UTILIZATION OF A SNP MICROARRAY FOR THE STUDY OF MULTIPLE MYELOMA PATIENTS: NOVEL FINDINGS AND BETTER STRATIFICATION OF PATIENTS

S Schepis, P Papenhausen and S Schwartz, Laboratory Corporation of America® Holdings
OBJECTIVES

• Delineate the usefulness of array as a diagnostic and prognostic tool in multiple myeloma
• Determine array effectiveness of the array if…
  – FISH is normal
  – FISH is abnormal
• Demonstrate the occurrence of copy-neutral loss of heterozygosity (CN-LOH)
• Determine the best way to approach laboratory testing for multiple myeloma
INTRODUCTION

• MM testing for most laboratories
  – Chromosome analysis
    • ~30 abnormal
  – FISH on enriched cells using multiple probes
    • ~80% abnormal
• Should consider array as 1st tier test in conjunction with MM translocation
  – Use on enriched cells
  – Reduce large number of FISH probes currently used
• 413 MM patients in this study

[INTERNAL DATA – MARCH, 2017]
MM Prognostic Factors

- Good prognostic factors
  - Hyperdiploidy
  - t(11;14)

- Poor prognostic factors
  - Hypodiploidy
  - 1q gain; 17p deletion
  - T(4;14); t(14;16)

- Array provides additional important prognostic information
OVERALL FINDINGS

• Array detected...
  – Abnormalities in 99.9% of samples
  – Additional abnormalities in 99.8% samples abnormal by FISH
  – Additional poor prognostic factors not detectable by standard FISH
    • 8q duplications (MYC)
    • 12p deletions (ETV6)
    • 16q deletions (MAF)
    • CN-LOH
    • Chromothripsis
  – Higher than expected frequency of copy-neutral loss of heterozygosity

[INTERNAL DATA – MARCH, 2017]
HYPERDIPLOIDY

• Hyperdiploidy - good prognosis
  – Only hyperdiploidy: 38 (~18%)
  – Additional findings not thought to be poor prognostic factors: 72 (~34%)

• Hyperdiploidy - with poor prognostic factors
  – 98 cases (~47%)
    • 1q gain
    • 8q gain (MYC)
    • 12p deletion (ETV6)
    • 16q deletion (MAF)
    • Chromothriipsis
    • CN-LOH

• Most common chromosomes found to be hyperdiploid by array
  – 3, 5, 7, 9, 11, 15, 19

[INTERNAL DATA – MARCH, 2017]
11;14 TRANSLOCATION

- 11;14 translocation – good prognosis
  - Array additional poor prognostic factors
    - 21 out of 51 cases (~41%)
    - 8q gain (MYC)
    - 12p deletion (ETV6)
    - 16q deletion (MAF)
    - CN-LOH
    - Chromothripsis

[INTERNAL DATA – MARCH, 2017]
CHROMOTHRIIPSIS

• Considered if 10 or greater gains or losses in chromosome/chromosome arm
• Believed to be associated with poor prognosis
• However may mask other poor prognostic factors
• No specific chromosome involvement
• Seen in ~6% of multiple myeloma patients

[INTERNAL DATA – MARCH, 2017]
MULTIPLE MYELOMA – CHROMOTHRIPSIS 1Q

[INTERNAL DATA – MARCH, 2017]
COPY-NEUTRAL LOSS OF HETEROZYGOSITY

• Seen in ~28% cases studied
• Exact significance – not always clear
• In some cases – poor prognostic factor
• As detailed yesterday – different from that seen in other hematological malignancies
  – More often whole chromosome involved
PATIENT WITH MM

- 67 year old female
- Chromosomes: No growth
- FISH – FGFR3/IGH rearrangement, MYC gain, CCND1 gain
- Array
  - Numerous abnormalities
  - Chromothripsis –3q and chromosome 8
  - Homozygous deletion – 11q
  - Complex clone

[INTERNAL DATA – MARCH, 2017]
CHROMOSOME 11 – NUMEROUS DELETIONS & HOMOZYGOUS DELETION – BIRC3

[INTERNAL DATA – MARCH, 2017]
SECOND DISEASE

• Some patients present with unclear findings and a diagnosis of myeloma/MDS
  – Can be studied with both positive and negative CD138 separated fractions

• Some cases two diseases detected
  – Myeloma and MDS
  – Myeloma and CLL

• Some case no myeloma – only MDS or CLL

• Some can be confusing if not studied separately

[INTERNAL DATA – MARCH, 2017]
SECOND DISEASE

• Positive
  – Multiple abnormalities consistent with more aggressive disease
    • MYC gains
    • CN-LOH 17p

• Negative
  – 7q deletion
  – 20q deletion
  – Consistent with MDS

• But what if not separated?
KARYOVIEW – NEGATIVE FRACTION

[INTERNAL DATA – MARCH, 2017]
BENEFITS OF ARRAY

• Advantages
  – Detection of abnormalities not available by FISH and not seen in chromosome analysis
    • Copy-neutral loss of heterozygosity
    • Chromothripsis
  – Ability to provide better prognostic information
  – Provides a mechanistic understanding of the formation of various abnormalities and underlying importance
CHALLENGES OF ARRAY

• Challenges
  – Cannot pick up balanced translocations
    • Important in MM testing
    • FISH needs to be done in conjunction
  – Sensitivity of the array
    • Have been able to detect 7-15% mosaicism
    • Dependent on size, location, type (del/dup)
    • Measure – using smooth signal (determine %)
  – Dependent on the enrichment
    • Percentage of plasma cells in patient
    • Age of the sample at time of enrichment
  – Detection of second disease
    • Can two diseases confuse diagnosis

[INTERNAL DATA – MARCH, 2017]
PATIENTS WITH MM - APPROACH

• Should consider array as 1\textsuperscript{st} tier test in conjunction with MM translocation
  – Use on enriched cells
  – Reduce large number of FISH probes currently used
  – Must do translocations probes – but other probes can be replace more effectively by array analysis

[INTERNAL DATA – MARCH, 2017]
ACKNOWLEDGEMENTS

LabCorp – Directors/Counselors
- Peter Papenhausen
- Stuart Schwartz
- Jim Tepperberg
- Inder Gadi
- Rachel Burnside
- Karen Phillips
- Hiba Risheg
- Rao Potluri
- Katie Rudd
- Justin Schleede
- Romela Pasion
- Huong Cabral
- Jennifer Shafer
- Laura Kline
- Margriet Johansen
- Sharon Molinari
- Michelle Pierce

LabCorp – Array Lab
- Carolyn Bullen
- Paul Colacicco
- Shorne Cox
- Jeanne Emery
- Zach Gillespie
- Bonnie Haines
- Joeven Nonato
- Nathan Raynor
- Zachary Riscica
- Keli Rodriguez
- Savanna Schepis
- Jesse Soileau
- Jessica Whaley-Davis
- Brian Williford
- Danielle Wright

LabCorp – FISH Lab
- Holly Goode
- Barbara Purvis
- Edison Chua
- Tracy Hummel