Genomics of multiple myeloma
Gareth J Morgan
Director NYULangone Myeloma Research Program
Talk outline

1. Introduction
2. Aetiologic events
3. The Myeloma Genome project
4. Epigenetic events
5. Risk stratification
6. Impact on the microenvironment
7. Structural events and identifying new drivers
We know it's illogical – "MULTIPLE" myelomas.

One size fits all.
Natural history of Multiple Myeloma.

Myeloma remains an incurable disease for most patients.

MGUS, monoclonal gammopathy of unknown significance; M protein, myeloma protein.

Personalize therapeutic decisions.

Younger, fit
• Achieving longest possible remission/sustained disease control while preserving QoL?

Elderly, fit
• Achieving and maintaining responses while preserving QoL?

Frail/comorbidities
• Tolerability while preserving QoL?

Very frail
• Palliative care while preserving QoL?
Personalized medicine
Multistep Progression System

Initiation

Germinal centre

Post-GC B cell

Bone marrow

MGUS

Progression

Smouldering Myeloma

Myeloma

Plasma Cell Leukemia

inherited variants

**PRIMARY GENETIC EVENTS**
- IGH TRANSLOCATIONS
- HYPERDIPLOIDY

**SECONDARY GENETIC EVENTS**
- COPY NUMBER ABNORMALITIES
- DNA HYPMETHYLATION
- ACQUIRED MUTATIONS
2. Aetiological Copy Number Abnormalities and Structural Variants
2. Aetiologic Events in Multiple Myeloma

<table>
<thead>
<tr>
<th>Primary genetic events (initiation events)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>IGH translocations (50%)</strong></td>
</tr>
<tr>
<td>• t(4;14)  FGFR3/MMSET (15%)</td>
</tr>
<tr>
<td>• t(6;14)  CCND3 (4%)</td>
</tr>
<tr>
<td>• t(11;14) CCND1 (20%)</td>
</tr>
<tr>
<td>• t(14;16) MAF (4%)</td>
</tr>
<tr>
<td>• t(14;20) MAFB</td>
</tr>
<tr>
<td><strong>Hyperdiploidy (50%)</strong></td>
</tr>
<tr>
<td>• Trisomies of chromosomes</td>
</tr>
<tr>
<td>3, 5, 7, 9, 11, 15, 19, 21</td>
</tr>
</tbody>
</table>

None 60%

- 11q13 (Cyclin D1) 15%
- 6p21 (Cyclin D3) 3%
- 4p16 (MMSET) 15%
- 16q23 (c-MAF) 5%
- 20q12 (MAFB) 2%

Morgan GJ, Walker BA and Davies FE. Nature Reviews Cancer. *In 2012*
“A unifying aetiologic classification of MM is based on the presence of chromosomal translocations and the deregulation of a D group cyclin either directly by a translocation or by an unknown mechanism.”

MM is comprised of at least six molecular categories.

G1/S checkpoint

Tumour initiating events
- t(14:16) MAF or MAFB
- t(14:20) MAF or MAFB
- t(4:14) MMSET
- Cyclin D2 CDK4 or CDK6
- Cyclin D1 CDK4 or CDK6
- Cyclin D3 CDK4 or CDK6

Secondary events
- MYC RAS
- 1q21 gain CKS1B

G1 phase
- Homozygous inactivation
  - 13q14 loss
  - Mutation

S phase
- RB2 E2F
- E2F
- RB2

Legend:
- Translocation
- Chromosomal gains by hyperdiploidy (HRD)
- Gene or protein-level expression changes
- Direct effect
- Indirect effect

Pawlyn C and Morgan GJ. Nat Rev Cancer. 2017;17(9):543-556
3. Myeloma Genome Project

DNA
- ~600 ex. panel
- ~1700 WES
- ~1000 WGS

RNA
- ~1100 RNAseq

Clinical

Molecular profiling leads to stratification of patients into groups:
- Group 1
- Group 2
- Group 3
- Group 4
Molecular Risk Stratification

N = 1273 exomes

1) Obtain significant features by univariate tests
2) Assess significant features by multivariate model

Stratified partitioning for age, ISS, study

Recursive Partitioning Model for Risk Stratification (n=784)
Bi-allelic Inactivation of Tumour Suppressor Genes
Multiple Myeloma

NFκB
G1S
P53
Pan-cancer Homozygous Deletions
Multiple Myeloma Driver Genes (n=63)

Walker et al. Blood 2018
Mutation Frequencies of 63 Driver Genes
Walker et al. Blood 2018
Mutation Frequencies of 63 Driver Genes

VEMURAFINIB
DEBRAFINIB

KRAS

RAS
Cancer Clonal Fraction of Driver Genes

Walker et al. Blood 2018
Myeloma as an Evolutionary Ecosystem

Ecosystem 1
- Single founder tumour initiating cell

Ecosystem 2
- Diffuse
- Focal

Ecosystem 3
- EMD

Ecosystem 4
-MGUS
- MM
- PCL

Selective pressures
Mutation Targeted Therapy; Implications of Sub-clonal Heterogeneity

Venetoclax t(11;14)

Target the trunk not the branches!
• Distribution of mutations and copy number variables is not random providing evidence for distinct subgroups of disease.
• The aetiologic events provide a distinct genetic background on which collaborating variants are super imposed.
Clustering by Copy Number Abnormality; Nine Copy Number Sub-Groups
4. Epigenetic Changes; t(4;14)

Immunoglobulin gene locus

Enhancer

Hybrid gene

FGFr3

Ig

NSD2

IgH

IGHD

IGHC

JAG2

D14S681E

D14S523

D14S398

RMS59237

D4S523

4p16

4q32

14q32

14

1 MB

600 KB

600 KB
NSD2 is H3K36me3 Wipes Out Repressive H3K27me3 Peaks to Activate Genes
Balance of H3K27me/Ac regulates key processes

The complex is frequently targeted by mutation in MM

Modified from Ezponda & Licht, Clin Cancer Res 2014
## Epigenetic Driver Mutations

<table>
<thead>
<tr>
<th>Gene</th>
<th>Cytoband</th>
<th>Full name</th>
<th>Function</th>
<th>Frequency (1273)</th>
<th>Chromosome</th>
<th>Score</th>
</tr>
</thead>
<tbody>
<tr>
<td>ARID1A</td>
<td>1p36.11</td>
<td>AT-rich Interaction Domain 1A</td>
<td>SWI/SNF</td>
<td>31/1273 (2.44%)</td>
<td>1p36.11</td>
<td>TSG score</td>
</tr>
<tr>
<td>ARID2</td>
<td>12p12</td>
<td>AT-rich Interaction Domain 2</td>
<td>PBAF complex</td>
<td>17/1273 (1.34%)</td>
<td>12q12</td>
<td>TSG score</td>
</tr>
<tr>
<td>CREBBP</td>
<td>16p13.3</td>
<td>CREB Binding Protein</td>
<td>BRD and HAT</td>
<td>30/1273 (2.36%)</td>
<td>16p13.3</td>
<td>TSG score</td>
</tr>
<tr>
<td>DNMT3A</td>
<td>2p23.3</td>
<td>DNA Methyltransferase a</td>
<td>DNA methylation</td>
<td>22/1273 (1.73%)</td>
<td>2p23.3</td>
<td>TSG score</td>
</tr>
<tr>
<td>EP300</td>
<td>22q13.2</td>
<td>E1A Binding Protein P300</td>
<td>HAT</td>
<td>25/1273 (1.96%)</td>
<td>22q13.2</td>
<td>TSG score</td>
</tr>
<tr>
<td>HIST1H1E</td>
<td>6p22.2</td>
<td>Histone Cluster 1 H1 Family Member E</td>
<td>Epigenetic</td>
<td>47/1273 (3.69%)</td>
<td>6p22.2</td>
<td>TSG score</td>
</tr>
<tr>
<td>IDH1</td>
<td>2q34</td>
<td>Isocitrate Dehydrogenase</td>
<td>DNA methylation</td>
<td>7/1273 (0.55%)</td>
<td>2q34</td>
<td>ONC score</td>
</tr>
<tr>
<td>IDH2</td>
<td>15q26.1</td>
<td>Isocitrate Dehydrogenase</td>
<td>DNA methylation</td>
<td>4/1273 (0.31%)</td>
<td>15q26.1</td>
<td>ONC score</td>
</tr>
<tr>
<td>KDM5C (Jarid1c)</td>
<td>Xp11.22</td>
<td>Lysine Demethylase 5C</td>
<td>H3K4 demethylase</td>
<td>21/1273 (1.65%)</td>
<td>Xp11.2</td>
<td>TSG score</td>
</tr>
<tr>
<td>KDM6A (UTX)</td>
<td>Xp11.3</td>
<td>Lysine Demethylase 6A</td>
<td>H3K27 demethylase</td>
<td>19/1273 (1.49%)</td>
<td>Xp11.3</td>
<td>TSG score</td>
</tr>
<tr>
<td>KMT2B (MLL2)</td>
<td>19q13.12</td>
<td>Lysine Methyltransferase 2B</td>
<td>H3K4 methylation</td>
<td>28/1273 (2.2%)</td>
<td>19q13.1</td>
<td>TSG score</td>
</tr>
<tr>
<td>KMT2C (MLL3)</td>
<td>7q36.1</td>
<td>Lysine Methyltransferase 2C</td>
<td>H3K4 methylation</td>
<td>34/1273 (2.67%)</td>
<td>7q36.1</td>
<td>TSG score</td>
</tr>
<tr>
<td>NCOR1</td>
<td>17p12</td>
<td>Nuclear Receptor Corepressor 1</td>
<td>gene repression</td>
<td>17/1273 (1.34%)</td>
<td>17p12</td>
<td>TSG score</td>
</tr>
<tr>
<td>SETD2</td>
<td>3p21.31</td>
<td>SET Domain Containing 2</td>
<td>H3K36 me3</td>
<td>24/1273 (1.89%)</td>
<td>3p21.31</td>
<td>TSG score</td>
</tr>
<tr>
<td>TET2</td>
<td>4q24</td>
<td>Hydroxylation 5MeC</td>
<td>DNA methylation</td>
<td>24/1273 (1.89%)</td>
<td>4q24</td>
<td>TSG score</td>
</tr>
</tbody>
</table>

Myeloma genome project Walker et al. Blood 2018
5. Risk Stratification

Interphase FISH

– Adverse – t(4;14), t(14;16), del17p.
– Standard – hyperdiploidy, t(11;14).
– Ultra high risk – adverse translocation plus del17p
Revised International Staging System R-ISS

<table>
<thead>
<tr>
<th>Stg</th>
<th>Factor</th>
<th>% of pts</th>
<th>5 yr PFS</th>
<th>5 yr OS</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>Absence of adverse factors (no high LDH, ISS 2 or 3, t(4;14) t(14;16) or del(17p))</td>
<td>28%</td>
<td>55%</td>
<td>82%</td>
</tr>
<tr>
<td>II</td>
<td>Not R-ISS I or III</td>
<td>62%</td>
<td>36%</td>
<td>62%</td>
</tr>
<tr>
<td>III</td>
<td>ISS 3 and either high-risk CA by iFISH or high LDH</td>
<td>10%</td>
<td>24%</td>
<td>40%</td>
</tr>
</tbody>
</table>

Non–transplantation-based regimens

Transplantation-based regimens

Immunomodulatory-based-based regimens

Proteasome inhibitor-based regimens

Double Hit MM a new disease segment.

“Double hit” MM
a. Biallelic P53
b. ISS III amp CKS1B
c. 6% NDMM
Comparison of Double Hit MM to IMWG Risk Groups

A.

- IMWG: low/standard risk, RP-Risk: Low Risk
  - Events / N: 65 / 357
  - 18-Month Estimate: 87% (84, 91)
  - Events / N: 126 / 296
  - 18-Month Estimate: 67% (62, 73)
- IMWG: low/standard risk, RP-Risk: Double Hit
  - Events / N: 15 / 24
  - 18-Month Estimate: 44% (23, 65)
- IMWG: high risk, RP-Risk: Low Risk
  - Events / N: 10 / 30
  - 18-Month Estimate: 69% (51, 88)
- IMWG: high risk, RP-Risk: Intermediate Risk
  - Events / N: 21 / 53
  - 18-Month Estimate: 74% (61, 87)
- IMWG: high risk, RP-Risk: Double Hit
  - Events / N: 18 / 24
  - 18-Month Estimate: 35% (15, 54)

Log-rank p-value < .0001

B.

- IMWG: low/standard risk, RP-Risk: Low Risk
  - Deaths / N: 21 / 357
  - 18-Month Estimate: 96% (94, 98)
  - Deaths / N: 59 / 296
  - 18-Month Estimate: 85% (81, 90)
- IMWG: low/standard risk, RP-Risk: Double Hit
  - Deaths / N: 10 / 24
  - 18-Month Estimate: 73% (53, 93)
- IMWG: high risk, RP-Risk: Low Risk
  - Deaths / N: 6 / 30
  - 18-Month Estimate: 88% (74, 100)
- IMWG: high risk, RP-Risk: Intermediate Risk
  - Deaths / N: 6 / 53
  - 18-Month Estimate: 94% (86, 100)
- IMWG: high risk, RP-Risk: Double Hit
  - Deaths / N: 16 / 24
  - 18-Month Estimate: 37% (16, 58)

Log-rank p-value < .0001
Integrating mutation and expression data
12 Stable Patient Clusters in NDMM

- Multi-dimensional clustering analysis identified 12 stable patient segments (all ≥5%) with distinct outcomes
- This is the first large-scale analysis of ndMM to perform unsupervised biomarker analysis to stratify pts based on SV, CNA, GEP, and SNV differences
Molecular Features of Cluster 8

- 24/59 (41%) were ISS3
- 19/59 (32%) were ≥ 65 yrs

Cytogenetics

- Del1p13/22-21 (1e-3)
- Amp 1q21-25 (1e-2)

- Del13q12-34 (1e-5/3)
- Del14q12-32 (1e-7)
- Del16q12-24 (1e-4)

Copy Number

GE profile driven by significant low-expression of transcripts and up-regulation of cell-cycle pathways

Identifying Molecular Drivers of High-Risk

**Computational**
- Validation
- Master regulators
- Bionetworks

**Genetic**
- CRISPR screens
- shRNA knockdowns
- Targeted gene manipulations
- Chemical inhibition

**Biological**
- Proliferation
- Cell cycle
- Apoptosis
- 3D & Co-culture assays
- Immune assays

**Proteomic**
- Interaction mapping
- Immunoprecipitation
- Mass Spectrometry
- ChIP-Seq

- Pathways, genes and functional networks
- Biomarker
- Preclinical / ex-vivo models for testing function
6. Impact on the Microenvironment

Recurrent myeloma mutations
Recurrent copy number changes

Normal cell

MGUS
SMM
MM
EMD

HR
PCL

MYC translocations
P53 inactivation
Amp1q
Whole arm translocations
RB1 inactivation
CDKN2C inactivation
Increase in tumour promoting cells

High-risk states

T-regs
pDCs
MDSCs

Normal cell

MGUS
SMM
MM
EMD

MYC translocations
P53 inactivation
Amp1q
Whole arm translocations
RB1 inactivation
CDKN2C
High-risk states

- MYC translocations
- P53 inactivation
- Amp1q
- Whole arm translocations
- RB1 inactivation
- CDKN2C inactivation

Decrease in tumour suppressing cells

NK cells
Cytotoxic T cells
B cells
Th cells

Normal cell
MGUS
SMM
MM
EMD
Do the tumour cells contain information which is able to disorganize the microenvironment?
De-convoluting the immune micro-environment.
The Rosetta project.

- Purity
- Transcription profile
- Gene expression signature matrix (LM22)
- Leukocyte signature matrix (LM22)
- Newman et al., Nat Meth, 2015

- Purity
- Transcription profile
- Gene expression signature matrix (MGSM27)
- Myeloma gene signature matrix (MGSM27)
- 6 additional cell types

- Purity
- Transcription profile
- Gene expression signature matrix (MGSM27)
- Myeloma gene signature matrix (MGSM27)
- 5 additional cell types
- Matched aspirate
- CD138+ cell type

- 10% Tumor
- 05% Dendritic
- 15% Macrophage
- 07% Osteoblast

- ... 04% T-regs

---

- Remove Tumor
- 05% Dendritic
- 15% Macrophage
- 07% Osteoblast
- ... 04% T-regs

---

- Rescale to 100%

Black bar = pathologist PC % IHC
Red dot = deconvolution estimate
Gray shade = PC % error estimate based on IHC and Flow data devi
Sub-groups identified with distinct features independent of the major molecular subgroups of disease.

Cluster 5.
Cluster 5. High-risk microenvironment

- T cells
- NK cells
- Macrophage
- Mast cells
- Eosinophils

M2
M1
Plasma cell features associated with high-risk microenvironment

T cells↑  NK cells↓  Macrophage  Mast cells↓  Eosinophils↓

M2  M1

Interferon response genes IFIT1/3, ISG15, IFI6, IFI44L

Hepatocyte growth factor

VCAM

Proteoglycan 2

Cancer testis antigen FAM133A

SMAD, RASSF6, GBP1, MITD1, MAD2L1, NDC80 APOBEC3B

Plasma cell

Extracellular matrix

Stromal cell
7. Identifying New Drivers

A Significant Percentage of Patients (15%) Have No Detectable Driver

Walker et al. Blood 2018
We are missing drivers either because they occur by a mechanism its difficult to find or they occur in a part of the genome we haven’t examined.
The Non-coding Regions of the Multiple Myeloma Genome

10x Chromium WGS

SVs
SNVs
Copy Number

Enriched for high risk molecular features

UAMS

10X Chromium WGS
(N = 95)

RNA-seq
(N = 85)

Analysis
Control-FREEC – copy number
Longranger – structural events

N = 95 whole genome sequencing
Structural Variants and Gene Deregulation

- Amplification
- Fusion genes
- Gene overexpression by superenhancers
- Gene knockout
1q amp and JT1q12

• 1q+ is present in 40% presenting MM and is a poor prognostic feature.
• Frequently amplified by duplication or breakage fusion breakage cycles.
• 1q12 site of hypomethylation which may be an important mechanism.
• Genes at locus include CKS1B, ANP32E, BCL9
• Translocates to receptor chromosomes and causes gain and loss of copy number
• Deregulates MYC, BCL2, WWOX, CYLD, 17p-

Sawyers et al 2014 Blood
Functional Groups of Fusion Genes

Identified fusion genes can be split into functional categories:

**Kinases** –
- *EML4-ALK* identified in NSCLC
- *BRAF* fusions usually result in dimerization and activation
- *NTRK3* fusions found in multiple cancer types at low frequencies
- *ROS1* fusions in NSCLC

**Transcription Factors** –
- *MYC* fusions due to common translocations in MM
- *EIF4E3-FOXP1* seen in breast cancer
- *CDC6-RARA* identified in NSCLC

<table>
<thead>
<tr>
<th>Kinase</th>
<th>Transcription Factor</th>
<th>Others</th>
</tr>
</thead>
<tbody>
<tr>
<td>EML4-ALK</td>
<td>EIF4E3-FOXP1</td>
<td>TBL1XR1-ATR</td>
</tr>
<tr>
<td>AGK-BRAF</td>
<td>EIF4E3-FOXP1</td>
<td>ATM-DLG2</td>
</tr>
<tr>
<td>GTF2I-BRAF</td>
<td>EIF4E3-FOXP1</td>
<td>MED15-EP300</td>
</tr>
<tr>
<td>ESYT2-BRAF</td>
<td>TXNDC5-MYC</td>
<td>KAT6A-EYS</td>
</tr>
<tr>
<td>KANK-BRAF</td>
<td>TXNDC5-MYC</td>
<td>MKL1-LTBR</td>
</tr>
<tr>
<td>SNX29-FGFR1</td>
<td>TXNDC5-MYC</td>
<td>SLC5A5-MYO18A</td>
</tr>
<tr>
<td>ARHGAP27-MAP3K14</td>
<td>FOXO3-MYC</td>
<td>EWSR1-PKDREJ</td>
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<tr>
<td>AKT1-MAPK14</td>
<td>CDC6-RARA</td>
<td>HDAC4-PLEKHM3</td>
</tr>
<tr>
<td>HNRNPA2B1-NTRK3</td>
<td>DIP2B-ATF1</td>
<td>ABL1-RBM18</td>
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<tr>
<td>UBE2R2-NTRK3</td>
<td>DUSP22-IRF4</td>
<td>STK11-RTDR1</td>
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<tr>
<td>TSPAN3-ROS1</td>
<td>SS18-FLI1</td>
<td>TBL1XR1-SLC9C1</td>
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<tr>
<td></td>
<td>ATF1-GALNT6</td>
<td>CREBBP-SLX4</td>
</tr>
<tr>
<td></td>
<td>RUNX1-LINCO0160</td>
<td>STT3B-TBL1XR1</td>
</tr>
<tr>
<td></td>
<td></td>
<td>RUNX1T1-TBL1XR1</td>
</tr>
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</table>
Translocations Drive Gene Overexpression via the Ig Gene Super-enhancers

Rearrangement of super-enhancers

Ig locus

FGFR3

Overexpression of FGFR3
Novel Ig Rearrangements

We detected five novel significantly overexpressed genes when looking at structural events involving IgK, IgH and IgL.

<table>
<thead>
<tr>
<th>symbol</th>
<th>cytoband</th>
<th>Adjusted P</th>
<th>Expression (event)</th>
<th>Expression (no event)</th>
<th>Fold Change</th>
<th>Percent</th>
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<tbody>
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<td>MAP3K14</td>
<td>17q21.31</td>
<td>2.18E-55</td>
<td>15.0</td>
<td>10.7</td>
<td>18.6</td>
<td>1.6%</td>
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<tr>
<td>CCND2</td>
<td>12p13.32</td>
<td>6.46E-06</td>
<td>18.4</td>
<td>8.4</td>
<td>1010.8</td>
<td>1.1%</td>
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<td>LRRC31</td>
<td>3q26.2</td>
<td>2.56E-16</td>
<td>8.0</td>
<td>0.9</td>
<td>137.4</td>
<td>0.4%</td>
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<tr>
<td>PAX5</td>
<td>9p13.2</td>
<td>0.00019762</td>
<td>15.8</td>
<td>8.2</td>
<td>187.9</td>
<td>0.4%</td>
</tr>
<tr>
<td>ZFP36L1</td>
<td>14q23.3,14q24.1</td>
<td>0.03199773</td>
<td>14.4</td>
<td>12.0</td>
<td>5.4</td>
<td>0.7%</td>
</tr>
</tbody>
</table>
Plasma Cell Super-Enhancers

Selective Inhibition of Tumor Oncogenes by Disruption of Super-Enhancers

Jakob Lovén,1,7 Heather A. Hoke,1,2,7 Charles Y. Lin,1,3,5,7 Ashley Lau,1,2 David A. Orlando,1 Christopher R. Vakoc,4 James E. Bradner,5,6 Tong Ihn Lee,1 and Richard A. Young1,2,*

1Whitehead Institute for Biomedical Research, 9 Cambridge Center, Cambridge, MA 02142, USA
2Department of Biology
3Computational and Systems Biology Program
Massachusetts Institute of Technology, Cambridge, MA 02139, USA
4Cold Spring Harbor Laboratory, 1 Bungtown Road, Cold Spring Harbor, NY 11724, USA
5Department of Medical Oncology, Dana-Farber Cancer Institute, 44 Binney Street, Boston, MA 02115, USA
6Department of Medicine, Harvard Medical School, 25 Shattuck Street, Boston, MA 02115, USA
*These authors contributed equally to this work
*Correspondence: young@wi.mit.edu
http://dx.doi.org/10.1016/j.cell.2013.03.036

BRD4 and MED1 ChIP-seq to identify super-enhancer elements in MM1s cells
Canonical and novel Ig and MYC rearrangements

MAP3K14 event: IGH IG1:MAP3K14_region (p = 0)

no event  with event
Chromatin Organization

“From a linear genome to a 3D structure.”

Topologically associated domains

Spielmann et al. Nature Reviews Genetics 2018
Maintenance of TAD Boundaries is Critical

Maintenance of TAD boundaries is crucial to maintaining correct gene expression profiles.

Impacted by multiple mechanisms
Maintenance of TAD boundaries is crucial to maintaining correct gene expression profiles.
Hypothesis
Translocations Break TAD Boundaries

“Structural rearrangements function mediating gene dysregulation by the hijacking of the super-enhancers of donor TADs and placing them within a receptor TAD to drive expression of oncogenes”

Spielmann et al. Nature Reviews Genetics 2018
Potential New Drivers Identified by Rearrangement to a Super Enhancer

<table>
<thead>
<tr>
<th>symbol</th>
<th>cytoband</th>
<th>Adjusted P</th>
<th>Expression (event)</th>
<th>Expression (no event)</th>
<th>Fold Change</th>
<th>Percent</th>
<th># IG</th>
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<tr>
<td>PAX5</td>
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<td>4.64E-05</td>
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<td>LIPG</td>
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<tr>
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<td>0.0400851</td>
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<td>0.0029752</td>
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<tr>
<td>MAP3K14</td>
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<tr>
<td>PPIL1</td>
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<td>10.3</td>
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<td>LRRC37A</td>
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</tr>
</tbody>
</table>
MM is a Disease of Abnormal Transcription

Bromodomains
- Recognize acetylation marks in histone tails
- Recruit transcriptional machinery
- Promote gene transcription

Bromodomain inhibitors
- Prevent bromodomains from interacting with acetyl group
- Downregulate gene transcription

Structural Events are Common

- Complex and simple.
- Median number of structural variants is 25 (range 1 – 182).
  - Intra chromosomal events more frequent median of 14 (range 1 - 179) (P<0.001).
  - Inter-chromosomal events median of 7 (range 0 - 29).
- Events seen on all chromosomes but most frequent
  - Chromosomes 14 – 64%.
  - Chromosome 8 – 53%.
  - Chromosome 1 - 44%.
  - Chromosome 6 – 42%.
Chromothripsis involve 1 or 2 chromosomes shattering and recombining. Thought to be a single catastrophic event.

We identified 21% of samples with chromothripsis.
Chromoplexy results in derivative chromosomes

An example where chromoplexy developed sequentially over time

(t(1;8;19))

(t(1;8;16;19))
Co-existence of chromoplexy and chromothripsis impacts survival

- Called rearrangements chromoplexy and chromothripsis on 500 cases with low depth whole genome sequencing.
- Samples with both chromoplexy and chromothripsis are associated with a shorter time to progression (P<0.001).
Genetic Events Associated With Multiple Myeloma

### Primary genetic events

**IGH translocations (50%)**
- t(4;14) FGFR3/MMSET (15%)
- t(6;14) CCND3 (4%)
- t(11;14) CCND1 (20%)
- t(14;16) MAF (4%)
- t(14;20) MAFB
- t(14;17) MAP3K14

**Hyperdiploidy (50%)**
Trisomies of chromosomes 3, 5, 7, 9, 11, 15, 19, 21
9 clusters

### Secondary genetic events

#### Copy number abnormalities

<table>
<thead>
<tr>
<th>Deletion</th>
<th>Gain</th>
</tr>
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<tbody>
<tr>
<td>Deletion 1p (30%) CDKN2C, FAF1, FAM65C</td>
<td>Gain 1q (40%) CKS1B, ANP32E</td>
</tr>
<tr>
<td>Deletion 6q (33%)</td>
<td>Gain LTBR</td>
</tr>
<tr>
<td>Deletion 8p (25%)</td>
<td>Gain TACI</td>
</tr>
<tr>
<td>Deletion 13 (44%) RB1, DIS3</td>
<td>Gain NIK</td>
</tr>
<tr>
<td>Deletion 11q (7%) BIRC2/BIRC3</td>
<td>Deletion 6q (33%)</td>
</tr>
<tr>
<td>Deletion 14q (38%) TRAF3</td>
<td>Deletion 8p (25%)</td>
</tr>
<tr>
<td>Deletion 13 (44%) RB1, DIS3</td>
<td>Deletion 14q (38%) TRAF3</td>
</tr>
<tr>
<td>Deletion 17p (8%) TPS3</td>
<td>Deletion 13 (44%) RB1, DIS3</td>
</tr>
</tbody>
</table>

#### Secondary structural rearrangements
- t(8;14)
- Fusion genes
- Non IGH translocations
- Chromoplexy
- Chromothripsis

### Mutational events and the molecular hallmarks of myeloma

- **A** – Immortalisation
- **B** – G1S abnormality
- **C** – Proliferation
- **D** – Resistance to apoptosis
- **E** – Abnormal localisation and bone disease
- **F** – Abnormal PC Differentiation
- **G** – Abnormal DNA repair
- **H** – RNA editing
- **I** – Epigenetic/PRC complex
- **J** – Abnormal Immune microenvironment
- **K** – Abnormal energy metabolism and ADME events

#### Epigenetic events

- Global hypomethylation from MGUS to myeloma
- Gene specific hypermethylation from myeloma to plasma cell leukaemia
- Epigenetic mutation and the compass complex

---

MyDrUG trial

Functional High Risk Patients

Profiling for alterations (NCT02884102)

- No detectable “actionable” alterations
- RAF/RAS mutations
- IDH activating mutations
- CDK pathway activating alterations
- FGFR3 activating alterations
- Other activating alterations
- t(11;14)

- No detectable “actionable” alterations
- MEKi + Dex
- IDHi + Dex
- CDKi + Dex
- FGFR3i + Dex
- Other + IPD
- 2 cycles

- Anti-CD38 + IPD
- Other + IPD
- MEKi + IPD
- IDHi + IPD
- CDKi + IPD
- FGFR3i + IPD
- Other + IPD
- BCLi + IPD

KRAS: 23%
NRAS: 26%
CDK activating: 6%
t(11;14): 7%
Others: 1%
None: 24%
IDH2: 3%
BRAF (V600E): 4%
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Michael Bauer
Niels Weinhold
Leo Rasche
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Samrat Roy Choudhury
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Thanendraraj
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