ACUTE MYELOID LEUKEMIA, WITHOUT MENTION OF HAVING ACHIEVED REMISSION; POSSIBLE RELAPSE

**Morphology**

- 10% MYELOID BLASTS; COMPATIBLE WITH RELAPSED ACUTE MYELOID LEUKEMIA
- INCREASED, LEFT-SHIFTED ERYTHROID PRECURSORS AND GRANULOCYTES
- MILD ERYTHROID AND MEGAKARYOCYTIC CHANGES

**Flow Cytometry**

- 13% ABNORMAL MYELOID BLASTS

**Cytogenetics**

46,XY,t(1;15)(p36.3;q11.2)[9]/46,XY,t(4;15)(q11.2;q25)[3]/46,XY,add(15)(q13)[3]/46,XY[3]

ABNORMAL KARYOTYPE WITH THREE CLONES HAVING UNRELATED REARRANGEMENTS OF CHROMOSOME 15 IN 17 OF 20 CELLS

**FISH**

- AML PANEL: NORMAL
- MDS PANEL: NORMAL

**Milderythroid andMegakaryocytic Changes**

Comments: Flow cytometry detected 13% myeloblasts without features of AML-M3. Morphologic evaluation reveals a hypercellular marrow for age (70-75% cellularity) with erythroid and megakaryocytic changes (atypical feature of 1 on a scale of 0 to 4), left shift of erythroid precursors and granulocytes and increased iron stores (4 on a scale of 0 to 4). CD34 immunostains shows 10% blasts in small clusters and single form. CD117 shows moderately increased immature myeloid cells. E-cadherin stain shows increased immature erythroid precursors. Conventional cytogenetics showed an abnormal karyotype. AML/MDS FISH panel did not detect any abnormality. Overall morphologic and immunophenotypic findings are compatible with Acute Myeloid Leukemia in relapse. The increase in erythroid precursors raises the possibility of Acute Erythroid Leukemia. However, complete diagnostic records are not available for comparison at this time. Preliminary findings discussed with the doctor.

**Flow Cytometry Scatter Plot**

**Hypercellular Marrow for Age**
Flow Cytometry Report

Patient Information

Patient Name: [Redacted]
Sex: Male
DOB: 01/06/1957 (57)
Specimen: Bone Marrow
Site: Iliac
Procedure:

Referring Physician

Physician:
Accession #: [Redacted]
Case #: [Redacted]
ID#: [Redacted]
Collected: 08/15/2014
Received: 08/16/2014
Reported: 08/19/2014

Specimen Information

Clinical Data

(205.00) Acute myeloid leukemia, without mention of having achieved remission; Possible relapse

Interpretation

- 13% ABNORMAL MYELOID BLASTS

Comment:
Flow cytometry detected 13% abnormal myeloid blasts. Flow cytometric findings are compatible with acute myelogenous leukemia in relapse. Morphologic evaluation and conventional cytogenetics are in process. Flow cytometric findings were discussed with the doctor.

Results

Populations Identified
Abnormal myeloid blast population identified, of variable cell size, comprising 13% of events analyzed, with the following antigenic profile:
POSITIVE FOR: CD7 (variable), CD11b (variable), CD13, CD33, CD34, CD38, CD45 (dim), CD117 (variable), intracellular MPO, HLA-DR
NEGATIVE FOR: CD2, CD3, CD4, CD5, CD8, CD10, CD14, CD16, CD19, CD20, CD36, CD56, CD64, CD79a, intracellular TDT, surface kappa, surface lambda

B-Cells: <1% surface-kappa:lambda ratio: 1:1 (too few events to reliably compute)
Hematogones: 0%
T-Cells: 5% CD4:CD8 ratio: 1.1:1 (consistent with benign T-Cell population)
NK-Cells: 1%
Plasma cells: <1%
Granulocytes: 70%
Monocytes: 3%

The remaining events consist of debris and non-staining forms.

Evaluated Markers
CD2, CD3, CD4, CD5, CD7, CD8, CD10, CD11b, CD13, CD14, CD16, CD19, CD20, CD33, CD34, CD36, CD38, CD45, CD56, CD64, CD117, HLA-DR, surface kappa (p), surface lambda (p), intracellular CD3, intracellular CD22, intracellular CD34, extracellular CD79a, intracellular MPO, intracellular TDT, PI (Total markers: 31)

Electronic Signature

Jacqueline O’Hare, DO
CPT Code(s): 88184, 88185(x30), 88189

The technical component was performed at Applied Diagnostics and the professional component was performed by the signer of this report.

This test was developed and its performance characteristics determined by Applied Diagnostics. It has not been cleared or approved by the U.S. Food and Drug Administration. The FDA has determined that such clearance is not necessary. This test is used for clinical purposes, it should not be regarded as investigational or for research. This laboratory is certified under the Clinical Laboratory Improvement Amendments of 1988 (CLIA-88) as qualified to perform high complexity clinical testing.
Clinical Data
(205.00) Acute myeloid leukemia, without mention of having achieved remission; Possible relapse

Interpretation

- 10% MYELOID BLASTS; COMPATIBLE WITH RELAPSED ACUTE MYELOID LEUKEMIA
- INCREASED, LEFT-SHIFTED ERYTHROID PRECURSORS AND GRANULOCYTES
- MILD ERYTHROID AND MEGAKARYOCYTIC CHANGES

Comments
Flow cytometry detected 13% myeloblasts without features of AML-M3. Morphologic evaluation reveals a hypercellular marrow for age (70-75% cellularity) with erythroid and megakaryocytic changes (atypical feature of 1 on a scale of 0 to 4), left shift of erythroid precursors and granulocytes and increased iron stores (4 on a scale of 0 to 4). CD34 immunostains shows 10% blasts in small clusters and single form. CD117 shows moderately increased immature myeloid cells. E-cadherin stain shows increased immature erythroid precursors. Overall morphologic and immunophenotypic findings are compatible with Acute Myeloid Leukemia in relapse. The increase in erythroid precursors raises the possibility of Acute Erythroid Leukemia. However, complete diagnostic records are not available for comparison at this time. Conventional cytogenetics and MDS/AML FISH panel are in process. Preliminary findings discussed with the doctor.

Stains

Immunohistochemistry
On core biopsy and clot section:
CD34 reveals 10% blasts (increased), occasionally forming small clusters.
CD117 shows moderately increased immature myeloid cells.
E-cadherin shows markedly increased immature erythroids.

Iron Stain
On core biopsy, clot section and aspirate smear:
Iron stores: 4 (scale of 0 to 4)
Ring sideroblasts: Absent

Gross Description
- 1 bony core + scanty clot, totaling 0.6 x 0.2 cm, lightly decalcified, all as A1
- 0.8 mL of dark red clot, all as A2
- 10 aspirate smears
- 2 sodium heparin tubes and 1 EDTA tube labeled as bone marrow aspirate

### Microscopic Description

**PERIPHERAL BLOOD SMEAR**

No peripheral blood smear received.

**Accompanying CBC report (08/15/2014):**

- WBC: 1.9 K/micL, Gran: 0.4 K/micL, Lymph: 1.2 K/micL, Mid: 0.3 K/micL, Hgb: 10.1 g/dL, MCV: 118.9 fL, RDW: 13.5%.
- Plt: 39 K/micL

**BONE MARROW**

**Adequacy and general findings**

- Core biopsy: Adequate
- Clot section: Adequate
- Aspirate smears: Adequate
- Cellularity: 70-75%, hypercellular for age
- Bone abnormality: Absent

**Hematopoietic elements**

- Myeloid: Mildly increased in number with left shift
- Erythroid: Increased in number with irregular nuclear contours, nuclear blebs and left shift
- Megakaryocytes: Decreased in number with occasional atypical nuclear lobulation

**Infiltrates**

- Blasts: Increased blasts (10% by immunostain) of medium to large cell size with slightly convoluted nuclei, dispersed chromatin pattern, one or a few nucleoli and minimal amount of cytoplasm, without Auer rods
- Monocytic cells: Not increased
- Plasma cells: Not increased
- Lymphoid aggregates: Absent
- Granulomas: Absent
- Metastases: Absent
- Fibrosis: Not identified on routine H&E stain
- Amorphous materials such as amyloid: Not identified on routine H&E stain

**Percentages of 200 cells counted (Reference ranges are in parentheses):**

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<tbody>
<tr>
<td>Neutrophils/Bands: 10 (25-40)</td>
<td>Eosinophils: 2 (1-3)</td>
<td>Basophils: 2 (0-1)</td>
<td>Lymphocytes: 10 (10-15)</td>
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<tr>
<td>Plasma Cells: 0 (0-1)</td>
<td>Monocytes: 1 (0-1)</td>
<td>Pronormoblasts: 28 (0-2)</td>
<td>Erythroblasts: 12 (15-25)</td>
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</table>

M:E ratio: 1.2:1 (1.5:1 - 4:1)

**Electronic Signature**

Annisa Lewis, MD

CPT Code(s): 85097,88305(x2),88311,88313(x3),G0461(x2),G0462(x2)

The Technical and Professional components were performed at Applied Diagnostics - Main Lab, 1140 Business Center Dr., Suite 370, Houston, TX 77043.
Patient Information

Patient Name: [Redacted]
Sex: Male
DOB: 01/06/1957 (57)
Specimen: Bone Marrow
Site: Iliac
Procedure: [Redacted]

Referring Physician

Physician: [Redacted]

Specimen Information

Accession #: [Redacted]
Case #: [Redacted]
ID#: [Redacted]
Collected: 08/15/2014
Received: 08/16/2014
Reported: 08/19/2014

Clinical Data

(205.00) Acute myeloid leukemia, without mention of having achieved remission; Possible relapse

Interpretation

AML PANEL: NORMAL

Comment:

\[ \text{nuc ish(RUNX1T1, RUNX1)x2[197],(KMT2A)x2[188],(PML,RARA)x2[191],(MYH11,CBFB)x2[188]} \]

A total of 200 interphase nuclei were scored for each acute myelogenous leukemia (AML) probe set on a direct harvest of bone marrow cells and revealed a normal result.

Analyzed Probes:

- RUNX1T1 (8q21.3) and RUNX1 (21q22.12) dual fusion probe set to detect t(8;21);
- KMT2A (11q23) break apart probe set to detect rearrangements, deletions and amplification of the MLL gene;
- PML (15q24.1) and RARA (17q21) dual fusion probe set to detect t(15;17);
- and CBFB (16q22) and MYH11 (16p13) dual fusion probe set to detect inv(16) and t(16;16).

(All probes CytoCell)

The scoring for this case was completed using a per cell nuclei approach to signal counting and was performed manually by a licensed technologist. Interphase FISH is performed to screen for the loci indicated above and does not detect other chromosomal abnormalities.

Electronic Signature

Annisa Lewis, MD
CPT Code(s): 88368(x8)

The Technical and Professional components were performed at Applied Diagnostics - Main Lab, 1140 Business Center Dr., Suite 370, Houston, TX 77043.
Patient Information

Patient Name: [Redacted]
Sex: Male
DOB: 01/06/1957 (57)
Specimen: Bone Marrow
Site: Iliac
Procedure:

Referring Physician

Physician:

Specimen Information

Accession #:
Case #:
ID#:
Collected: 08/15/2014
Received: 08/16/2014
Reported: 08/19/2014

Clinical Data

(205.00) Acute myeloid leukemia, without mention of having achieved remission; Possible relapse

Interpretation

MDS PANEL: NORMAL

Comment:
nuc ish [D5S630/DSS2064,EGR1)x2[199],(D7Z1x2)[199],(D7S796,D7S486)x2[200],(D8Z2x2)[198],
(D20S108,MYBL2)x2[197]

A total of 200 interphase nuclei were scored for each myelodysplastic syndrome (MDS) probe set on a
direct harvest of bone marrow cells and revealed a normal result.

Analyzed Probes:
D5S630/DSS2064 (5p15.31) and EGR1 (5q31.2) probe set to detect deletions of EGR1 and monosomy of chromosome 5;
D7S796 (7q22) and D7S486 (7q31) probe set to detect deletions of the long arm of chromosome 7 (7q); D7Z1 (7p11.1-7q11.1) to detect monosomy of chromosome 7; D8Z2 (8p11.1-8q11.1) to detect polysomy of chromosome 8; and 20S108 (20q12) and MYBL2 (20q13.12) probe set to detect deletions of the long arm of chromosome 20 (20q). (All probes
CytoCell)

The scoring for this case was completed using a per cell nuclei approach to signal counting and was performed manually by a licensed
technologist. Interphase FISH is performed to screen for the loci indicated above and does not detect other chromosomal abnormalities.

Electronic Signature

Annisa Lewis, MD
CPT Code(s): 88368(x8)

The Technical and Professional components were performed at Applied Diagnostics - Main Lab,
1140 Business Center Dr., Suite 370, Houston, TX 77043.

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Cytogenetics Report

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Clinical Data
(205.00) Acute myeloid leukemia, without mention of having achieved remission; Possible relapse

Interpretation
ABNORMAL KARYOTYPE WITH THREE CLONES HAVING UNRELATED REARRANGEMENTS OF CHROMOSOME 15 IN 17 OF 20 CELLS

KARYOTYPE: 46,XY,t(1;15)(p36.3;q11.2)[9]/46,XY,t(4;15)(q11.2;q25)[3]/46,XY,add(15)(q13)[3]/46,XY[3]

Chromosome analysis was performed on G-banded metaphase spreads prepared from unstimulated 24 hour and 72 hour cultures. Three abnormal clones were identified. The mainline consisted of nine cells with a translocation between 1p36.3 and 15q11.2. The first subclone had a translocation between 4q11.2 and 15q25. The second subclone of 3 cells had extra material of unknown origin at 15q13. Three cells were interpreted as normal.

Metaphases Counted: 20  Metaphases Analyzed: 20  Metaphases Karyotyped: 6
Culture Type: 48HR, 72HR  Banding Technique: GTG  Banding Resolution: 425

Electronic Signature
Katy Phelan, PhD
CPT Code(s): 88237(x2),88264,88280(x2),88291
The Technical and Professional components were performed at Applied Diagnostics - Main Lab, 1140 Business Center Dr., Suite 370, Houston, TX 77043.
This test was developed and its performance characteristics determined by Applied Diagnostics. It has not been cleared or approved by the U.S. Food and Drug Administration. The FDA has determined that such clearance is not necessary. This test is used for clinical purposes. It should not be regarded as investigational or for research. This laboratory is certified under the Clinical Laboratory Improvement Amendments of 1988 (CLIA-88) as qualified to perform high complexity clinical testing.