Next-Generation Sequencing for the Detection of Somatic Mutations in Myeloid Neoplasms

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Genomic Testing for Haematological Malignancies

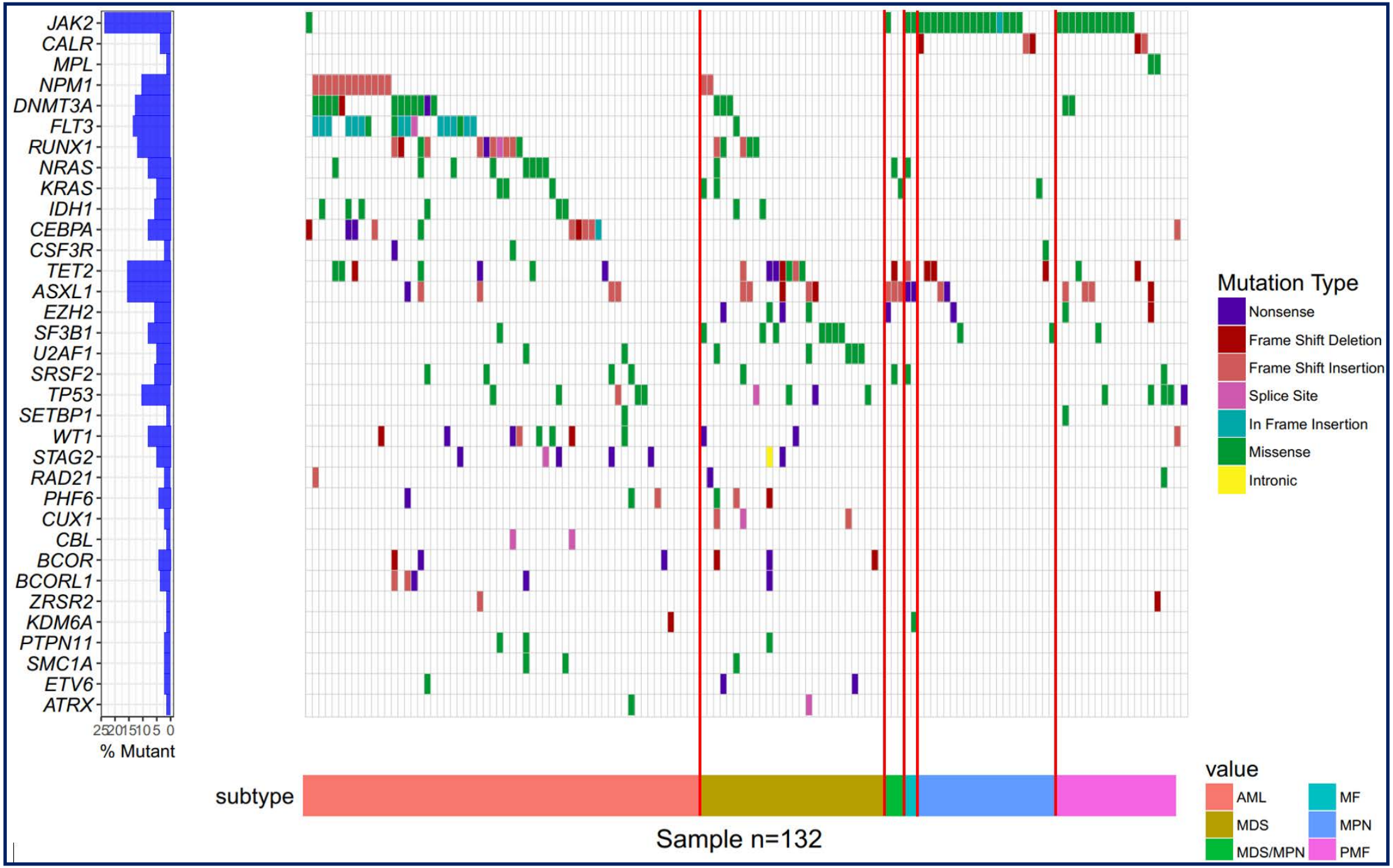
Myeloid Panel in Queen Mary Hospital

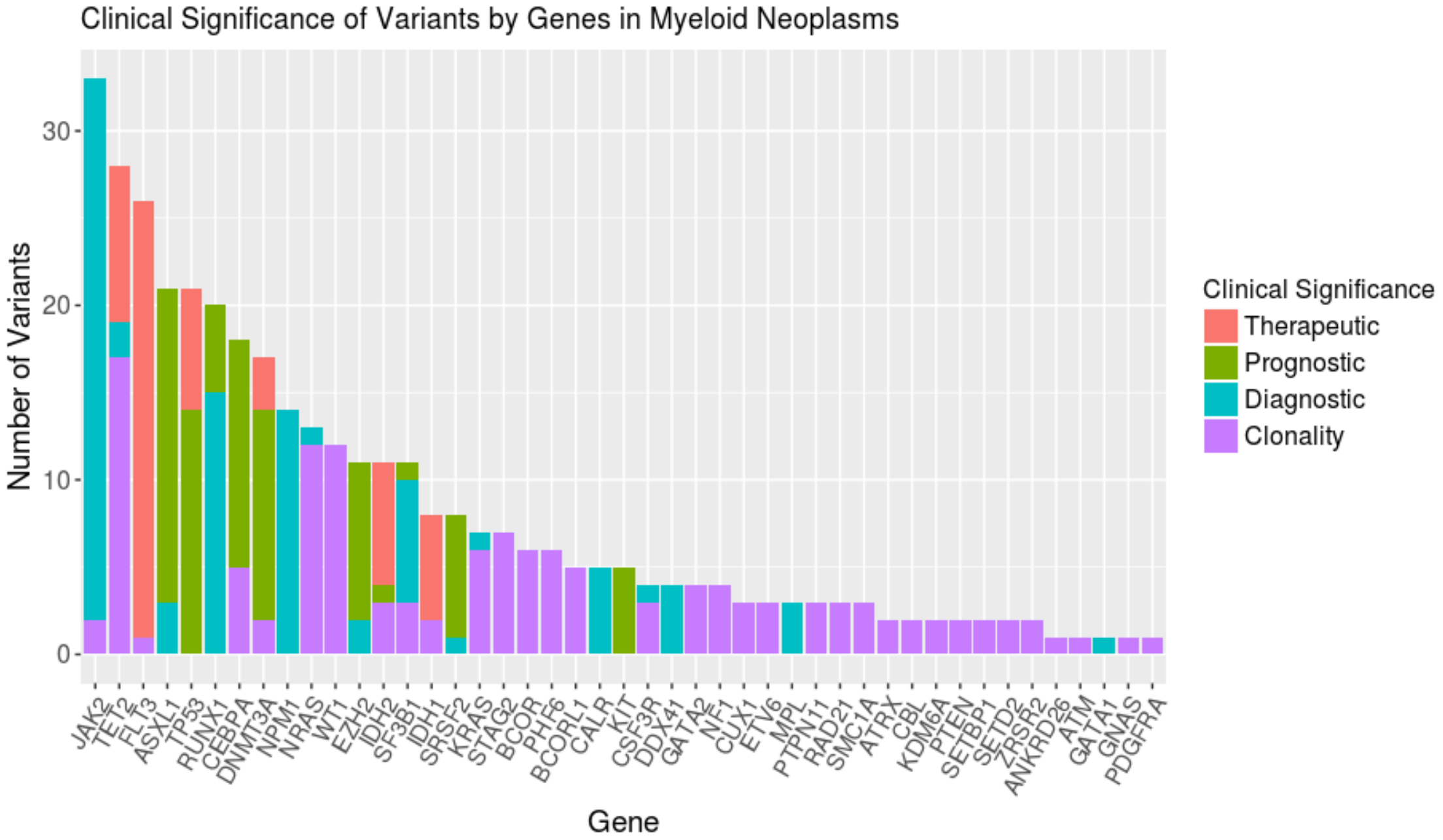
- 67 genes implicated to be of diagnostic, prognostic and therapeutic significance in myeloid neoplasms
  - Acute myeloid leukaemia (AML)
  - Myelodysplastic syndrome (MDS)
  - Myeloproliferative neoplasms (MPN)
  - Myelodysplastic/myeloproliferative neoplasms (MDS/MPN)

- Hybridisation-based capture approach

- Paired-end sequencing by Illumina MiniSeq sequencer

- Tumour sample: Peripheral blood or bone marrow aspirate
- Matched normal sample: Buccal swab


JAK2
CALR
MPL
NPM1
DNMT3A
FLT3
RUNX1
NRAS
KRAS
IDH1
CEBPA
CSF3R
TET2
ASXL1
EZH2
SF3B1
U2AF1
SRSF2
TP53
SETBP1
WT1
STAG2
RAD21
PHF6
CUX1
CBL
BCOR
BCORL1
ZRSR2
KDM6A
PTPN11
SMC1A
ETV6
ATRX
25 20 15 10 5 0
% Mutant
Mutation Type
Nonsense
Frame Shift Deletion
Frame Shift Insertion
Splice Site
In Frame Insertion
Missense
Intronic
subtype
value
AML
MDS
MDS/MPN
MF
MPN
PMF
Sample n=132


Clinical Significance of Variants by Genes in Myeloid Neoplasms
Number of Variants
30
20
10
0
JAK2
TET2
FLT3
ASXL1
TP53
RUNX1
CEBPA
DNMT3A
NPM1
NRAS
WT1
EZH2
IDH2
SF3B1
IDH1
SRSF2
KRAS
STAG2
BCOR
PHF6
BCORL1
CALR
KIT
CSF3R
DDX41
GATA2
NF1
CUX1
ETV6
MPL
PTPN11
RAD21
SMC1A
ATRX
CBL
KDM6A
PTEN
SETBP1
SETD2
ZRSR2
ANKRD26
ATM
GATA1
GNAS
PDGFRA
Gene
Clinical Significance
Therapeutic
Prognostic
Diagnostic
Clonality

Clinical Roles of NGS Myeloid Panel

Myeloid Neoplasms with Germline Predispositions

Myeloid neoplasm classification
Myeloid neoplasms with germ line predisposition without a preexisting disorder or organ dysfunction
AML with germ line *CEBPA* mutation
Myeloid neoplasms with germ line *DDX41* mutation*
Myeloid neoplasms with germ line predisposition and preexisting platelet disorders
Myeloid neoplasms with germ line *RUNX1* mutation*
Myeloid neoplasms with germ line *ANKRD26* mutation*
Myeloid neoplasms with germ line *ETV6* mutation*
Myeloid neoplasms with germ line predisposition and other organ dysfunction
Myeloid neoplasms with germ line *GATA2* mutation
Myeloid neoplasms associated with BM failure syndromes
Myeloid neoplasms associated with telomere biology disorders
JMML associated with neurofibromatosis, Noonan syndrome or
Noonan syndrome-like disorders
Myeloid neoplasms associated with Down syndrome*

- M/75
- AML with minimal differentiation
- No family history
- Normal karyotype
- *DDX41* germline p.Gln306*
- *DDX41* somatic p.Arg525His

- M/57
- MDS-EB-1
- Father died of acute leukaemia at 60 y.o.
- Normal karyotype
- *DDX41* germline p.Tyr259Cys
- *DDX41* somatic p.Arg525His
- Referred for HSCT
- 12 siblings. 3 HLA-matched.

Arber DA et al. The 2016 revision to the World Health Organization classification of myeloid neoplasms and acute leukemia. *Blood* 2016;127:2391-2405.

Clinical Roles of NGS Myeloid Panel

Summary

- High positive rate (91%) for variants in myeloid neoplasms
- Most detected variants have some forms of clinical significance
- Facilitate detection of variants in multiple genes simultaneously
- Cost reduction per gene tested