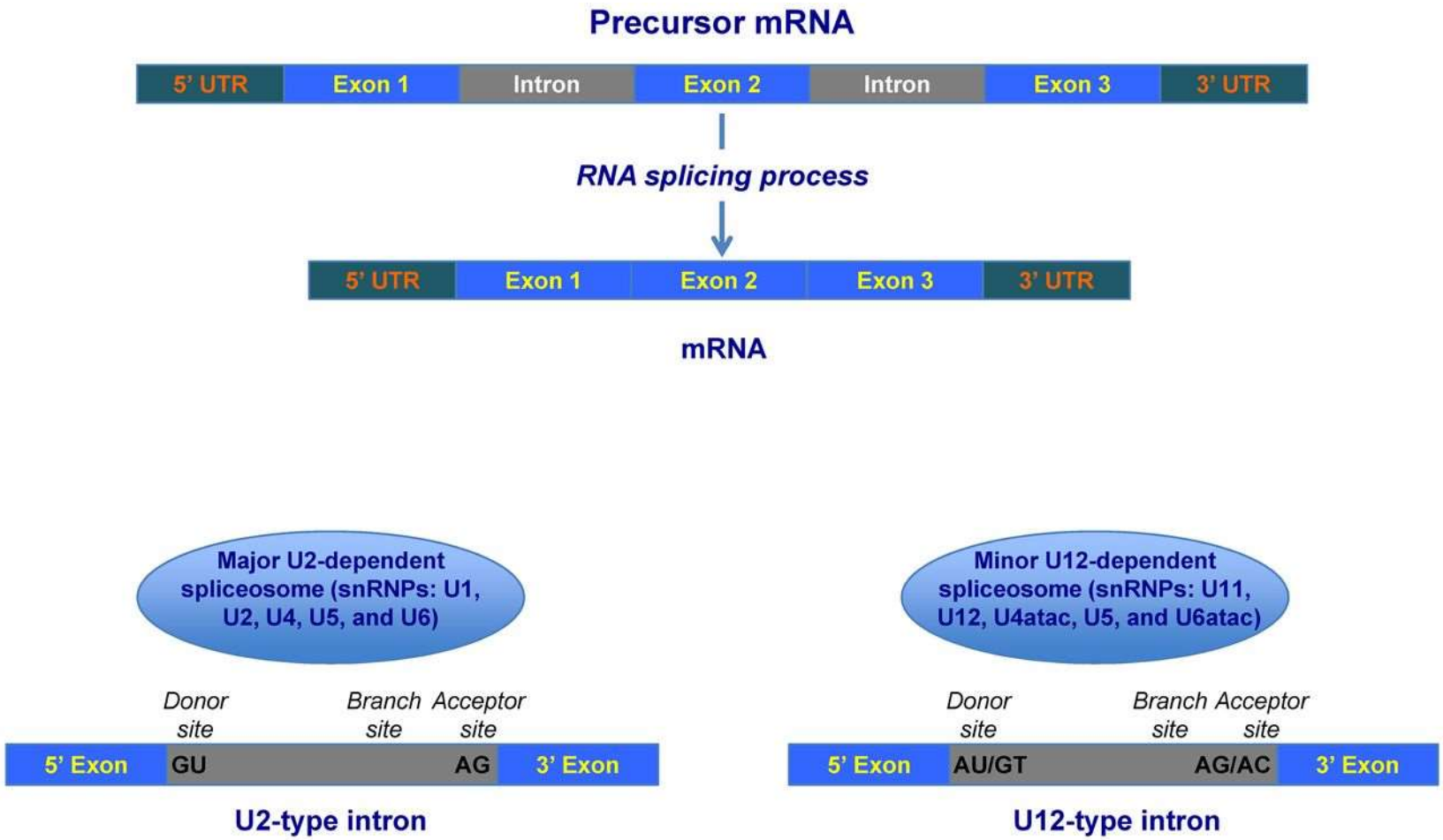
Splicing factors



The SF3B1 protein is a core component of the U2 snRNP, which recognizes the 3′ splice site at intron–exon junctions

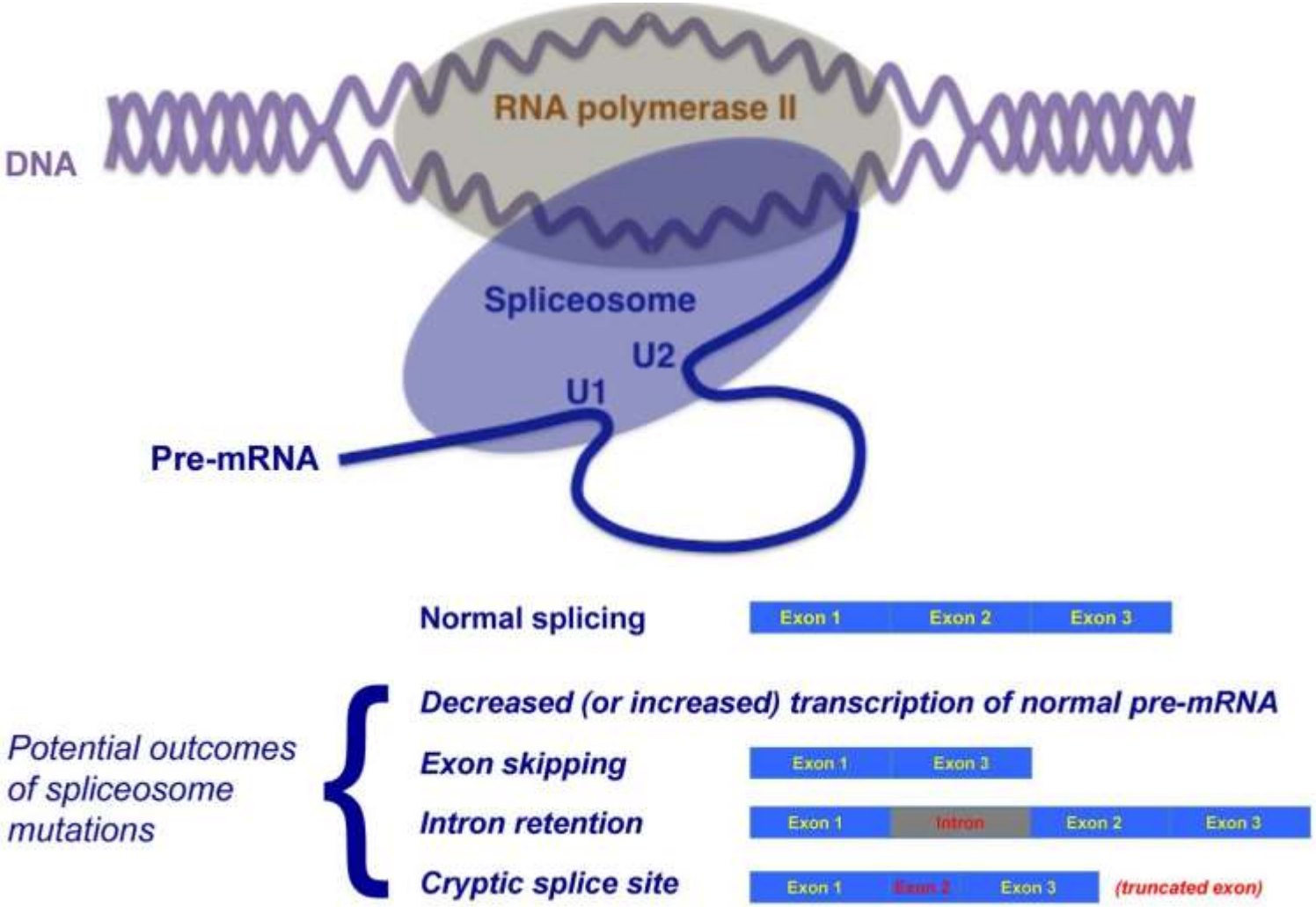
Yoshida et al. Nature. 2011 Sep 11;478(7367):64-9

Precursor mRNA (pre-mRNA) splicing



Cazzola et al. Blood. 2013 Jan 10;121(2):260-9

Co-transcriptional RNA splicing and potential outcomes of mutations of genes encoding proteins of the spliceosome



Cazzola et al. Blood. 2013 Jan 10;121(2):260-9

Relationship between somatic *SF3B1* mutations and ring sideroblasts

Quantitative enumeration of ring sideroblasts: 325 MDS patients

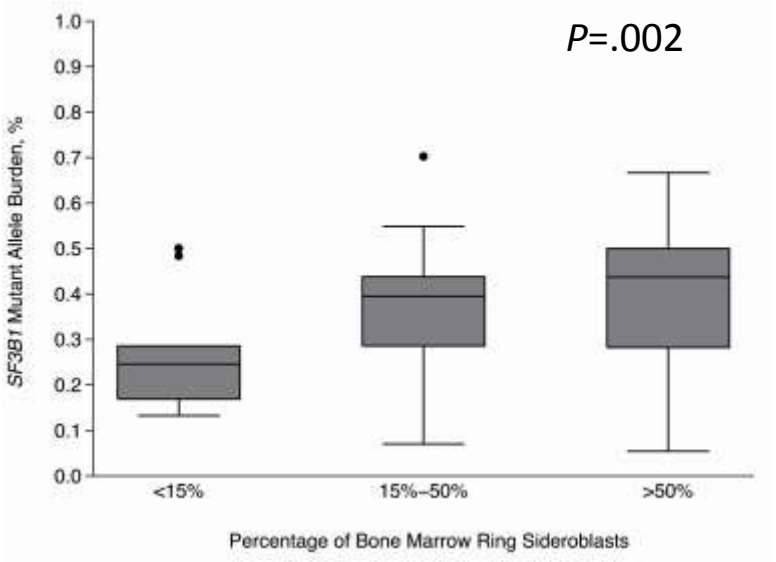
↓

101 (31%) patients with mutation in *SF3B1*

↓

91 patients >15% ring sideroblasts,
7 patients 1-14%,
3 patients no ring sideroblasts

↓

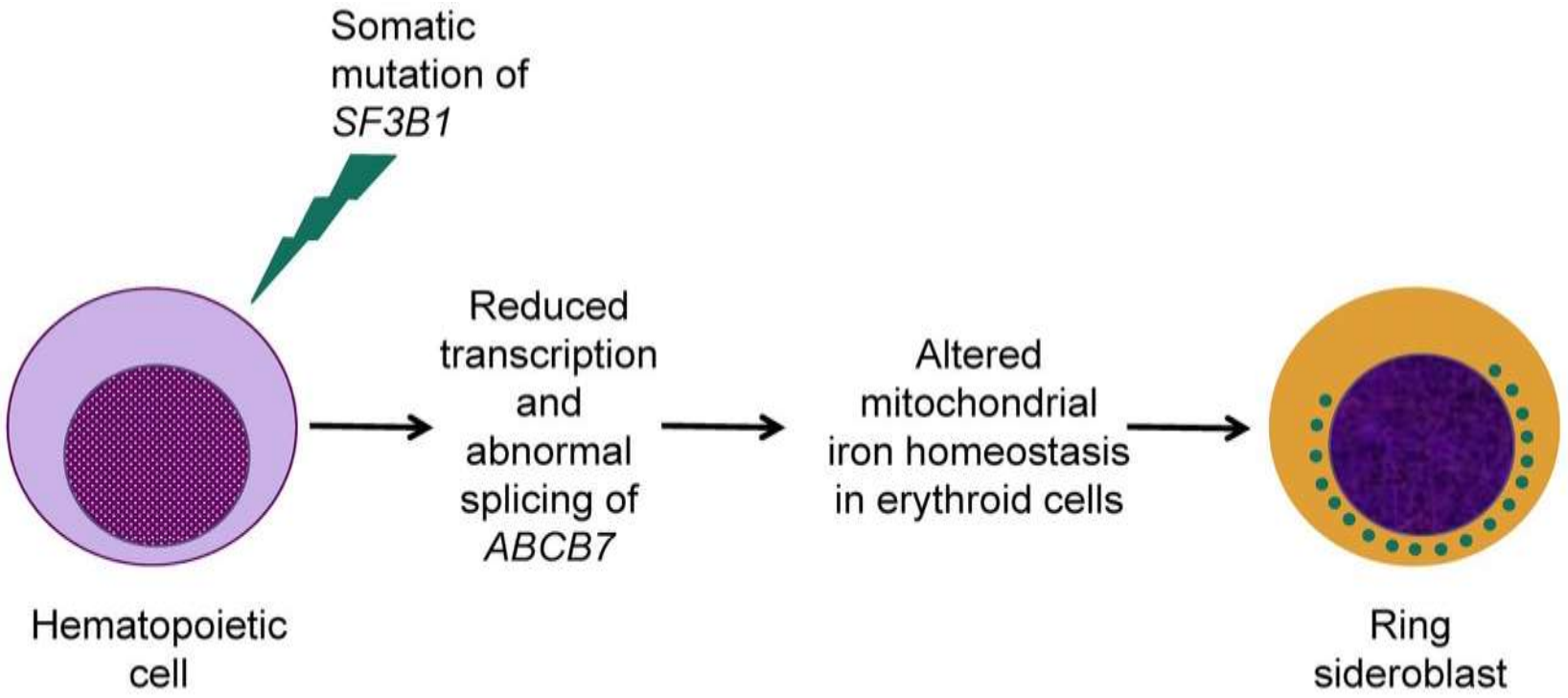


***SF3B1* mutation: positive predictive value for ring sideroblasts 97.7%**

Absence of ring sideroblasts: negative predictive value for *SF3B1* mutation 97.8%

Malcovati et al. Blood. 2011 Dec 8;118(24):6239-46

Relationship between the occurrence of a somatic *SF3B1* mutation and the formation of ring sideroblasts in patients with RARS



Cazzola et al. Blood. 2013 Jan 10;121(2):260-9

Comprehensive analysis of aberrant RNA splicing in myelodysplastic syndromes

- RNA sequencing of CD34+ cells revealed 230 splicing events significantly enriched in *SF3B1*-mutated cases, of which 206 (90%) were caused by misrecognition of 3' splice sites.

- About 50% of these altered 3' splice sites resulted in frameshift, indicating that *SF3B1* mutations cause deleterious effects in many genes simultaneously.

- Altered splice sites were found in genes involved in heme biosynthesis, cell cycle progression, and DNA repair

Shiozawa et al. ASH 2014, abstract #826

Novel disease paradigm

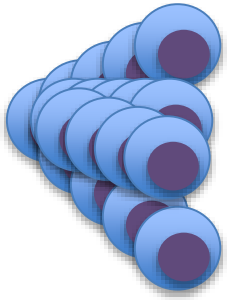
Occurrence of *SF3B1* mutation in a multipotent hematopoietic stem cell

Mutation detectable by DNA sequencing

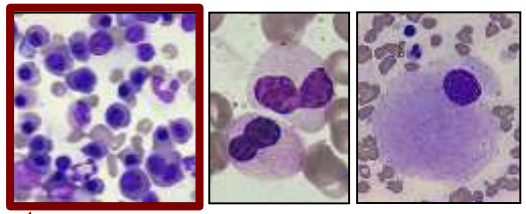
Misrecognition of 3' splice sites and frameshift in hundreds of genes

Mutations detectable only by RNA seq

Gain of function at hematopoietic stem cell level



Stem cells



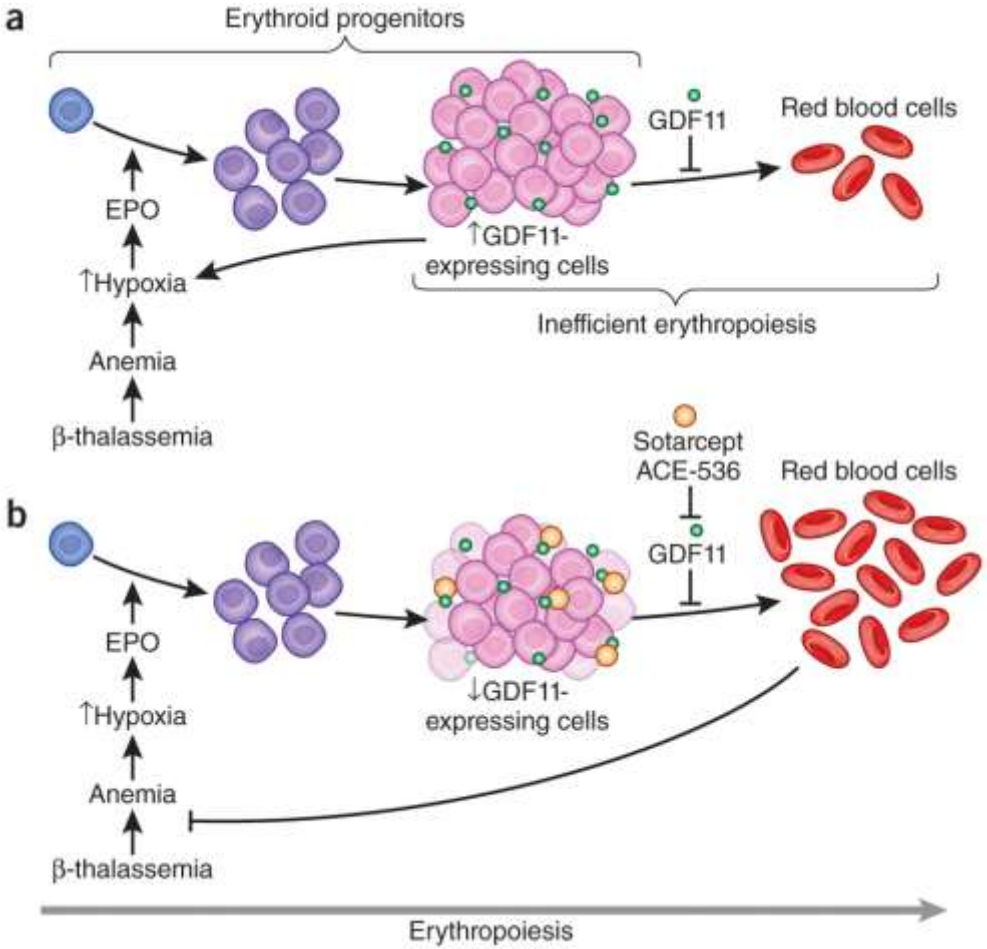
Hematopoietic precursors

Loss of function at hematopoietic precursor level

Ineffective erythropoiesis

Shiozawa et al. ASH 2014, abstract #826

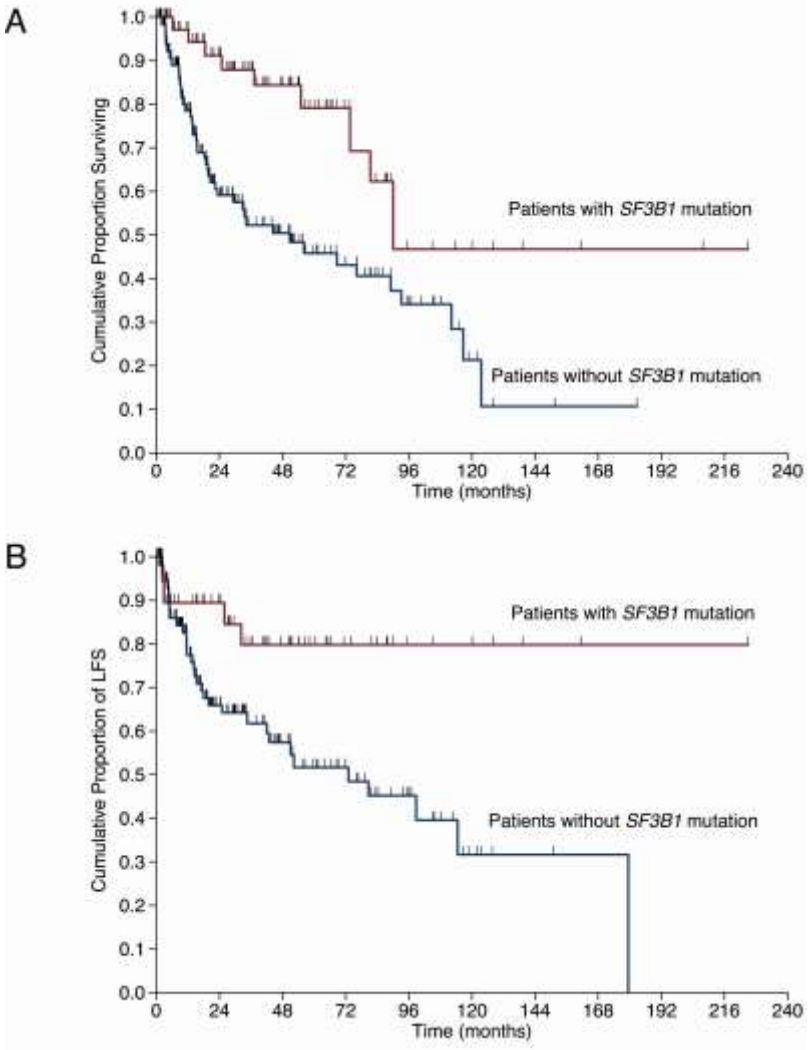
The ability TGF-β superfamily ligand-trapping proteins to alleviate anemia with ineffective erythropoiesis in a mouse model of MDS



Paulson RF. Nat Med. 2014 Apr;20(4):334-5

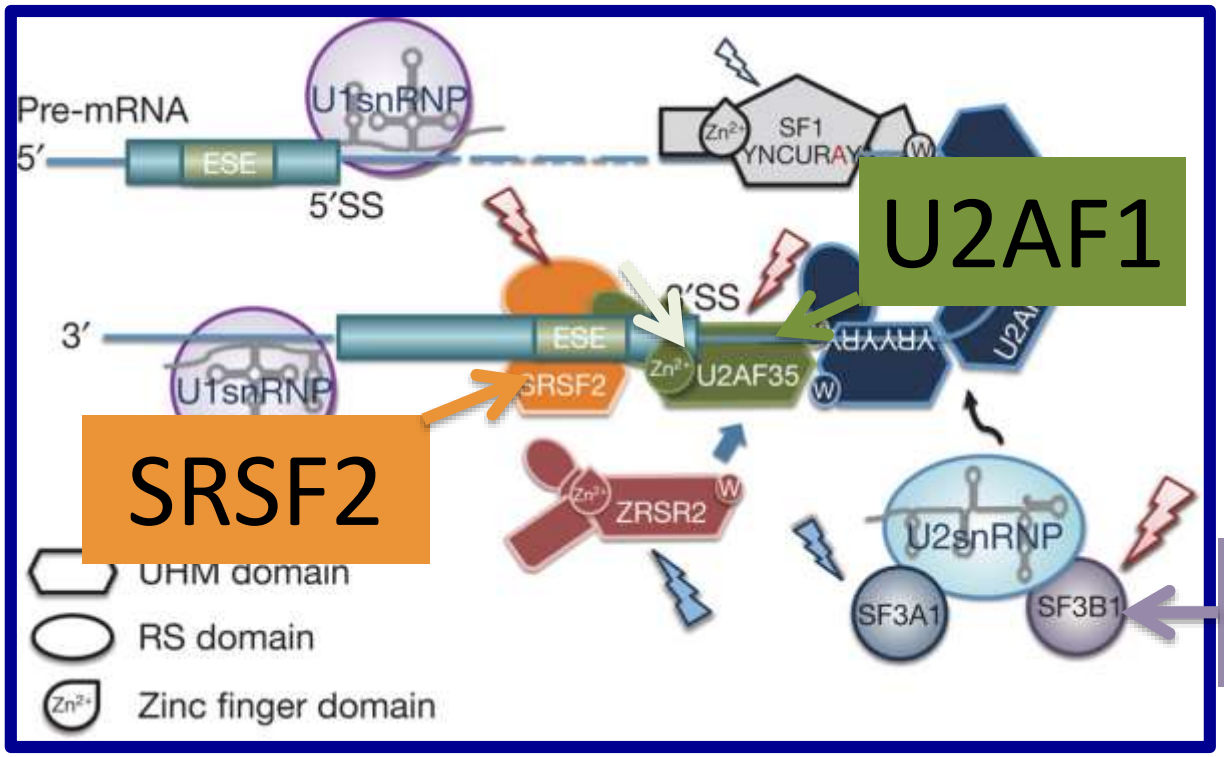
Suragani et al. Nat Med. 2014 Apr;20(4):408-14

Clinical significance *of SF3B1* mutation in MDS



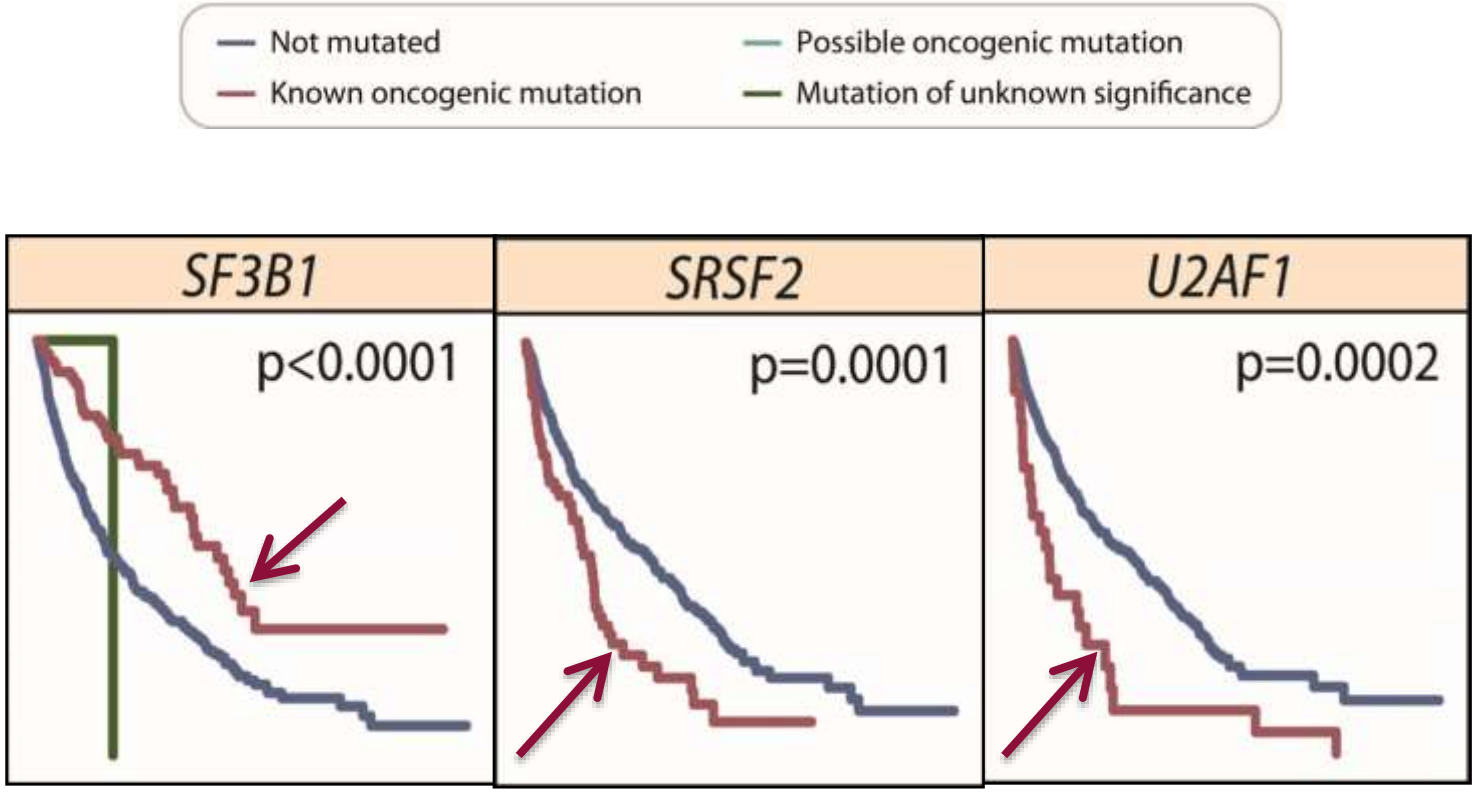
Malcovati et al. Blood. 2011 Dec 8;118(24):6239-46

RNA splicing factors: *SF3B1*, *SRSF2* and *U2AF1*



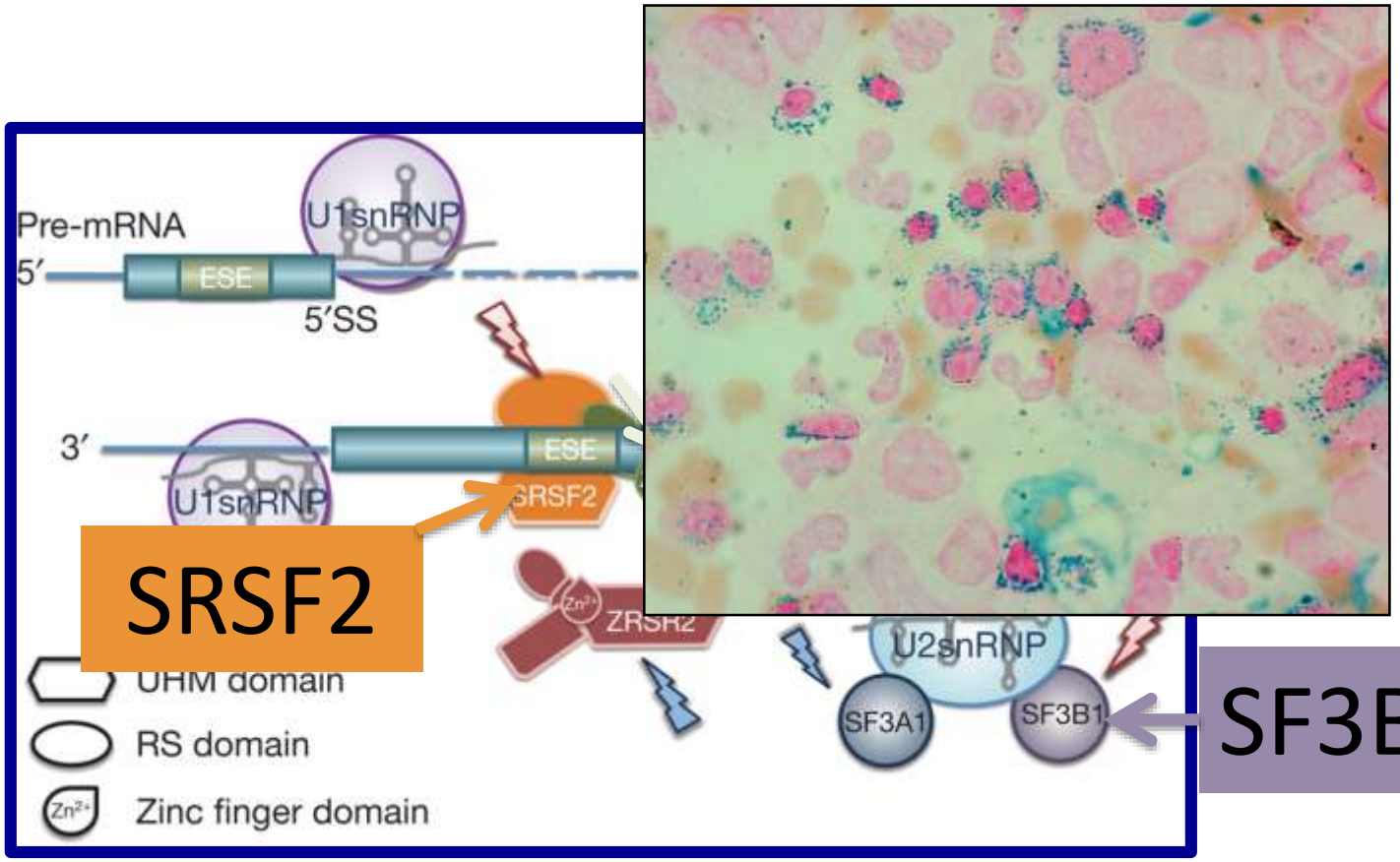
Yoshida et al. Nature. 2011 Sep 11;478(7367):64-9

Clinical effect of spliceosome pathway gene mutations in myelodysplastic syndromes



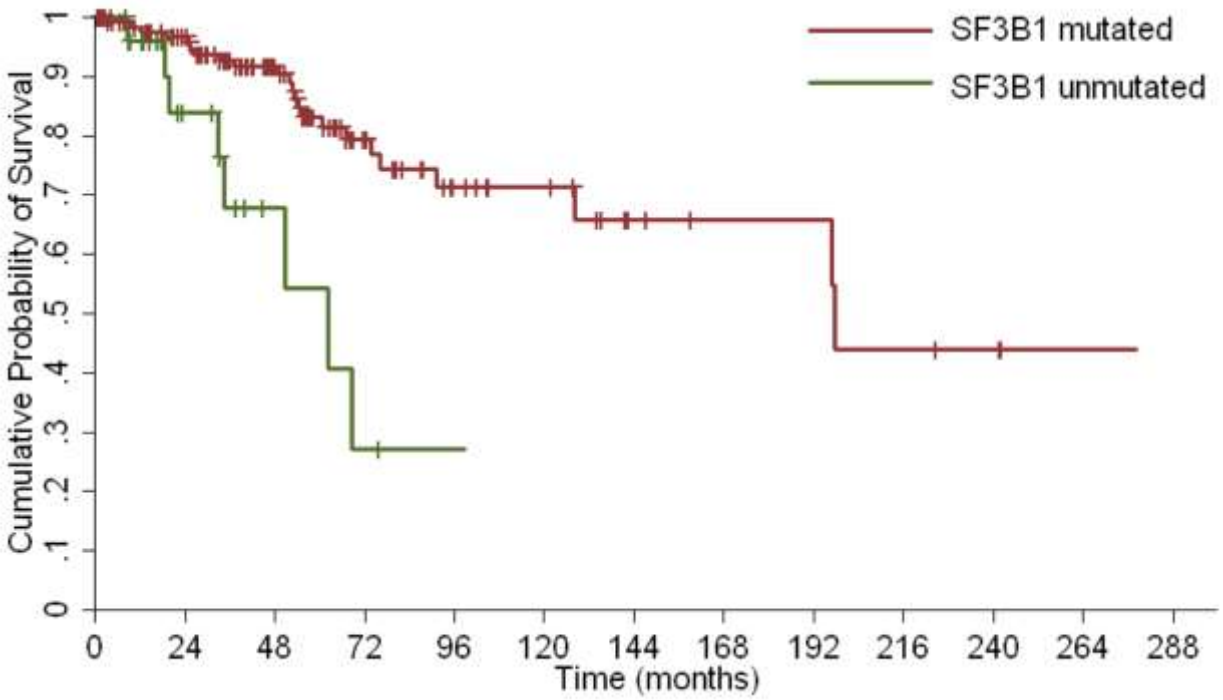
Papaemmanuil et al. Blood. 2013 Nov 21;122(22):3616-27

Clinical effect of spliceosome pathway gene mutations in MDS with ring sideroblasts



Yoshida et al. Nature. 2011 Sep 11;478(7367):64-9

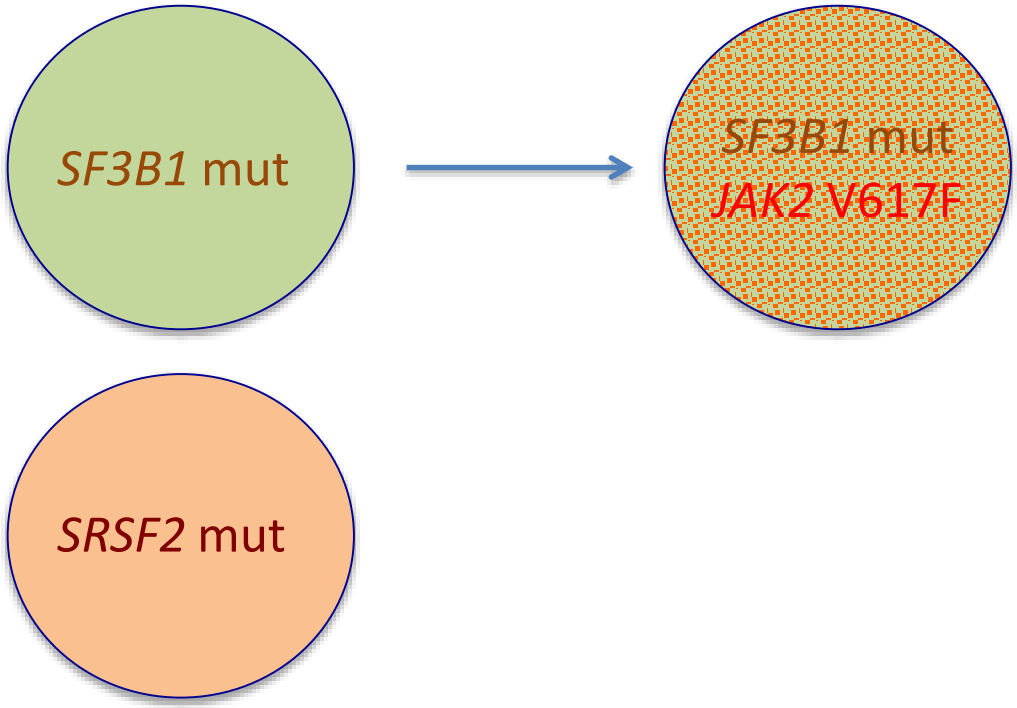
SF3B1-mutant MDS as a distinct nosologic entity



Malcovati et al. ASH 2014, abstract #826

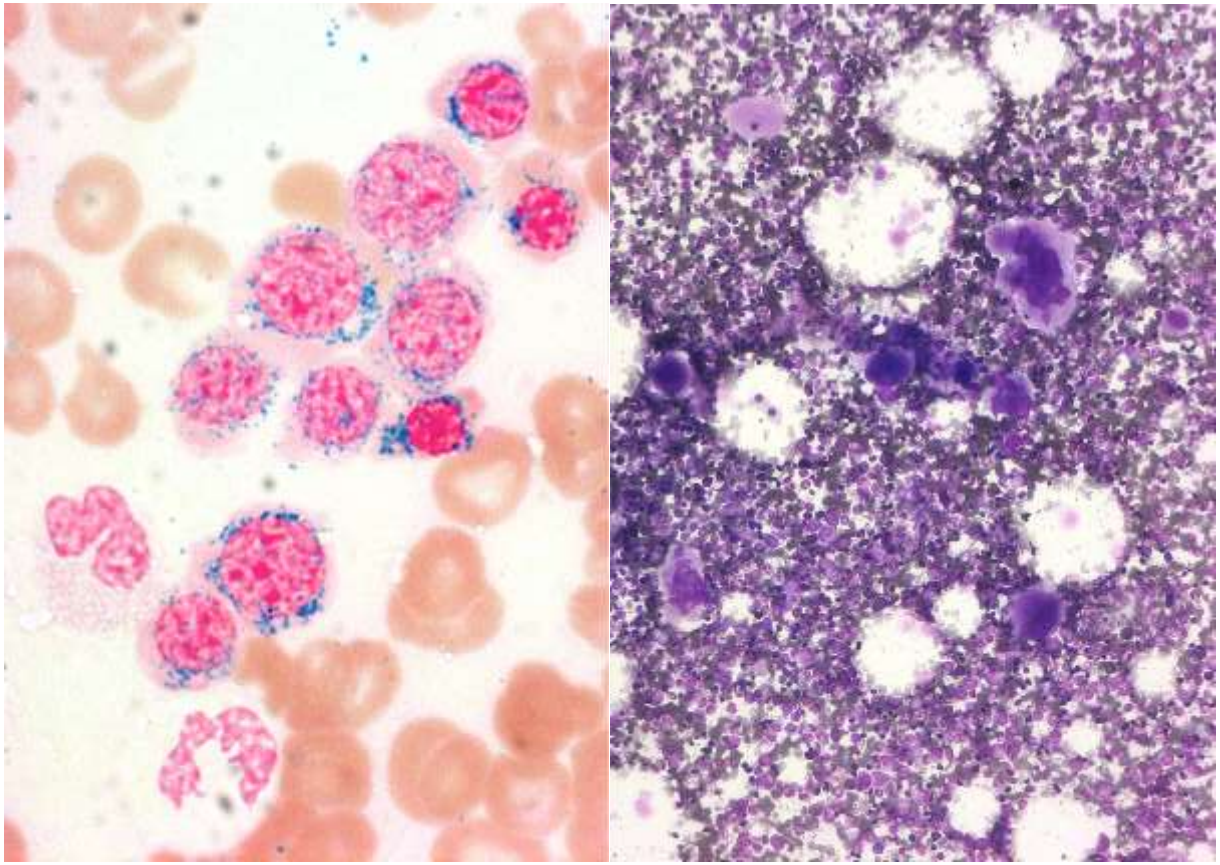
Genetic predestination

Genetic “predestination”: early founding driver mutations shape the future trajectories of clonal evolution of a cancer through constraints on the repertoire of cooperating subclonal genetic lesions



Papaemmanuil et al. Blood. 2013 Nov 21;122(22):3616-27

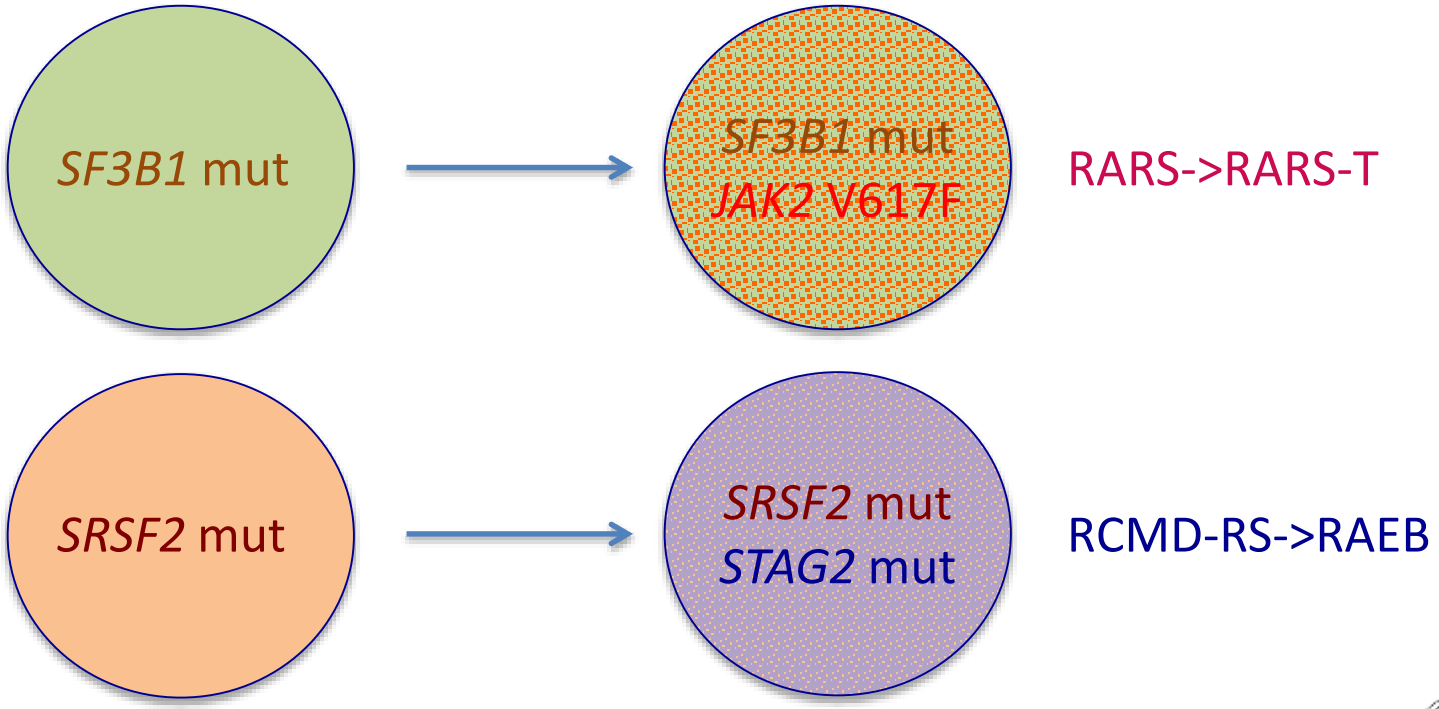
Transition from RARS to RARS-T



Malcovati et al. Blood. 2009 Oct 22;114(17):3538-45

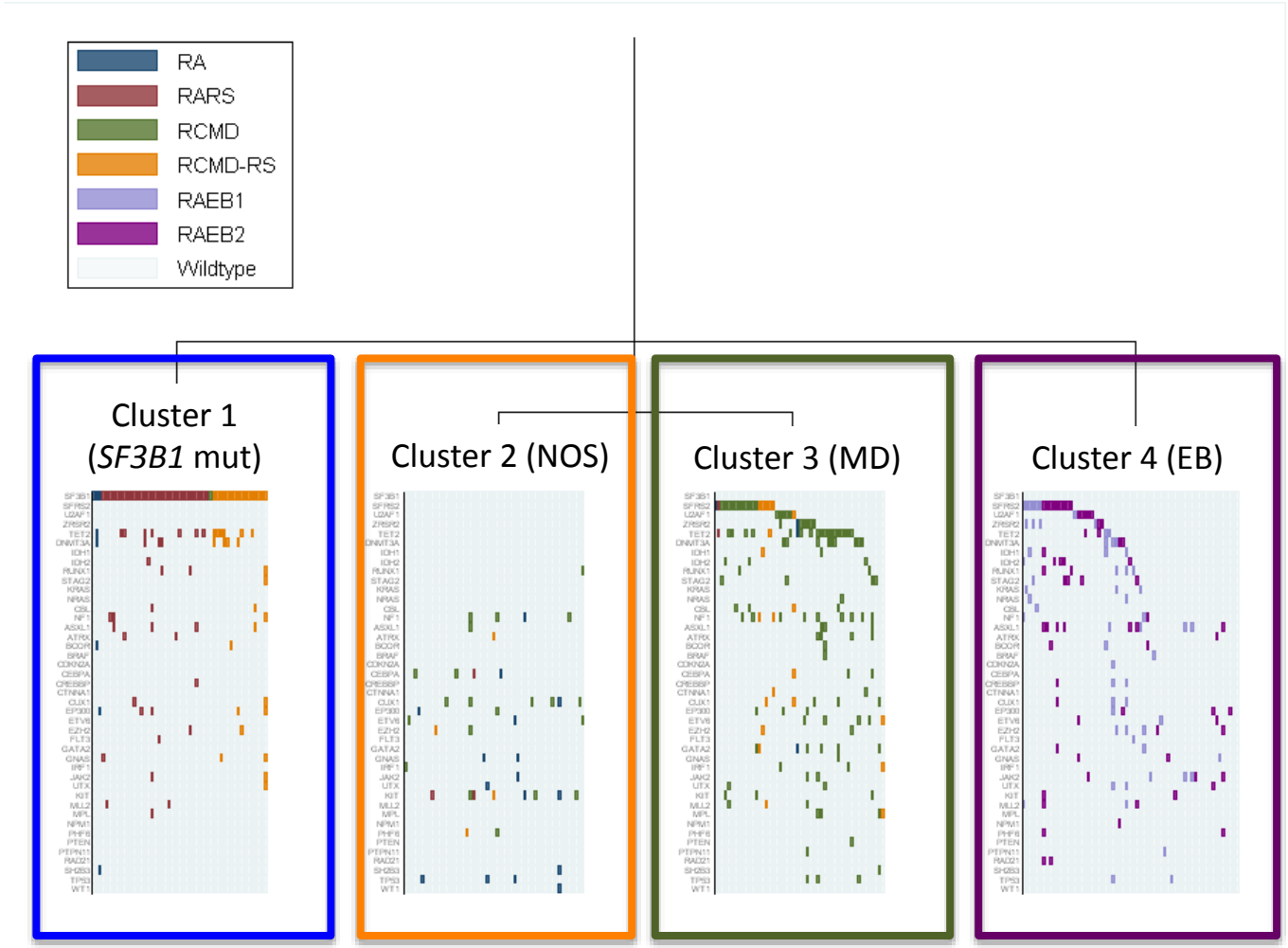
Genetic predestination

Genetic “predestination”: early founding driver mutations shape the future trajectories of clonal evolution of a cancer through constraints on the repertoire of cooperating subclonal genetic lesions



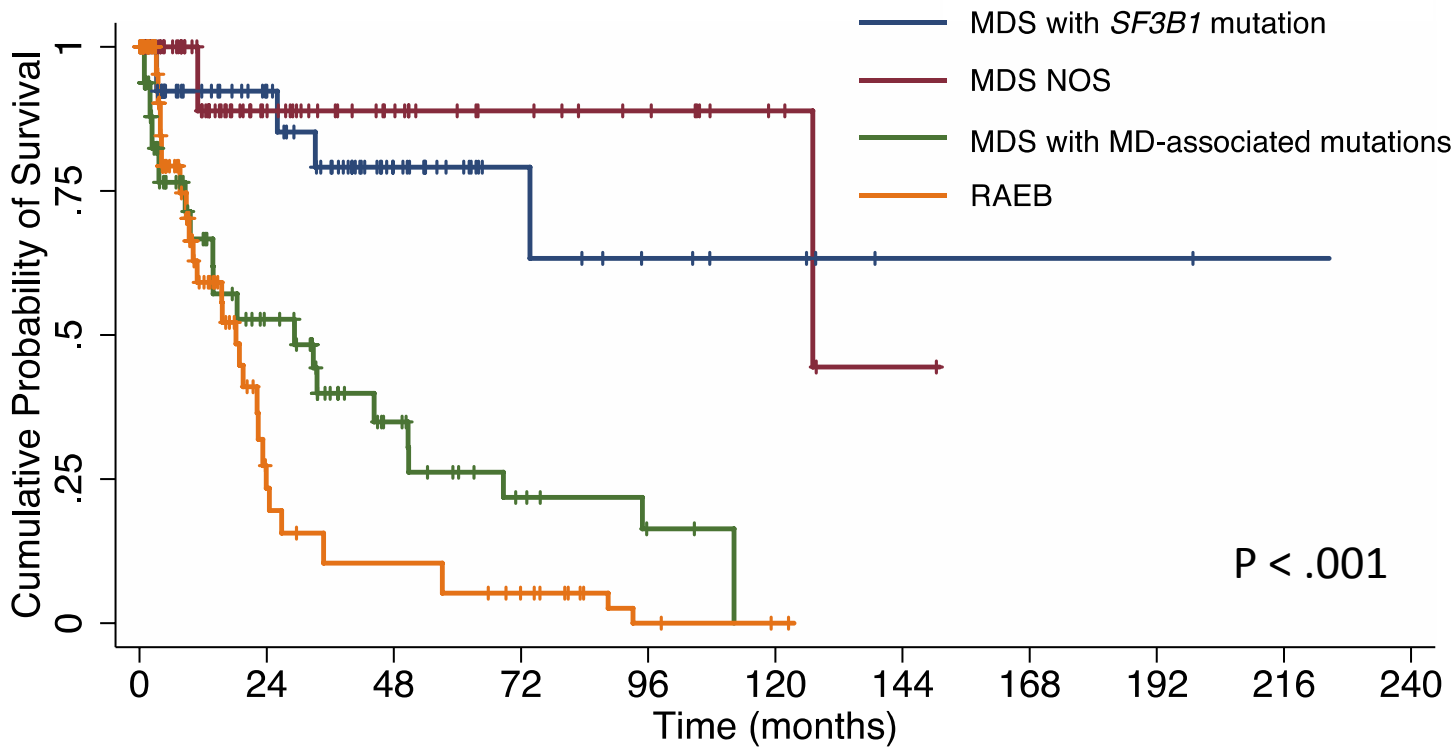
Papaemmanuil et al. Blood. 2013 Nov 21;122(22):3616-27

Unsupervised hierarchical clustering analysis of MDS patients



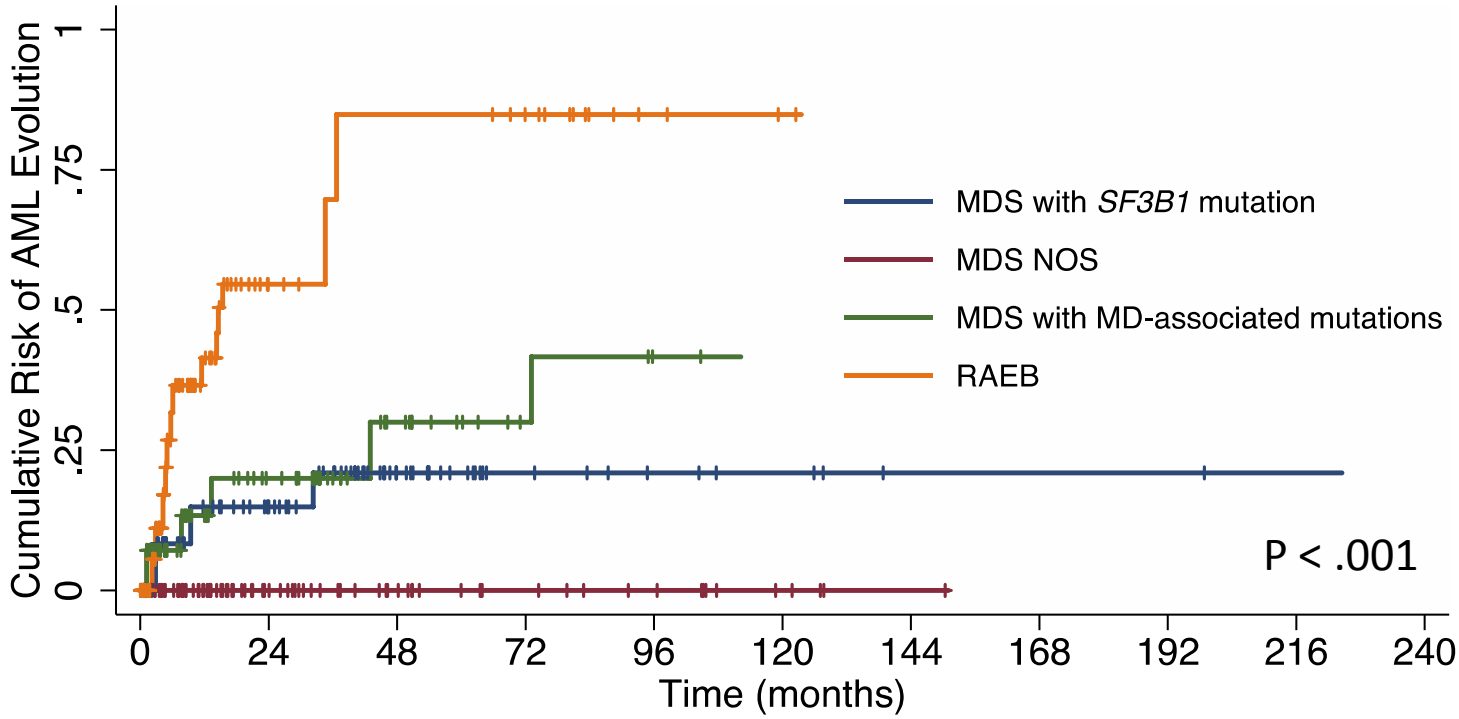
Malcovati et al. Blood. 2014 Aug 28;124(9):1513-21

Overall survival of MDS patients stratified according to genotype and blast excess



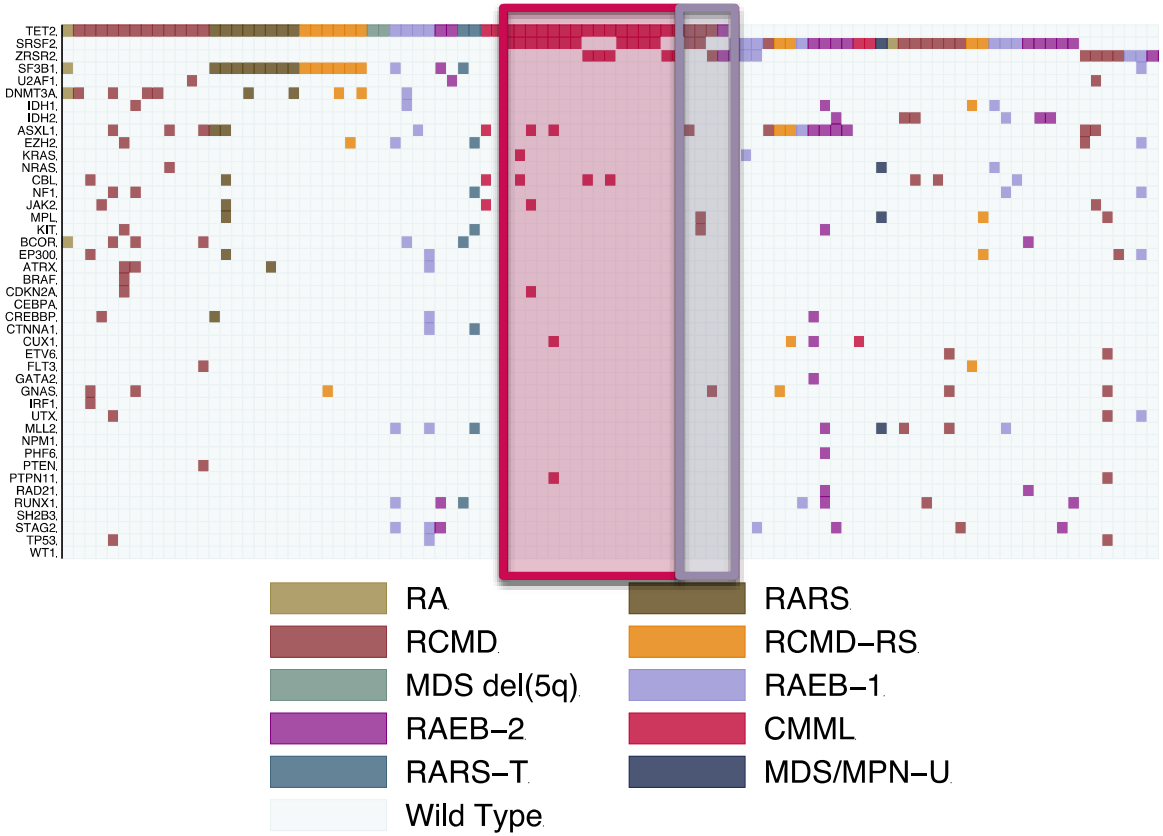
Malcovati et al. Blood. 2014 Aug 28;124(9):1513-21

Progression to AML in MDS patients stratified according to genotype and blast excess



Malcovati et al. Blood. 2014 Aug 28;124(9):1513-21

Co-occurrence of *TET2* and *SRSF2* (or *ZRSR2*) mutations is highly specific for myelomonocytic phenotype



CMML: Monocyte count ≥ 1.5 x 10^9/L or *TET2/SRSF2* co-mutation?

Malcovati et al. Blood. 2014 Aug 28;124(9):1513-21

Somatic mutations of *ASXL1*, *RUNX1* and *SETBP1* improve prognostic stratification of CMML

- *TET2* (44%), *SRSF2* (43%), *ASXL1* (34%), *KRAS* (11%), *NRAS* (10%), *CUX1* (10%), *CBL* (9%), *RUNX1* (7%), *SETBP1* (7%), *JAK2* (6%), *SF3B1* (6%), and *U2AF1* (5%)

- Lasso Cox regression model for genetic variable selection. The statistically significant variables were CPSS-specific cytogenetic risk groups (HR=2.49, P=.001), mutations in *ASXL1* (HR=2.77, P=.018), *RUNX1* (HR=5.39, P=.009) and *SETBP1* (HR=3.96, P=.013).

- CPSS-Mol performed better than the original CPSS cytogenetic risk classification

Elena et al. ASH 2014, abstract #1915

Conclusions

- The identification of somatic mutations of RNA splicing machinery has provided a paradigm shift

- Already established genotype/phenotype relationships include
  - *SF3B1*-mutant MDS
  - *TET2/SRSF2*-comutant MDS/MPN (CMML)

- The time has come for us to develop a genotype-based (molecular) classification of MDS

Acknowledgments

mds foundation
the myelodysplastic syndromes foundation, inc.
MDS - A bone marrow failure disorder

REL
RETE
EMATOLOGICA
LOMBARDA

AIRC

Associazione Italiana per la Ricerca sul Cancro

Pavia:

Luca Malcovati
Matteo G. Della Porta
Cristiana Pascutto
Ilaria Ambaglio
Antonio Bianchessi
Elisa Bono
Chiara Elena
Anna Gallì
Erica Travaglino
Marta Ubezio
Emanuela Boveri
Rosangela Invernizzi

International collaborations:

- Eva Hellström-Lindberg & Karolinska investigators
- Jackie Boultwood & Oxford investigators
- Elli Papaemmanuil & Cambridge investigators
- Ulrich Germing & Duesseldorf Investigators
- Guillermo Sanz & Spanish Investigators
- Seishi Ogawa & Kyoto Investigators