Pancytopenia in a 38 year old male: ICUS to CCUS to MDS

SOCIETY FOR HEMATOPATHOLOGY CASE SH2017-0050

MARY ANN THOMPSON ARILDSEN, MD.PH.D.

VANDERBILT UNIVERSITY MEDICAL CENTER, NASHVILLE, TN

Clinical Presentation

38 year old male who was found to have leukopenia and thrombocytopenia when hospitalized for a snowboarding accident

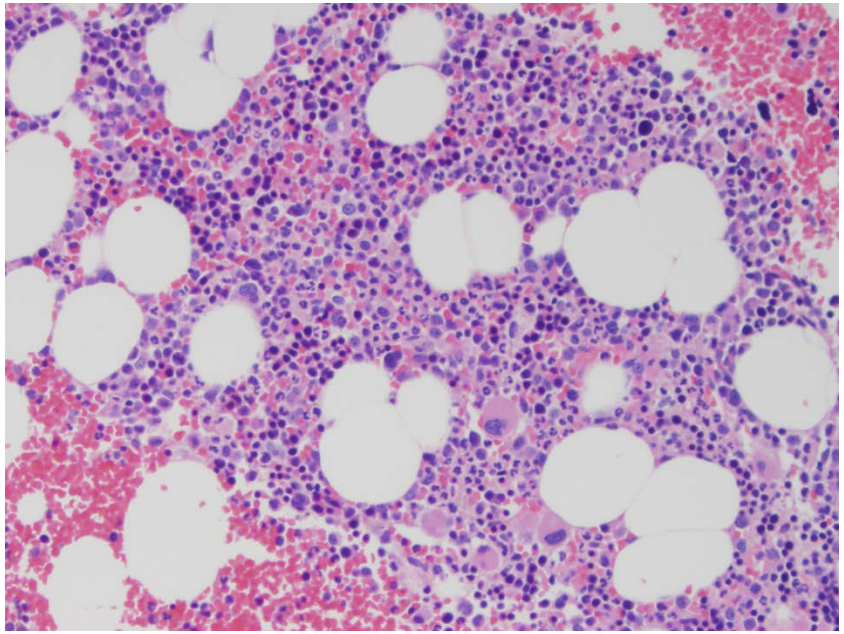
Follow-up CBC in November 2015 at hematologist's office:

- WBC 1.4 K/μl, Hct 39%, plts 92 K/μl ANC 280/μl, MCV 80.8 fL

Outside bone marrow biopsy dated 11/18/15: MODERATELY HYPERCELLULAR MARROW WITH MILD ERYTHROID AND MEGAKARYOCYTIC HYPERPLASIA

- Mild erythroid and megakaryocytic atypia
- Peripheral blood flow cytometry: no significant blast population and no monotypic B-cell or aberrant T cell population
- Karyotype: 46, XY

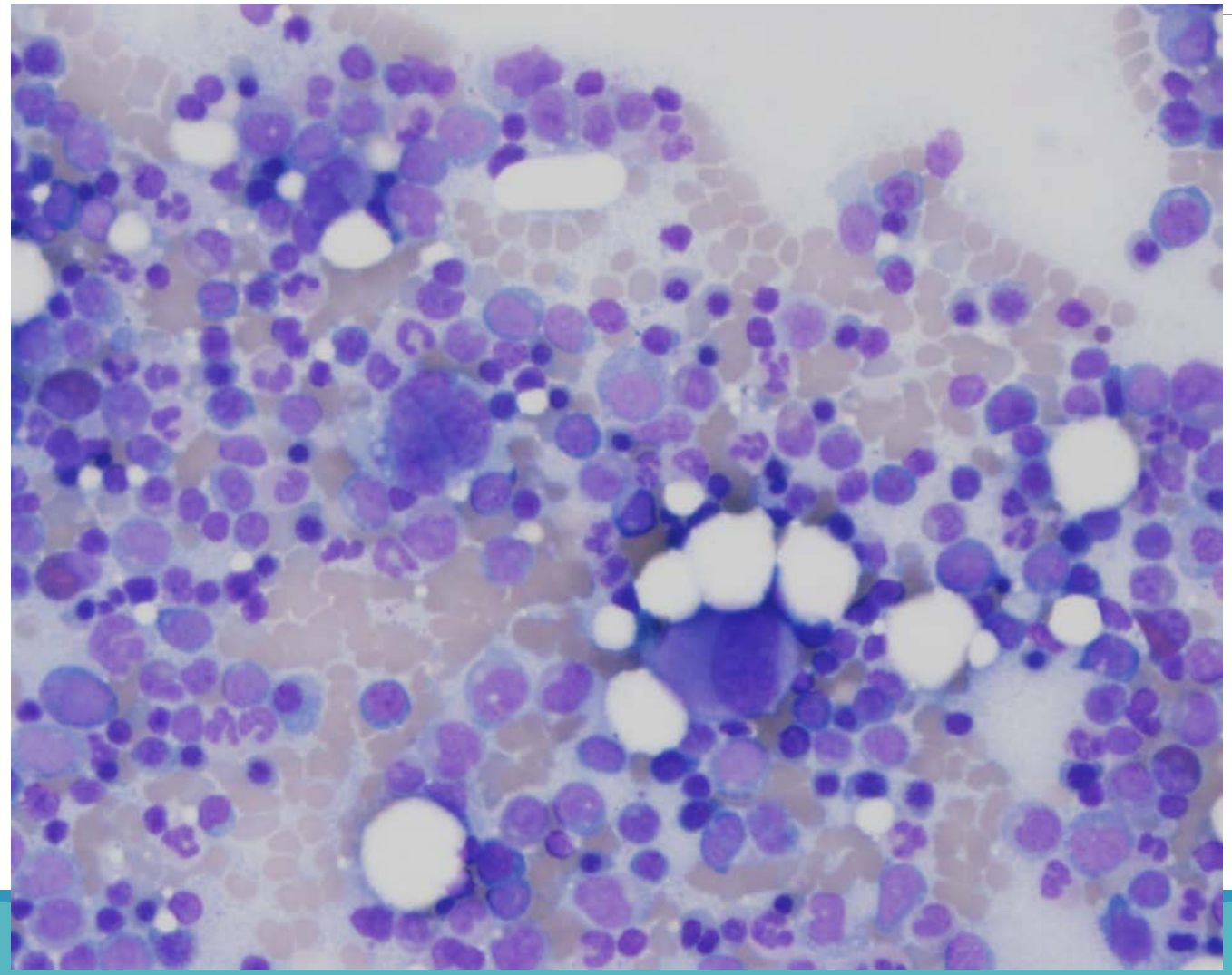
- ICUS: idiopathic cytopenia of uncertain significance

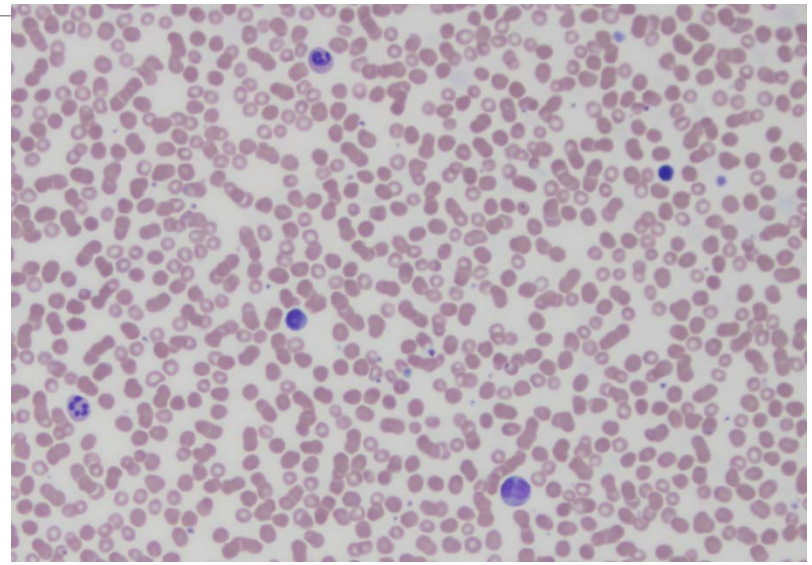


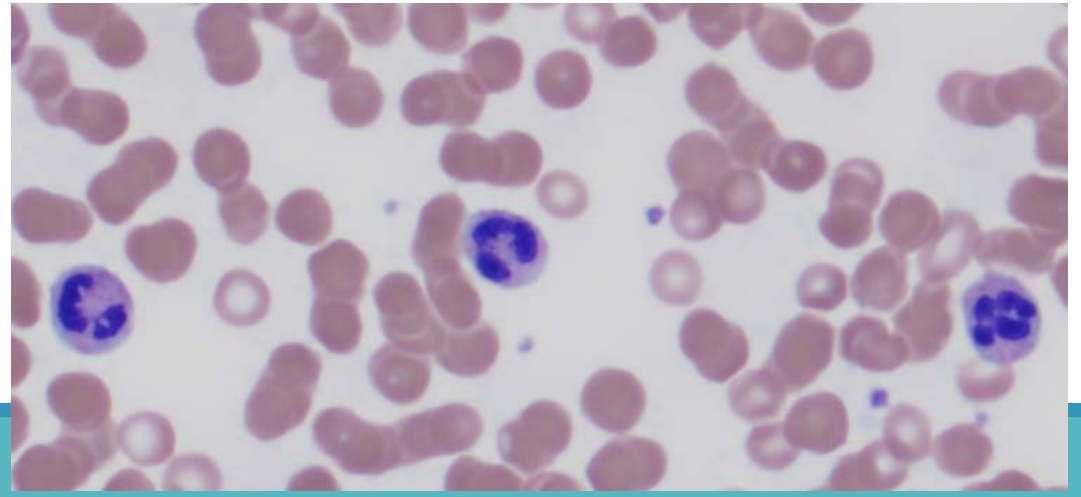
Clinical Presentation

- Patient was referred to VUMC for persistent pancytopenia (8/24/16, 10 mo. later)
  - No fatigue, night sweats, or weight loss
  - CBC: WBC 2.3 K/ul, Hct 40%, plts 104 K/ul
- Bone marrow diagnosis:
  - NORMOCELLULAR MARROW WITH TRILINEAGE HEMATOPOIESIS; MILD MEGAKARYOCYTIC DYSPLASIA, NO INCREASE IN BLASTS
  - Occasional hypolobated neutrophils in the peripheral blood
  - No increase in blasts by morphology, flow cytometry, or immunohistochemistry
  - Single interstitial lymphoid aggregate, mix of T and B cells by immunohistochemistry

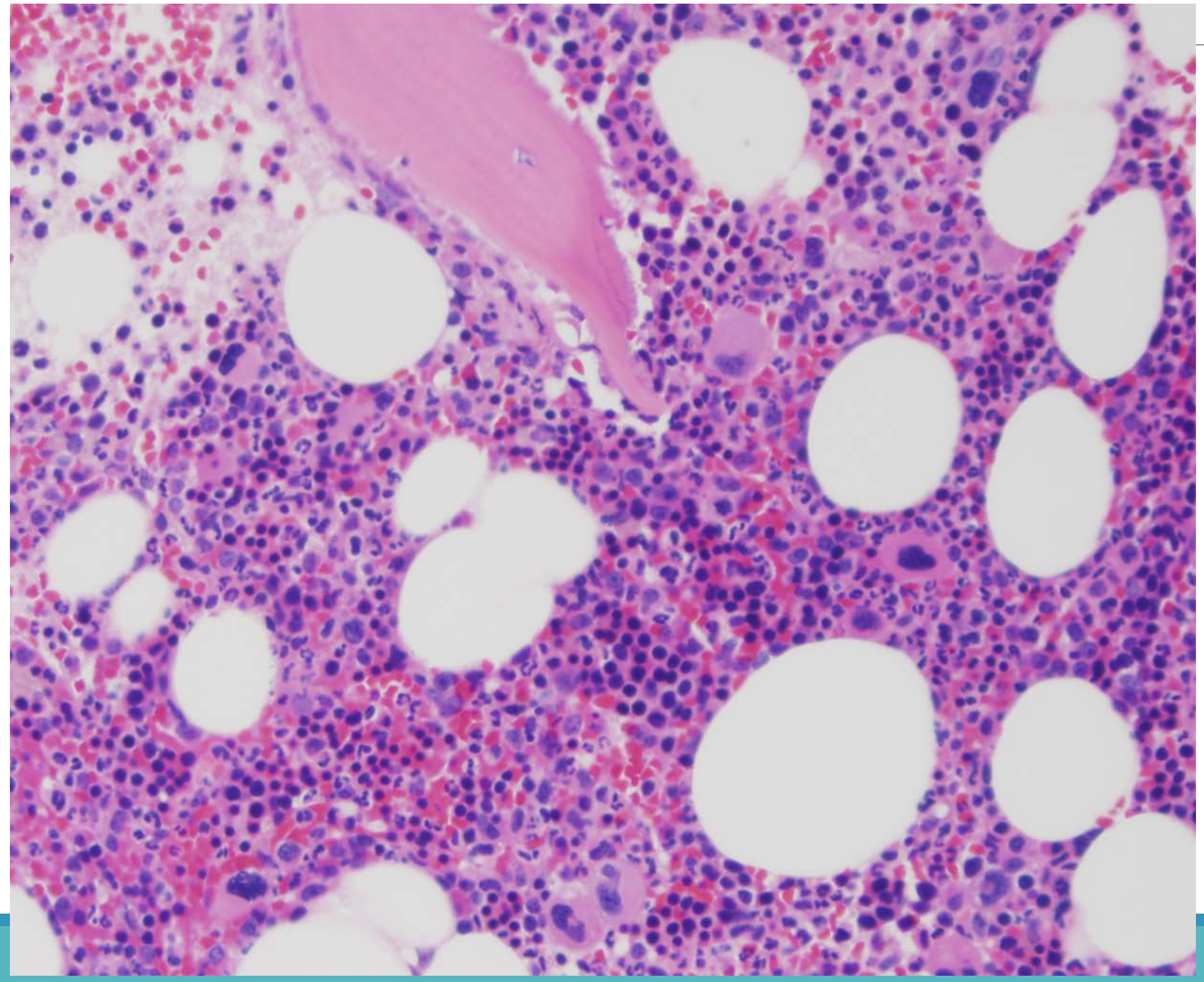
8/24/16 bone marrow aspirate and peripheral smear

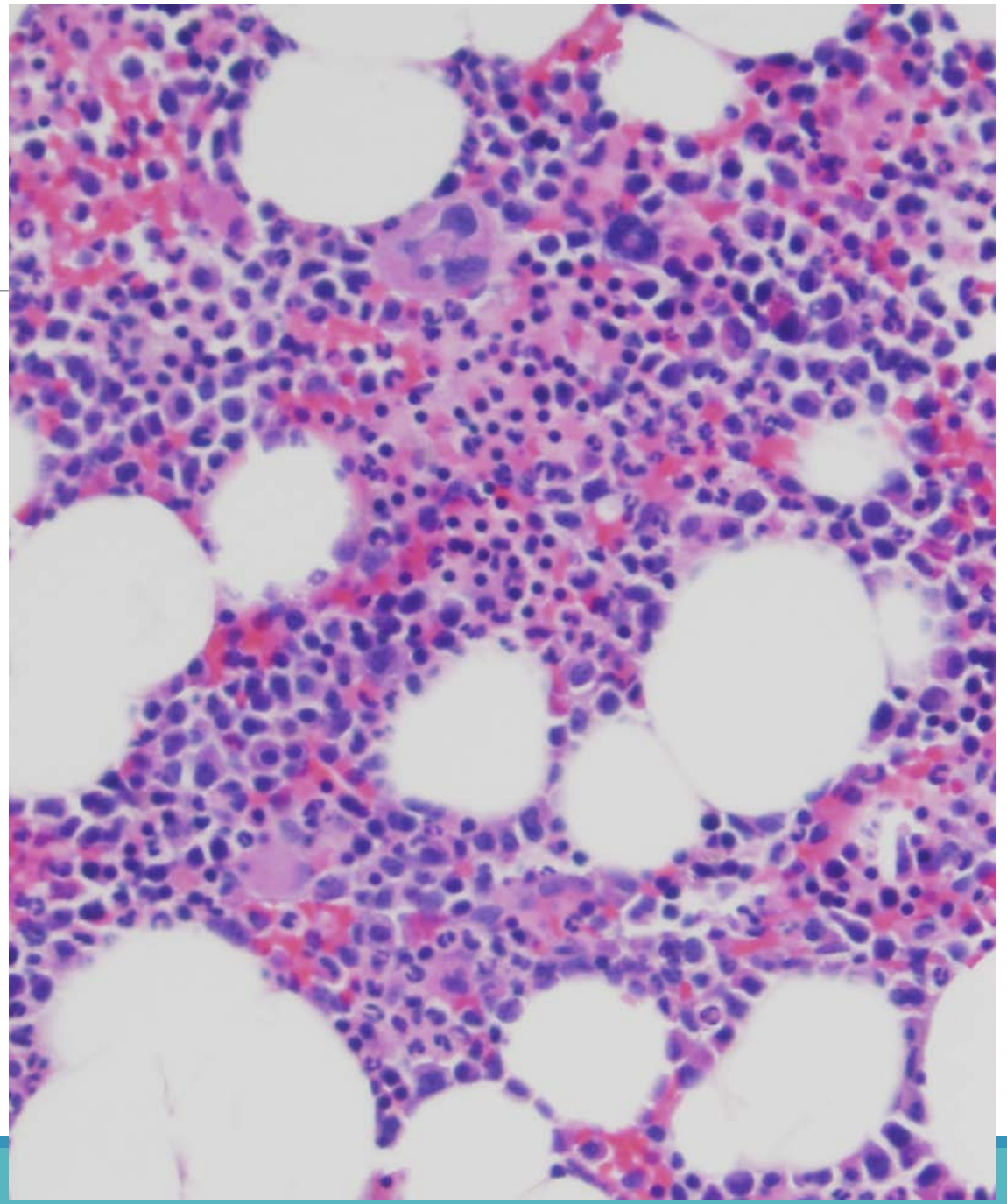






8/24/16 bone marrow biopsy and particle-H&E stain





Ancillary studies on 8/24/16 marrow

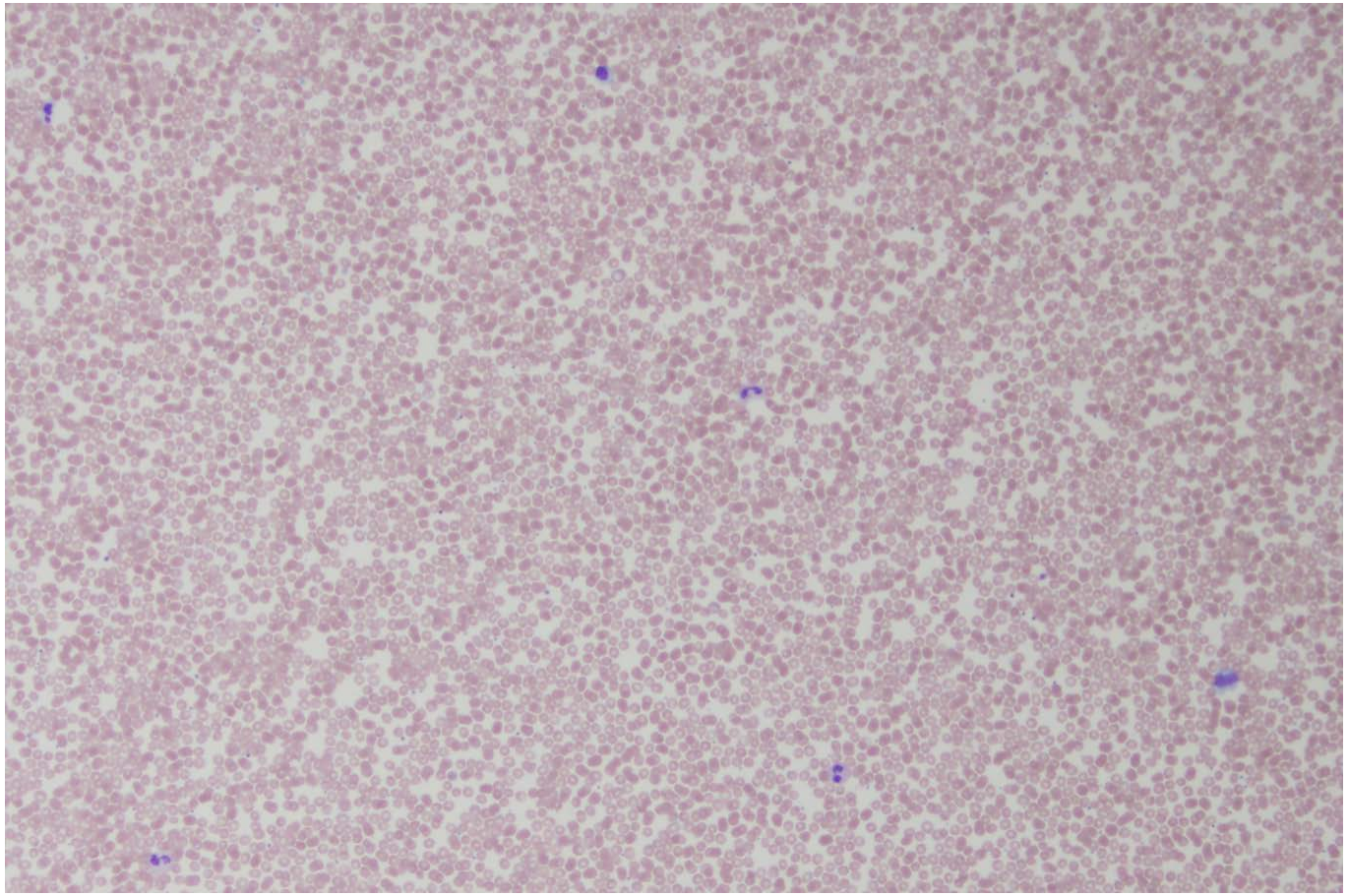
- Flow cytometry: 3.3% myeloblasts with a normal immunophenotype
- Karyotype: 46, XY
- FISH: Normal signal patterns using standard panel of MDS probes (5p15.2, 5q31, cen7, 7q31, cen8, 20q12)
- What next? The clinician called and said, “Look, I know this isn’t your usual protocol, but I would like to do NGS”.
- OnkoSight BRLI myeloid disease panel of 37 genes
  - SRSF2 p.Pro95Leu (43.15%)
  - RUNX1 p.Arg169Lysfs*44 (11.59%)
  - IDH1 p.Arg132Cys (42.26%)
  - NRAS p.Gly12Asp (11.87%)
  - BCOR p.Met1020Val—unclear variant (19.85%)

ICUS ➡ CCUS

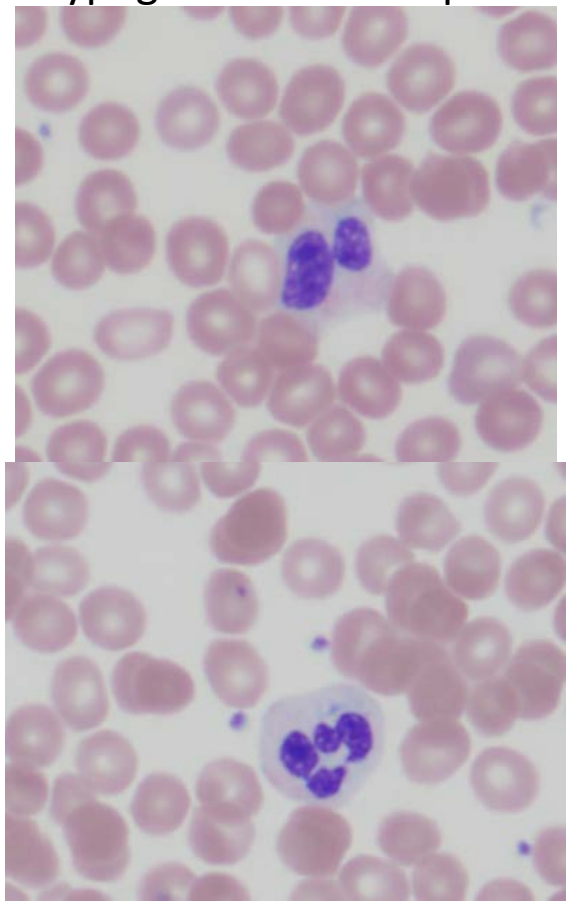
- BONE MARROW: NORMOCELLULAR MARROW WITH TRILINEAGE HEMATOPOIESIS; MILD MEGAKARYOCYTIC DYSPLASIA; NO INCREASE IN BLASTS; MULTIPLE MUTATIONS PRESENT CONSISTENT WITH CCUS

- CCUS: Clonal cytopenias of undetermined significance

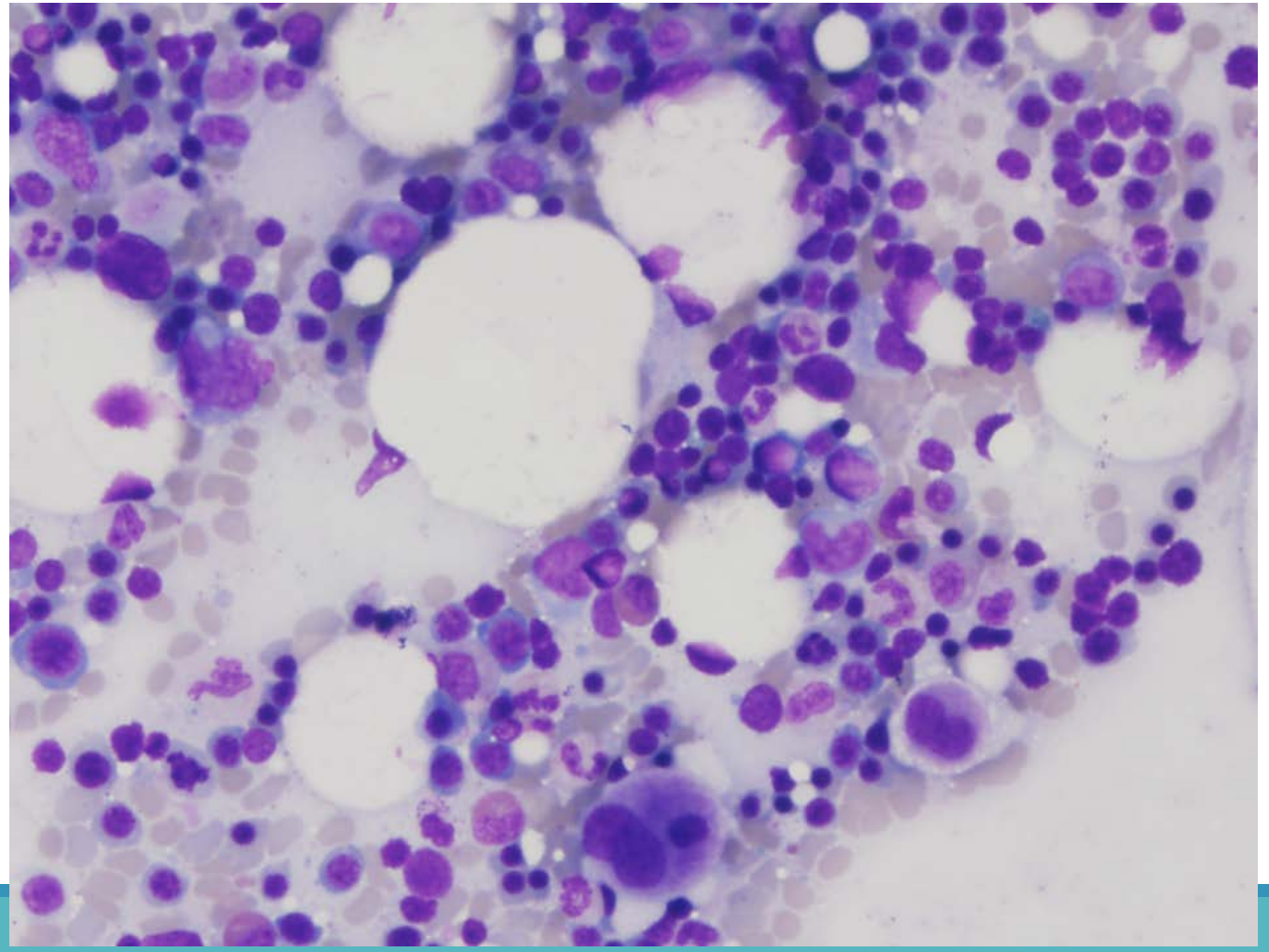
1/25/17 peripheral blood smear
CBC: WBC 0.9K/μl, Hct 38%, Plts 183 K/μl ANC 280/μl

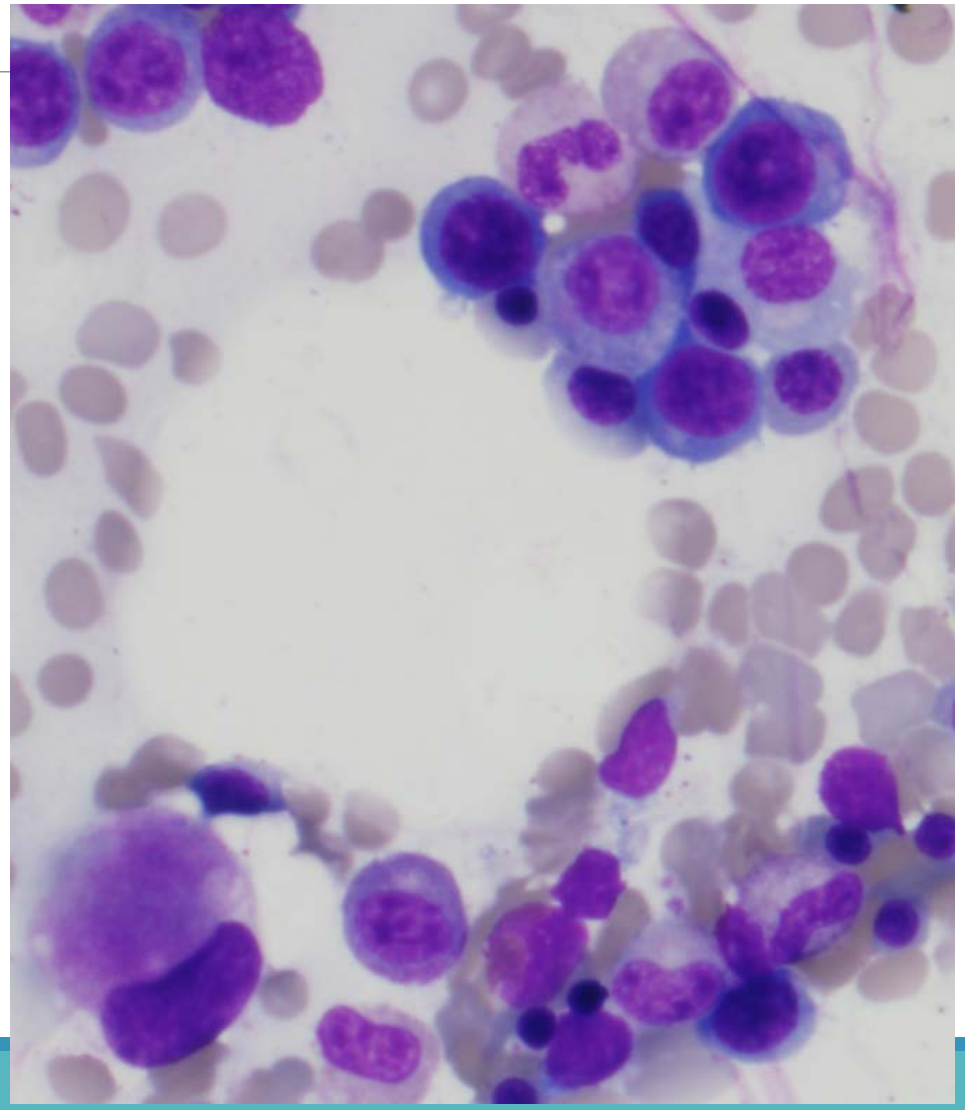


Hypogranular neutrophils

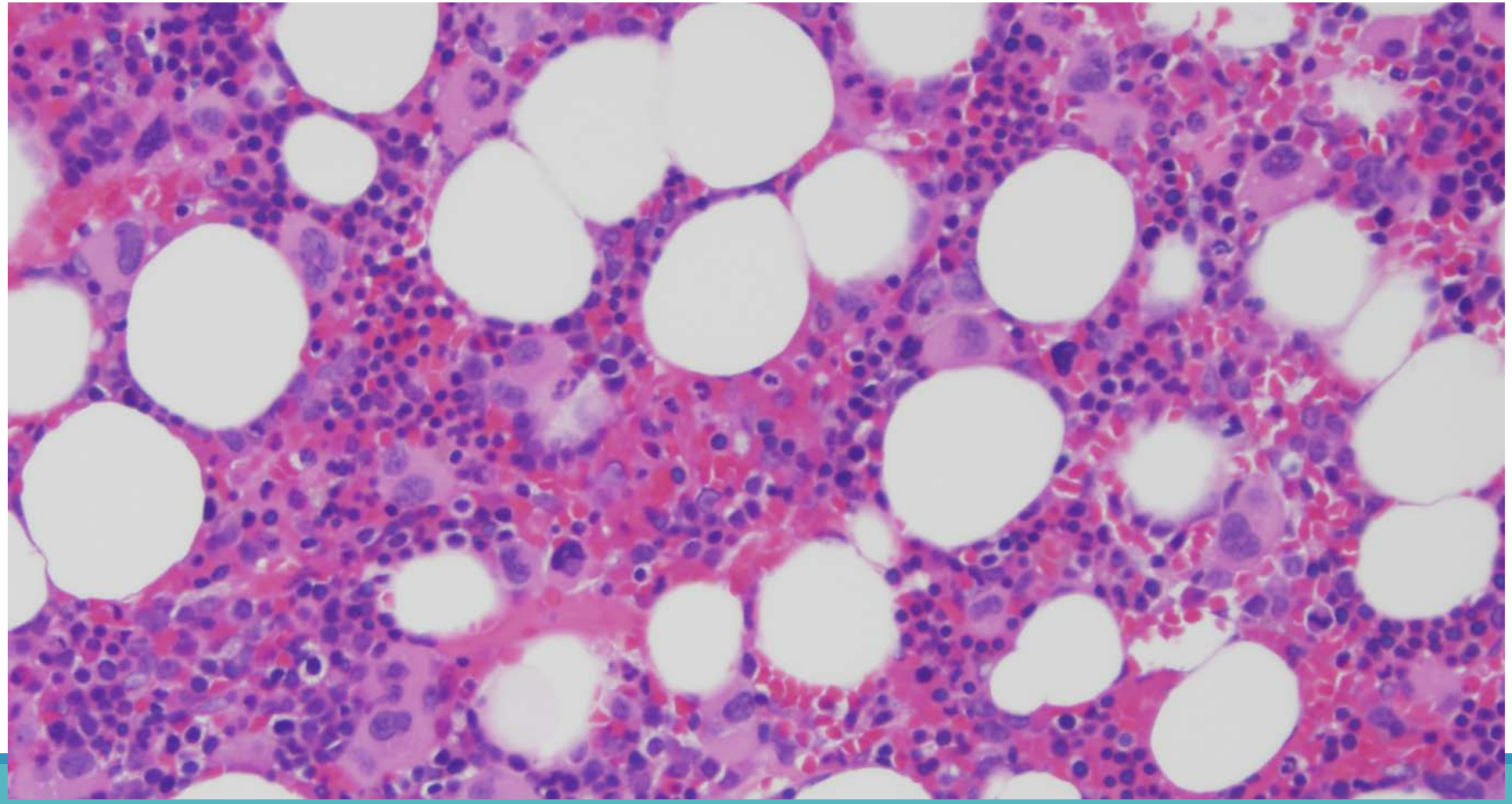


1/25/17 bone marrow aspirate





1/25/17 bone marrow biopsy

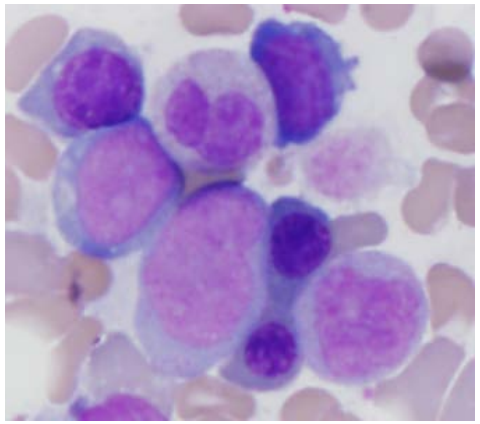


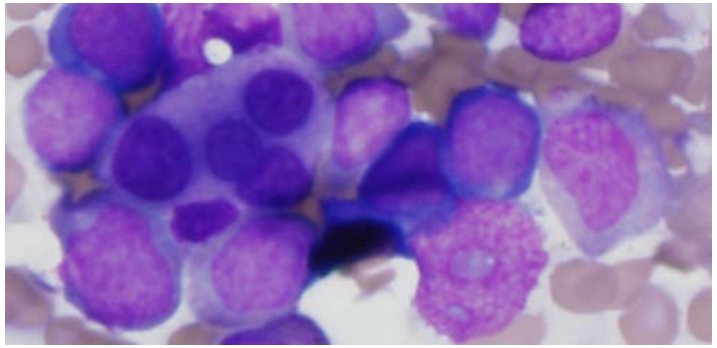
ICUS → CCUS → MDS

- Bone marrow on 1/25/17, interval of 5 months
  - CBC: WBC 0.9K/μl, Hct 38%, Plts 183 K/μl ANC 280/μl
  - BONE MARROW: NORMOCELLULAR MARROW WITH BILINEAGE DYSPLASIA; NO INCREASE IN BLASTS; CONSISTENT WITH MYELODYSPLASTIC SYNDROME
    - Best classified as MDS with multilineage dysplasia (MDS-ML)
    - Myeloid and megakaryocytic dysplasia
    - No increase in myeloblasts by flow cytometry
    - Normal karyotype, 46, XY

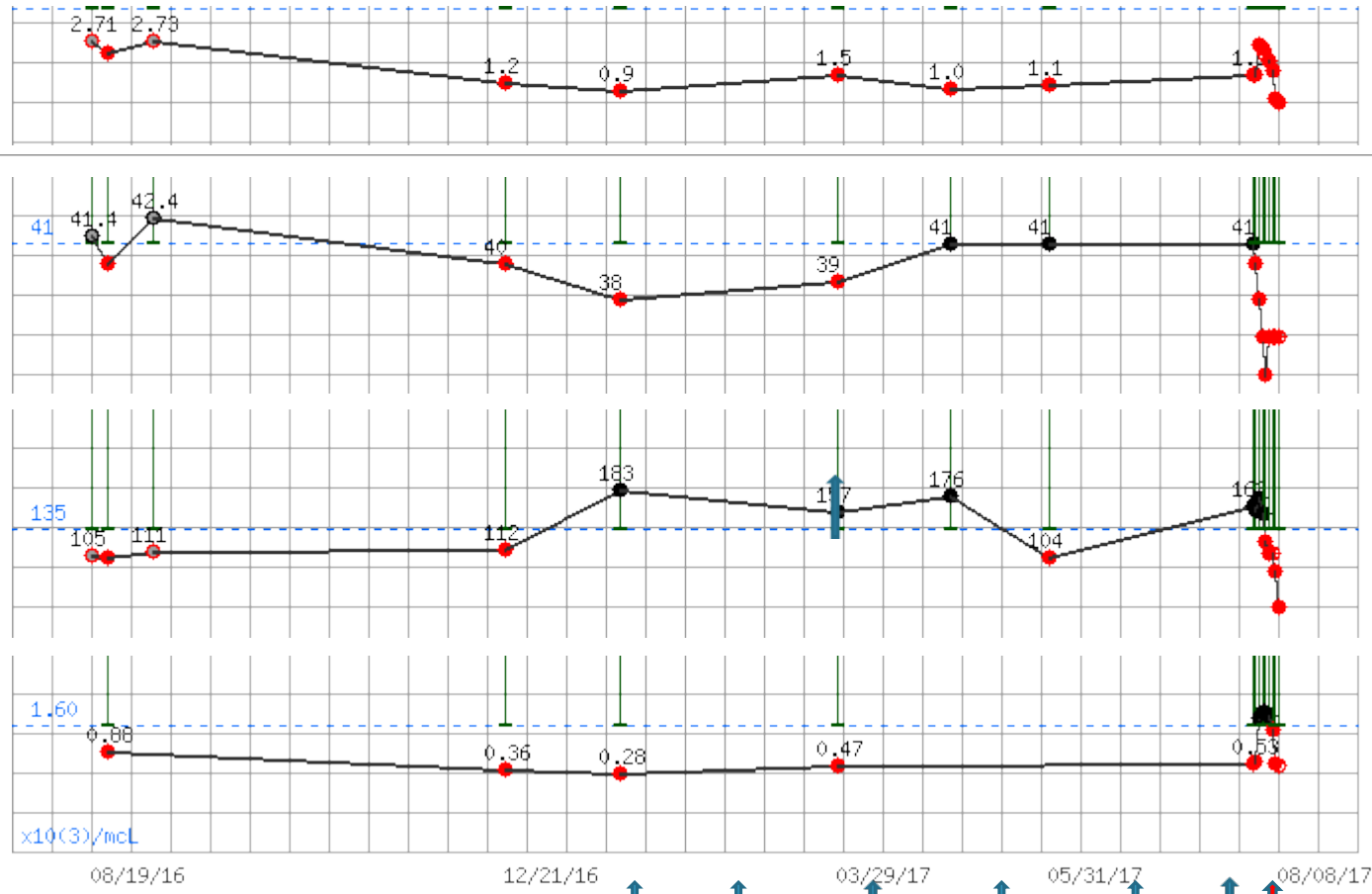
Progression of the patient's MDS

- Bone marrow 6/1/17: NORMOCELLULAR MARROW WITH ERYTHROID HYPERPLASIA, BILINEAGE DYSPLASIA, AND INCREASED BLASTS (10% BY IHC), CONSISTENT WITH MYELODYSPLASIA WITH EXCESS BLASTS
- Bone marrow 7/4/17: NORMOCELLULAR MARROW WITH PERSISTENT INVOLVEMENT BY MYELODYSPLASTIC SYNDROME WITH EXCESS BLASTS-1 (9.2% BY FLOW CYTOMETRY)
- Bone marrow 7/24/17: NORMOCELLULAR MARROW WITH MILD TRILINEAGE DYSPLASIA, CONSISTENT WITH PERSISTENT MYELODYSPLASTIC SYNDROME; NO INCREASE IN BLASTS





Progression of cytopenias



TREATMENT

Decitabine
6 cycles

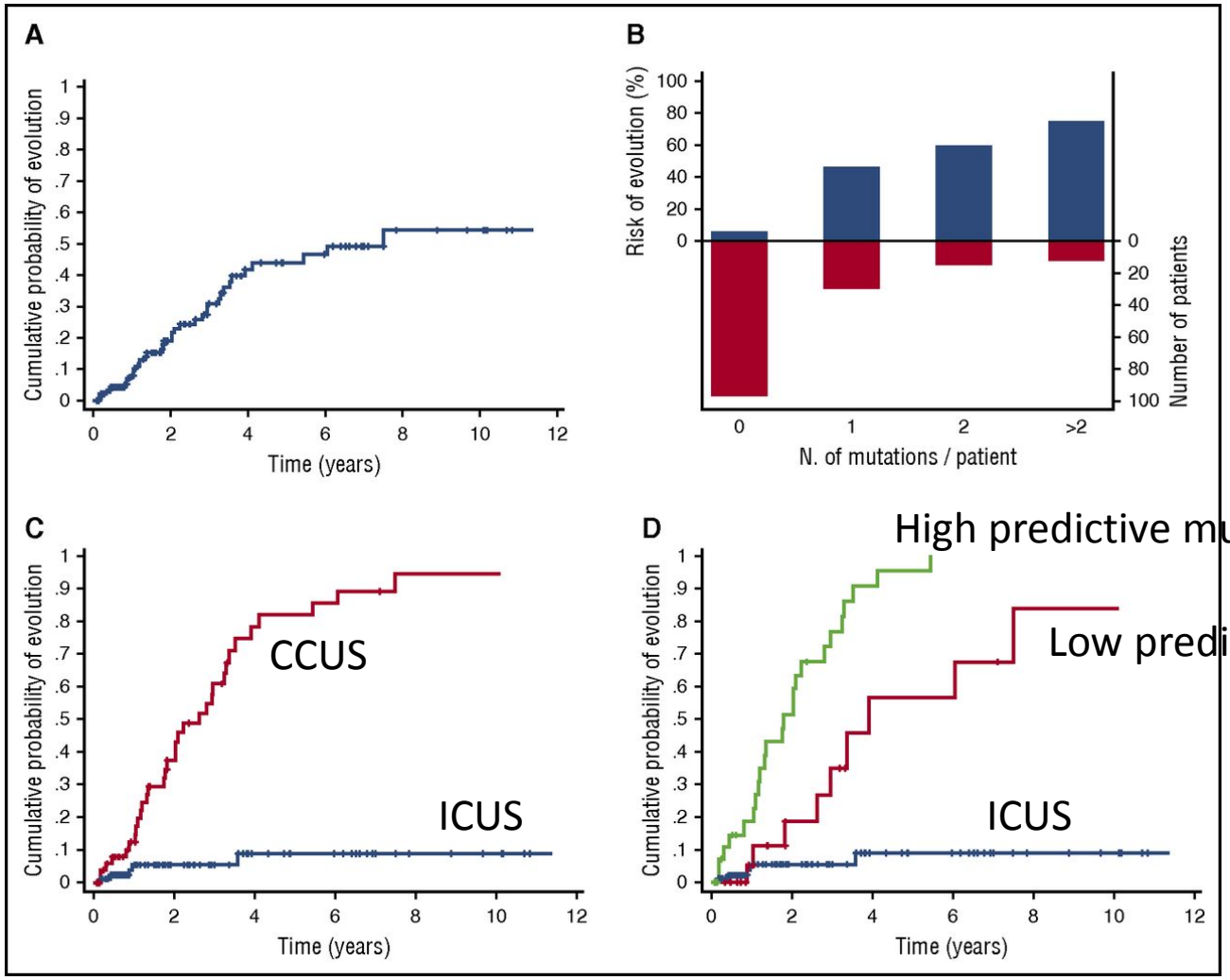
Pretransplant. Day 0=8/8/17
Mismatched unrelated ablative PBSCT

Clinical significance of somatic mutation in unexplained blood cytopenia

Luca Malcovati,[1,2] Anna Gallì,[2] Erica Travaglino,[2] Ilaria Ambaglio,[2] Ettore Rizzo,[3] Elisabetta Molteni,[1] Chiara Elena,[1,2] Virginia Valeria Ferretti,[1] Silvia Catricalà,[2] Elisa Bono,[1,2] Gabriele Todisco,[1,2] Antonio Bianchessi,[1,2] Elisa Rumi,[1,2] Silvia Zibellini,[2] Daniela Pietra,[2] Emanuela Boveri,[4] Clara Camaschella,[5,6] Daniela Toniolo,[5] Elli Papaemmanuil,[7] Seishi Ogawa,[8] and Mario Cazzola[1,2]

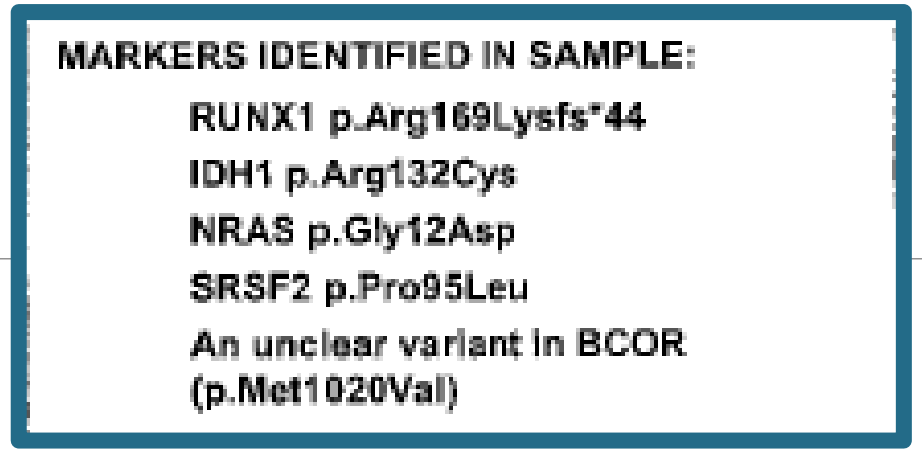
- Study design:
  - Learning cohort of 683 consecutive patients being worked up for unexplained cytopenias.
  - Targeted NGS using a 40 gene panel was performed (Illumina TruSight Myeloid Sequencing Panel).
  - Predictive values of mutation analysis were confirmed in an independent validation cohort of patients with a suspected diagnosis of myeloid neoplasm (190 patients).
- Results:
  - Standard work up of the cytopenic patients demonstrated that 60% of patients had a myeloid neoplasm, 22.5% had ICUS, 17.5% had other cause of cytopenia
  - 64% of patients with cytopenias had a somatic mutation in at least 1 of the 40 genes
  - Predictive value of mutations for myeloid neoplasm group
    - Spliceosome genes (SF3B1, SRSF2, U2AF1), JAK2, and RUNX1 mutations had highest predictive value for myeloid neoplasm (0.88-0.97)
    - Having 2 or more mutations had an odds ratio of 4.69 for having MDS or another myeloid neoplasm
  - Of the patients with original diagnosis of ICUS, 25% developed a myeloid neoplasm
    - 36% of ICUS patients were found to have 1 or more mutation (CCUS)
    - Allele frequencies of 10% or greater were significant
    - CCUS had a hazard ratio of 13.9 compared to ICUS for developing a myeloid neoplasm
    - CCUS: 5 yr and 10 yr cumulative probability of progression to myeloid neoplasm: 82% and 95% compared to 9% for unmutated ICUS
    - Highly predictive mutation pattern: Spliceosome gene mutations, or one of TET2, ASXL1, or DNMT3A with additional mutations

Probability of progression to myeloid neoplasm of patients receiving a provisional diagnosis of ICUS, according to mutation status and pattern.



Luca Malcovati et al. Blood 2017;129:3371-3378

Our patient



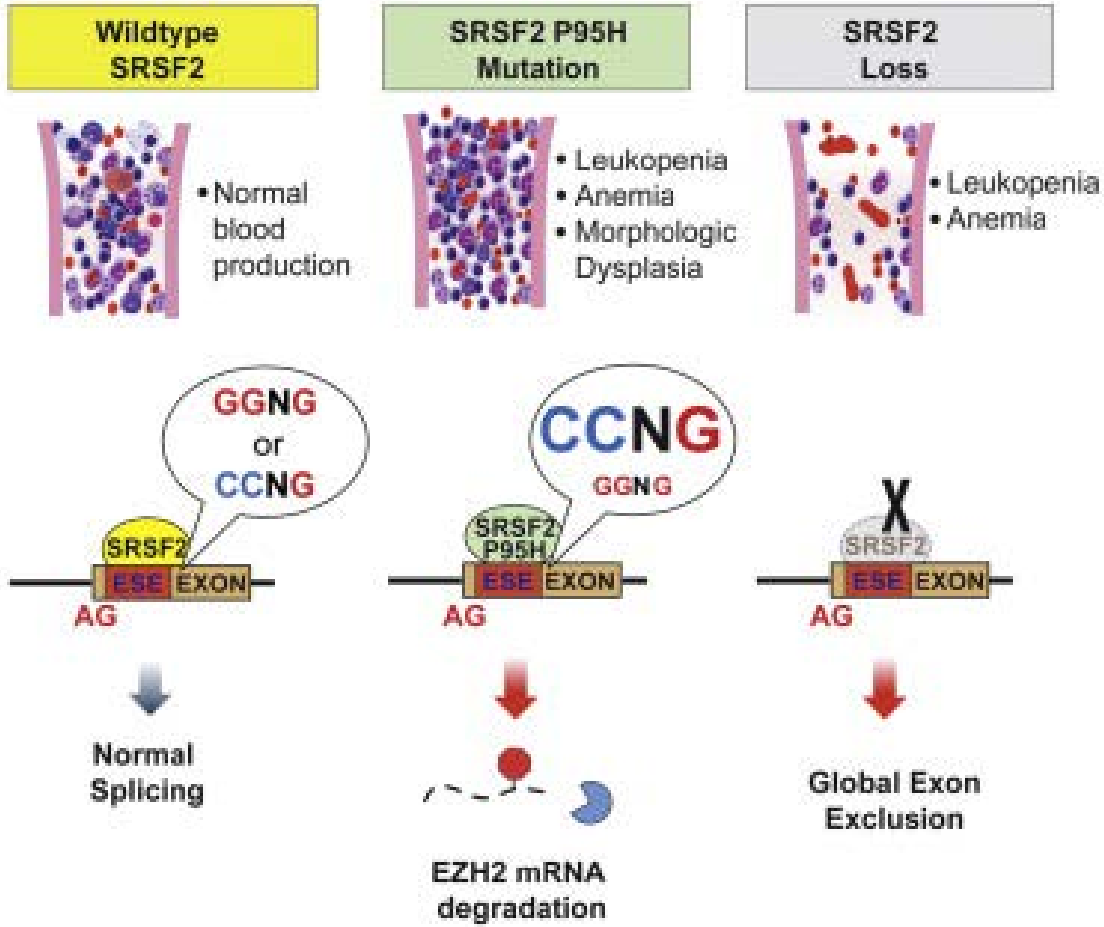
- Highly predictive mutation pattern
  - Splicesome mutation: SRSF2
  - 2 or more mutations (5)
  - RUNX1, IDH1/IDH2, and BCOR in the list of most frequent co-occurring mutations associated with development of myeloid neoplasm
- Time interval between ICUS and MDS: approximately 15 months
- Initial NGS performed due to ICUS diagnosis changed treatment approach
  - Frequent monitoring by CBC and bone marrow biopsy
  - Treatment with Decitabine
  - Allo PBSCT: potential cure

Questions raised by these studies

- What is the mechanism by which spliceosome mutations lead to MDS?
- Should NGS be performed on all patients with unexplained cytopenias?

How do spliceosome mutations cause MDS?

- SRSF2 P95H mutation affects specificity of pre-mRNA binding
- Mis-splicing of known hematopoietic regulators, among other targets
- Specifically, *EZH2* splicing is altered, introducing a premature termination codon and accelerated mRNA degradation
- *EZH2* encodes a histone H3K27 methyl-transferase commonly mutated in MDS
- SRSF2 and EZH2 mutations are mutually exclusive in mutational studies of MDS patients



Eunhee Kim, Janine O. Ilagan, Yang Liang, et al **SRSF2 Mutations Contribute to Myelodysplasia by Mutant-Specific Effects on Exon Recognition**
Cancer Cell, Volume 27, Issue 5, 2015, 617–630 http://dx.doi.org/10.1016/j.ccell.2015.04.006

Should NGS be performed on all patients with unexplained cytopenias?

- Screening for other causes essential
  - Nutritional
  - Infectious
  - Autoimmune
- Age cut-off?
  - Age range in the Malcovati study was 18-92 y.o., median 66 y.o., data not broken down by age
  - Previous study of 12,380 patients unselected for cancer or hematologic disease:
    - Genovese et al NEJM 371: 26, 2014
    - Whole exome sequencing of peripheral blood cell DNA
    - 10% incidence of somatic mutations in patients older than 65 years.
    - 1% incidence of somatic mutations in patients younger than 50 years.

Final panel diagnoses

- 8/24/16 bone marrow: Clonal cytopenia of undetermined significance (CCUS)
- 1/25/17 bone marrow: Myelodysplastic syndrome
- Subsequent bone marrow biopsies demonstrate progression to myelodysplastic syndrome with excess blasts-2.