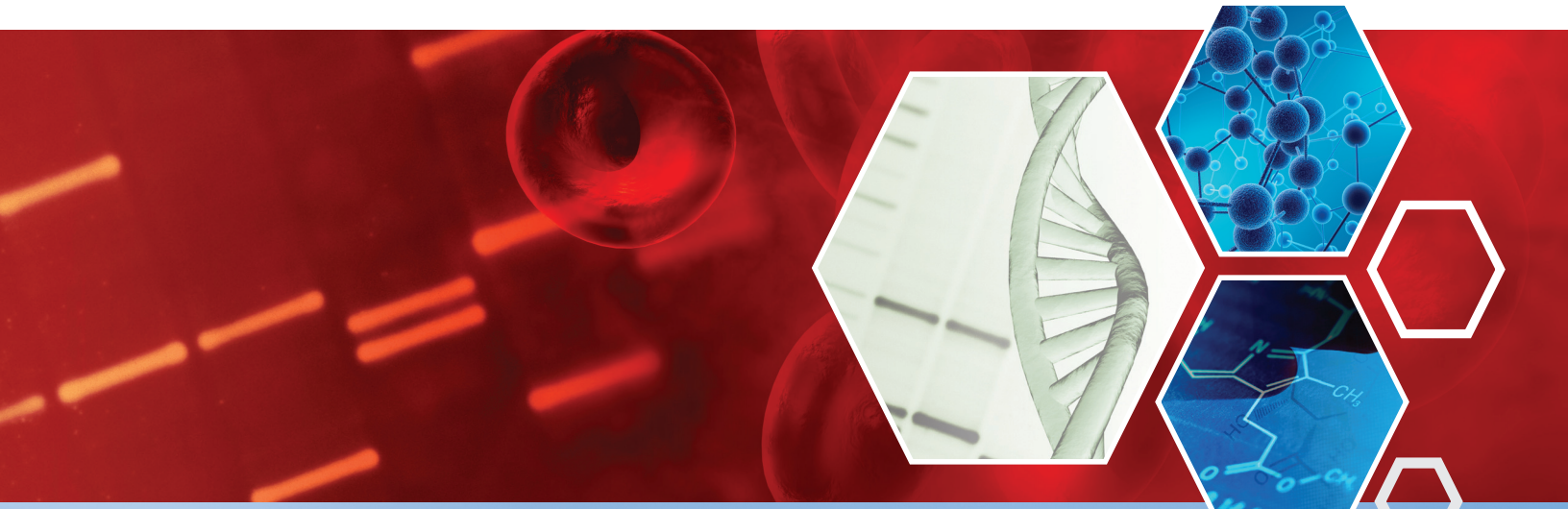
med fusion
clin-labs • clin-trials
www.medfusionservices.com

Hematopathology Case Study



Clinical Presentation:

Clinical Information: A 42 year old male with history of chronic myelogenous leukemia (CML) presents with an elevated white blood cell count. After physician consultation with patient, it was determined that the patient had not been compliant with Gleevec treatment.

Evaluation of the peripheral blood smear revealed a mild neutrophilic leukocytosis with left shift to the myelocyte stage as well as basophilia.

Examination of the marrow aspirate smears revealed trilineage hematopoiesis without significant dyspoiesis. There was myeloid hyperplasia with a myeloid to erythroid ratio of 9.6:1. The core biopsy revealed a hypercellular marrow (80%) with frequent small megakaryocytes.

Flow cytometric analysis demonstrated increased CD56 positive granulocytes, with no definitive immunophenotypic evidence of high-grade hematopoietic neoplasia, lymphoproliferative disease or plasma cell dyscrasia. There was no increase in myeloblasts.

These initial findings are consistent with the history of CML in chronic phase in this patient who has been noncompliant with therapy.

Chromosome analysis showed two related abnormal cell lines. The stemline contained trisomy 8 and the Philadelphia chromosome [t(9;22)]. An extra copy of derivative chromosome 22 was also observed in the sideline.

Fluorescence in situ hybridization (FISH) analysis also showed two abnormal signal patterns. The first represents the stemline with a deletion of BCR/ABL1 fusion gene on the der(9) and the 'typical' signal pattern represents the sideline in which the extra copy of Philadelphia chromosome [der(22)] gives the second fusion signal.

BCR and/or ABL1 deletion on the der(9) is a recurrent cytogenetic abnormality in CML, and patients with BCR/ABL1 deletion show significantly shorter overall survival (OS) and event-free survival (EFS), compared to patients without BCR or ABL1 gene deletions.

Diagnosis and Interpretation:

FINDINGS CONSISTENT WITH CHRONIC MYELOGENOUS LEUKEMIA, BCR-ABL POSITIVE; POSITIVE FOR CLONAL EVOLUTION.

In Conclusion:

The morphologic and flow cytometric findings are typical of CML. The disease is defined by the presence of the Philadelphia chromosome. Cytogenetic analysis on the patient revealed additional abnormalities.

The presence of clonal cytogenetic evolution occurring after the initial diagnostic karyotype is diagnostic of the accelerated phase of CML (disease progression). Drug therapy is effective in the chronic phase of CML; however, the emergence of sub-clones of leukemic progenitor cells can lead to drug resistance in patients in accelerated phase.

BCR/ABL1 FISH is positive and RT-PCR for BCR/ABL1 demonstrates an IS of 53.41% which is not indicative of major molecular response. Additionally, chromosomal analysis demonstrates Philadelphia chromosome (Ph+), deletion of BCR/ABL1 on derivative chromosome 9 and clonal evolution. Deletion of BCR and/or ABL1 gene on derivative chromosome 9 is associated with an unfavorable prognosis. These cytogenetic findings are worrisome for progression to the accelerated phase of CML. However, the original karyotype is not currently available for review. If in fact this is a new finding, this is most compatible with CML, accelerated phase. Please see full cytogenetics report for further details. The diagnoses are unchanged.

Patient Name:	TEST PATIENT			Accession #:	1234567
Med. Rec. #:	000000			Collected:	11/6/2013
DOB:	6/14/1971 (Age: 42)			Received:	11/6/2013
Gender:	M	Billing #: 0		Reported:	11/12/2013 12:06
Physician(s):	TEST PHYSICIAN	Copy To:			

HEMATOPATHOLOGY REPORT

DIAGNOSIS
Date Reported: 11/12/2013 12:06

BLOOD:
MILD NEUTROPHILIC LEUKOCYTOSIS WITH LEFT SHIFT (ANC = 6.7K/uL, ALC = 5.3K/uL, AMC = 1.3K/uL).
MODERATE THROMBOCYTOSIS (PLATELETS = 678K/uL).

CBC : WBC 14.8K/uL, RBC 4.84M/uL, Hgb 15.6g/dL, Hct 49.1%, MCV 101.4fL, MCH 32.3pg, MCHC 31.9g/dL, RDW 18.2%, Plts 678K/uL, MPV 7.9fL .
Manual diff(%): Segs+bands 45, Lymphs 36, Monos 9, Eos 3, Basos 3, Meta 3, Myelo 1.

BONE MARROW:
CONSISTENT WITH MYELOPROLIFERATIVE NEOPLASM, CHRONIC MYELOGENOUS LEUKEMIA (CML), BCR-ABL POSITIVE
HYPERCELLULAR MARROW FOR AGE (80%) WITH MYELOID HYPERPLASIA (M:E RATIO = 9.6:1)

Aspirate diff(%), 500 cells: Blasts <1, Promyelocytes <1, Myelocytes 10, Metamyelocytes 19, Segs+bands 51, Lymphs 4, Monos 2, Eos 2, Mast cells/basos 2, Plasma cells 1.
Myeloid total 86, RBC precursors 9, M:E ratio 9.6:1

COMMENT
Per discussion with Test Physician, the patient has a history of chronic myelogenous leukemia (CML) and has been noncompliant with Gleevec medication. The morphologic findings are compatible with CML, chronic phase as there are no morphologic findings which indicate progression.

Dr. Javed Gill has also reviewed this case and concurs. Test Physician was notified of the results on 11/12/13 at 10:15 AM

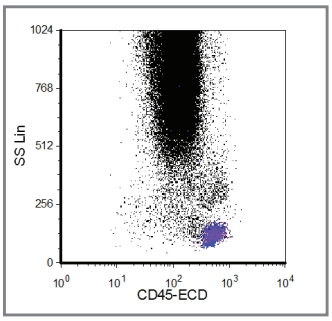
Cytogenetic analysis is pending; upon completion, a final comprehensive diagnosis will be issued.

Specimen: blood, marrow aspirate, biopsy, touch prep
Procedure: marrow aspiration and biopsy
Aspiration site: unspecified
Biopsy site: unspecified
Immunophenotype: flow cytometry performed; see body of the report

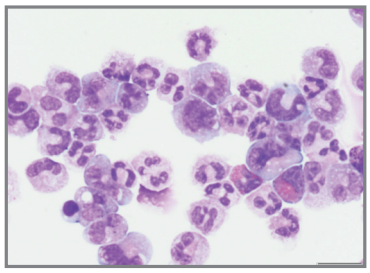
****Electronically Signed Out*** 11/7/2013 Latoya Keglovits, M.D.*

ANCILLARY STUDIES

Flow Cytometry - Leukemia/Lymphoma Profile:
Increased CD56 positive granulocytes. No definitive immunophenotypic evidence of high-grade hematopoietic neoplasia, lymphoproliferative disease or plasma cell dyscrasia.



CD45 vs SS



cs 50x.

*11/7/2013 11:49 ***Electronically Signed Out*** Latoya Keglovits, M.D.*

Technical services performed at: med fusion, 2501 South State Hwy 121, Suite 1100, Lewisville, TX 75067. Professional services performed at PBM Lewisville, 2501South State Hwy 121, Suite 1210, Lewisville, TX 75067.

BCR/ABL1 by Quant RT-PCR: Major(p210) and Minor(p190):
This specimen is positive for both the t(9;22) BCR/ABL1 Major (p210) and Minor (p190) fusion transcripts. The percent Major BCR/ABL1 transcript on the international scale (IS) is 53.41%. The percent Minor BCR/ABL1 transcript is 0.4172%. An IS of 100% Major transcript is designated as a universal baseline applicable to all patients. A 3 log decrease from this standardized baseline, or 0.1% IS, represents a Major Molecular Response (MMR). Achieving MMR by 18 months of treatment is associated with better outcome.

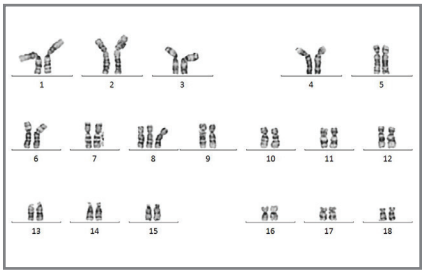
Current and Previous Test Results			
Accession #	**Collection Date**	**BCR/ABL1 Major (% IS)**	**BCR/ABL1 Minor (%)**
ABC (BM)	11/6/2013	53.41	0.4172

*11/14/2013 22:31 ***Electronically Signed Out*** Kristen J. Champion, Ph.D., FACMG*
Technical and Professional services performed at: med fusion, 2501 South State Hwy 121, Suite 1100, Lewisville, TX 75067

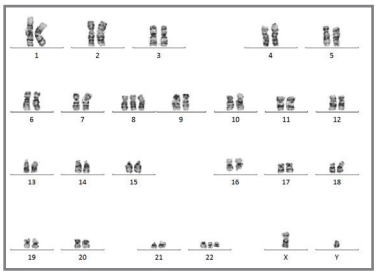
FISH Analysis:
Fluorescence in situ hybridization (FISH) was positive for the typical BCR/ABL1 gene rearrangement (2F1R1G) in 150 of the 200 interphase cells examined (75%). FISH was also positive for an atypical BCR/ABL1 gene rearrangement (1F1R1G) in 46 of 200 cells examined (23%). Chromosome analysis for this specimen was performed and will be reported upon completion.

In CML, BCR and/or ABL1 deletion on the der(9) is a recurrent cytogenetic abnormality, and patients with BCR/ABL1 deletion have significantly shorter overall survival (OS) and event-free survival (EFS), compared to the patients without any BCR or ABL gene deletions.

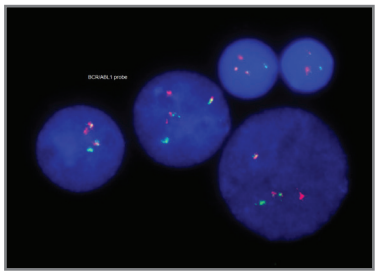
Cytogenetic Analysis:
Abnormal male bone marrow analysis with Philadelphia chromosome (Ph+), deletion of BCR/ABL1 genes on derivative chromosome 9 [der(9)], and clonal evolution. In chronic myelogenous leukemia (CML), deletion of BCR and/or ABL1 gene on the derivative chromosome 9 is associated with an unfavorable prognosis. Clonal evolution is usually associated with disease progression.



Stemline



Sideline



COMMENT
Chromosome analysis showed two related abnormal cell lines. The stemline contained trisomy 8 and t(9;22)(q34;q11.2) (Ph+). In sideline, an extra copy of derivative chromosome 22 [der(22)] was also observed. Fluorescence in situ hybridization (FISH) using dual color dual fusion probes for BCR and ABL1 gene loci also showed two abnormal FISH signal patterns (1F1R1G and 2F1R1G). The results of chromosome and FISH analyses indicate that the atypical FISH signal pattern (1F1R1G) represents the stemline with a deletion of BCR/ABL1 genes on the der(9) and the "typical" signal pattern represents the sideline in which the extra copy of der(22) gives the second fusion signal.

Reference: Lee et al., Deletion of any part of the BCR or ABL gene on the derivative chromosome 9 is a poor prognostic marker in chronic myelogenous leukemia, Cancer Genetics and Cytogenetics 166 (2006) 65-73

*11/12/2013 11:00 ***Electronically Signed Out*** Zhenjun Lou, Ph.D., DABMG and Thomas Lohmann, M.D*
Technical and Professional services performed at: med fusion, 2501 South State Hwy 121, Suite 1100, Lewisville, TX 75067

MORPHOLOGY REPORT

Clinical History
200.38; Patient with history of CML

Microscopic Description
Peripheral Smear:

WBC:	left shift to the myelocyte stage; morphologically mature granulocytes, lymphocytes and monocytes also observed; basophils increased in number
RBC:	mild anisopoikilocytosis, minimal polychromasia
Platelets:	estimate agrees with the reported count, granulation is normal, average size is not increased

Marrow aspirate smear/touch prep:

Particles:	cellular, well dispersed/touch prep is adequate
Myeloid Cells:	progressive maturation; mildly increased number of basophils observed
Erythroid precursors:	normoblastic maturation
Megakaryocytes:	present
Dyspoiesis:	insignificant
Lymphocytes:	morphologically mature
Plasma cells:	morphologically unremarkable

Marrow Core Biopsy:

Core Biopsy:	4mm with 1mm hematopoietic marrow, bone spicules well formed, periosteal soft tissue without infiltrate
Overall cellularity:	80%
Megakaryocytes:	adequate in number; frequent megakaryocytes are small with hypolobated nuclei
M/E ratio:	estimate agrees with aspirate differential count which shows myeloid hyperplasia
Granulomas:	absent
Lymphoid Aggregates:	not demonstrated
Infiltrates:	absent

Ancillary Stain(s):

Iron Stain:	performed on aspirate smear and biopsy
Storage iron:	adequate
Ring Sideroblasts:	none demonstrated
Reticulin:	within normal range (grade 0/3)
IHC:	performed on biopsy
CD3:	highlights scattered T-cells, <5% of the cellularity
CD20:	highlights scattered B-cells, <5% of the cellularity
Adequacy:	all controls show appropriate reactivity

Specimen(s) Received
1: Bone marrow Biopsy
2: Peripheral 4unst, Aspirate 4unst, Touch 2unst
3: EDTA Tube
4: EDTA Tube
5: NaHep Tube
6: NaHep Tube

Gross Description
Specimen #1 received in formalin labeled “PATIENT” and “bone marrow biopsy”, consists of a 0.6 cm in length x 0.2 cm in diameter single core of bony tissue, entirely submitted after decal.

Specimen #2 labeled “slides”, consists of 4 unstained peripheral slides, 4 unstained aspirate slides, 2 unstained touch preps. Also received are 2 purple-topped tubes and 2 green-topped tubes with blood.

11/7/2013
Unless otherwise stated and when applicable, the quality of the H&E and other stains is satisfactory. Technical services performed at PBM Lewisville, 2501 South State Hwy 121, Suite 1210, Lewisville, TX 75067 Professional services performed at PBM Lewisville, 2501 South State Hwy 121, Suite 1210, Lewisville, TX 75067.

FINAL COMPREHENSIVE DIAGNOSIS

FINDINGS CONSISTENT WITH CHRONIC MYELOGENOUS LEUKEMIA, BCR-ABL POSITIVE; POSITIVE FOR CLONAL EVOLUTION (SEE COMMENT).

COMMENT
BCR/ABL1 FISH is positive and RT-PCR for BCR/ABL1 demonstrates an IS of 53.41% which is not indicative of major molecular response. Additionally, chromosomal analysis demonstrates Philadelphia chromosome (Ph+), deletion of BCR/ABL1 on derivative chromosome 9 and clonal evolution. Deletion of BCR and/or ABL1 gene on derivative chromosome 9 is associated with an unfavorable prognosis. These cytogenetic findings are worrisome for progression to the accelerated phase of CML. However, the original karyotype is not currently available for review. If in fact this is a new finding, this is most compatible with CML, accelerated phase. Please see full cytogenetics report for further details. The diagnoses are unchanged.

****Electronically Signed Out*** Latoya Keglovits, M.D.*