17TH ULTMANN
CHICAGO LYMPHOMA SYMPOSIUM

THE UNIVERSITY OF CHICAGO
MEDICINE & BIOLOGICAL SCIENCES

17TH INTERNATIONAL ULTMANN CHICAGO LYMPHOMA SYMPOSIUM

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chicagolymphoma.com

Diving into the Diagnosis

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Disclosure Information

- CF is a clinical consultant for Duke Lifepoint, UPHSM.
- JG is a speaker for Sanofi Genzyme, BMS and has served on an advisory board for Celgene.
- No discussion of off label use and/or investigational use included in this presentation

Learning Objectives

- Understand the significance of various laboratory test results in different subtypes of lymphoma
- Review the clinical implications of lab results on patient management

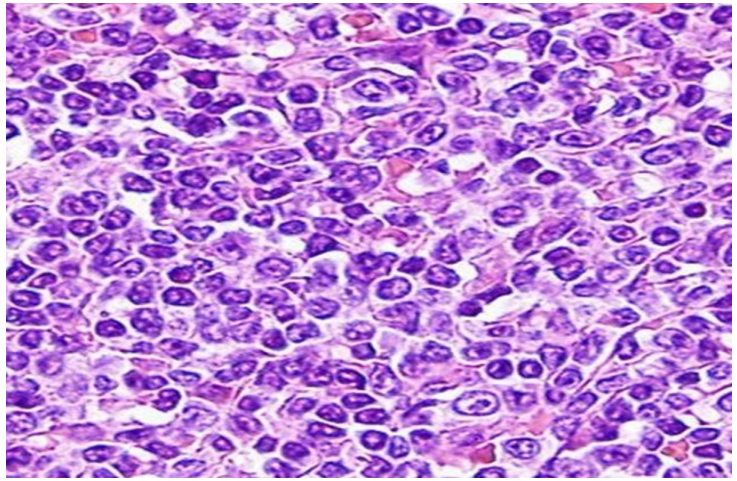
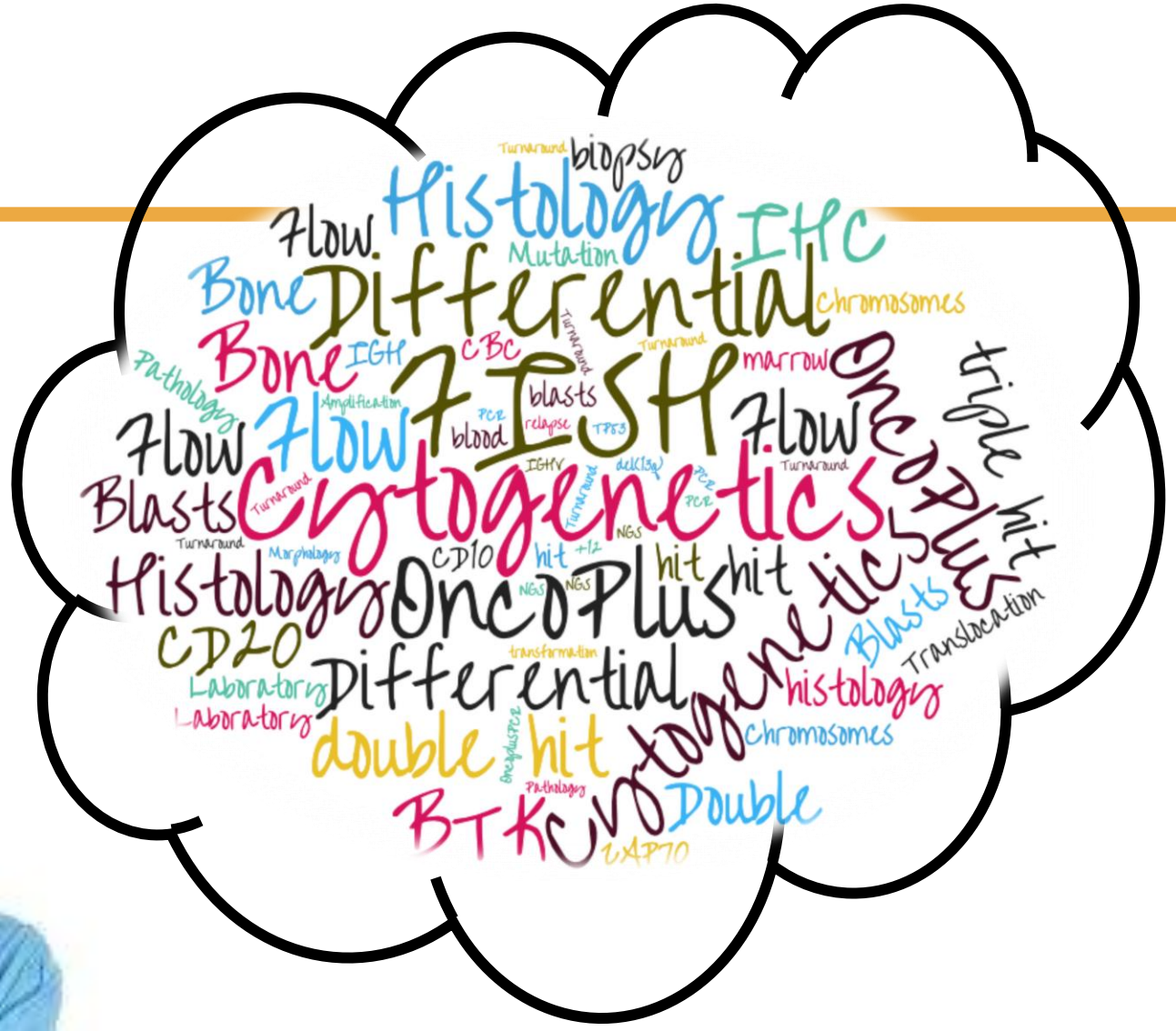


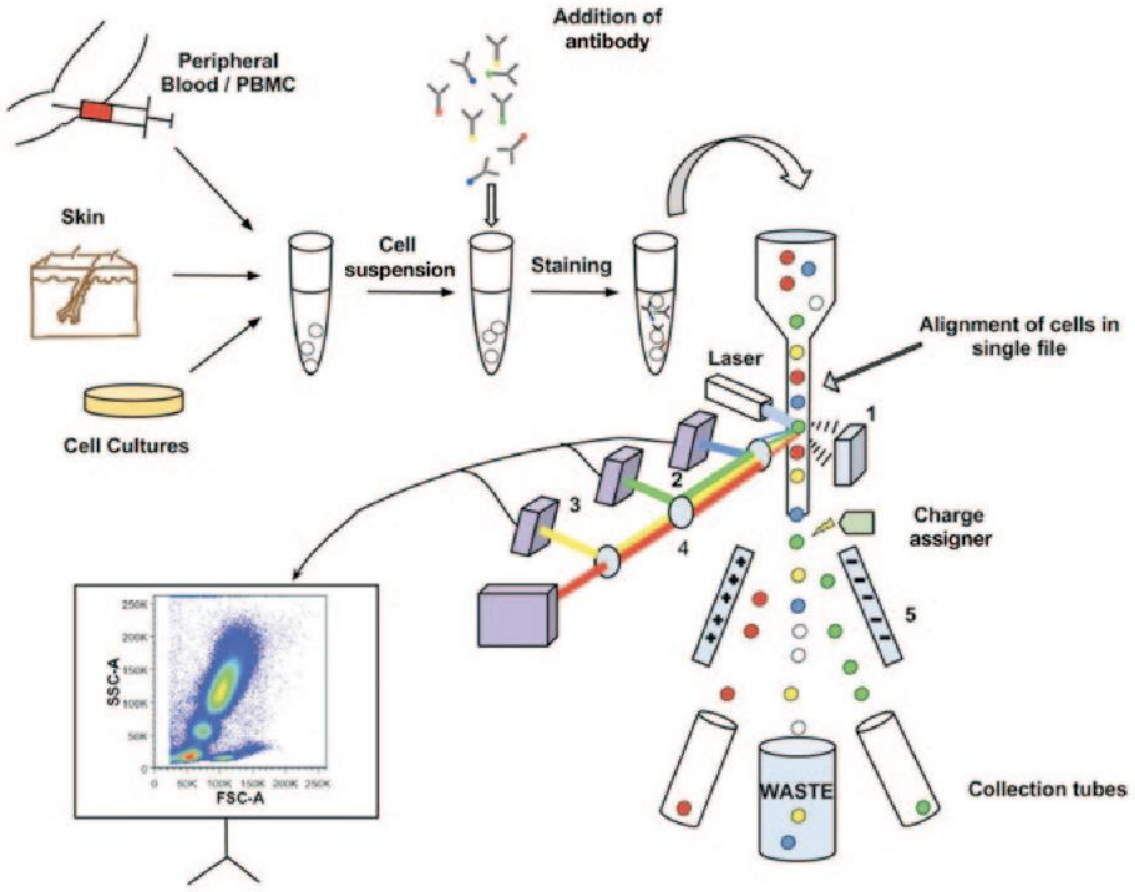
Image credit V. Shankar, 2016

Lab test overload!



Lab tests for Lymphoma: The Big Picture

- **Flow cytometry:** a suspension of cells are sorted by size and cell surface markers. Helpful for determining lineage of new leukemias/lymphomas, and very sensitive for detecting small populations (MRD)



Jahan-Tigh, R.R., Ryan, C., Obermoser, G., & Schwarzenberger, K. (2012). Techniques made simple Flow Cytometry.

Lab tests for Lymphoma: The Big Picture

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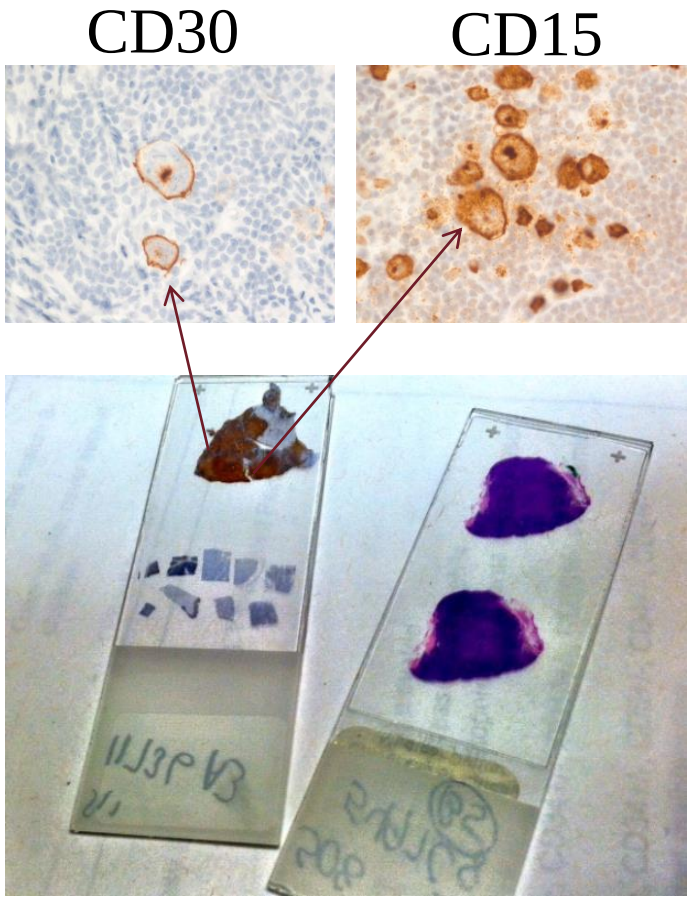
- **IHC**: similar to flow, identifies cells on a slide based on protein expression.



Immunohistochemistry

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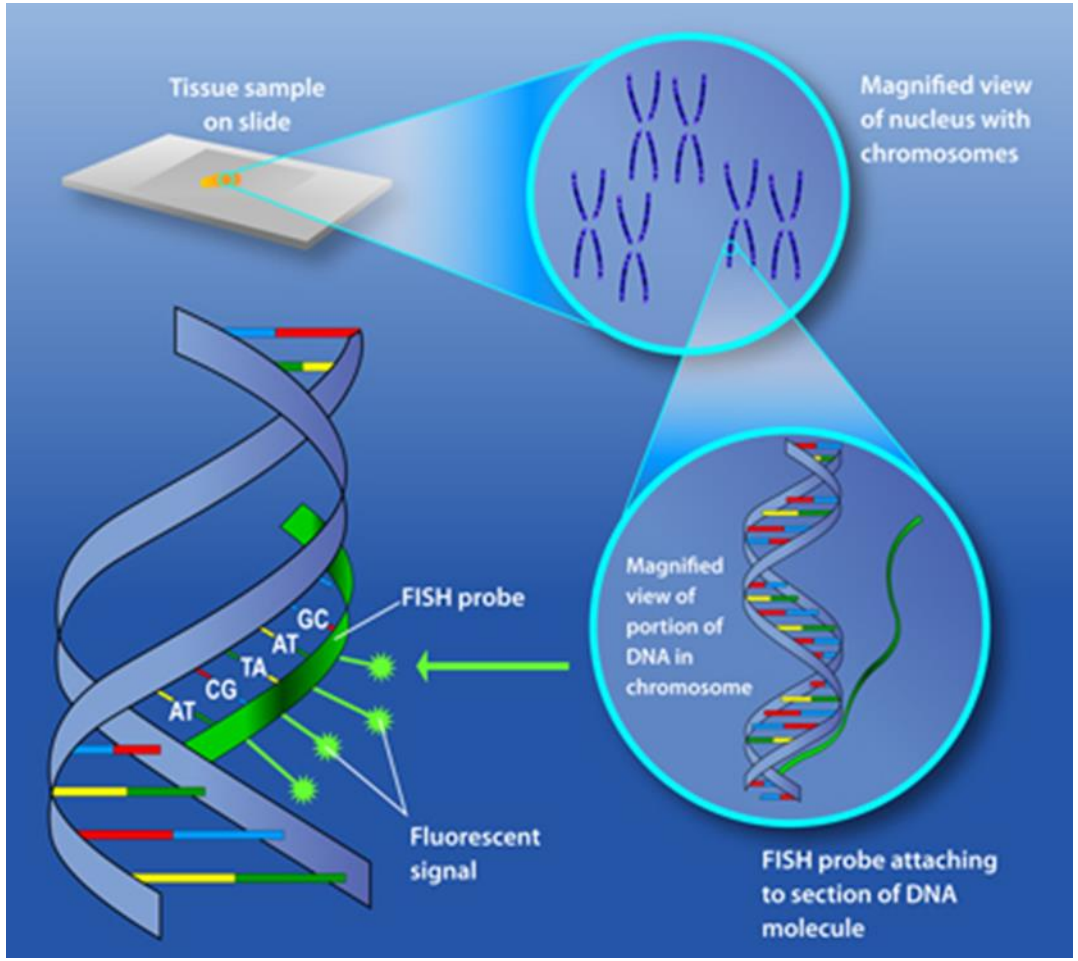
- **FISH:** Uses fluorescently labeled fragments of DNA to recognize complementary sequences inside cells affixed to a slide.



NEED REF

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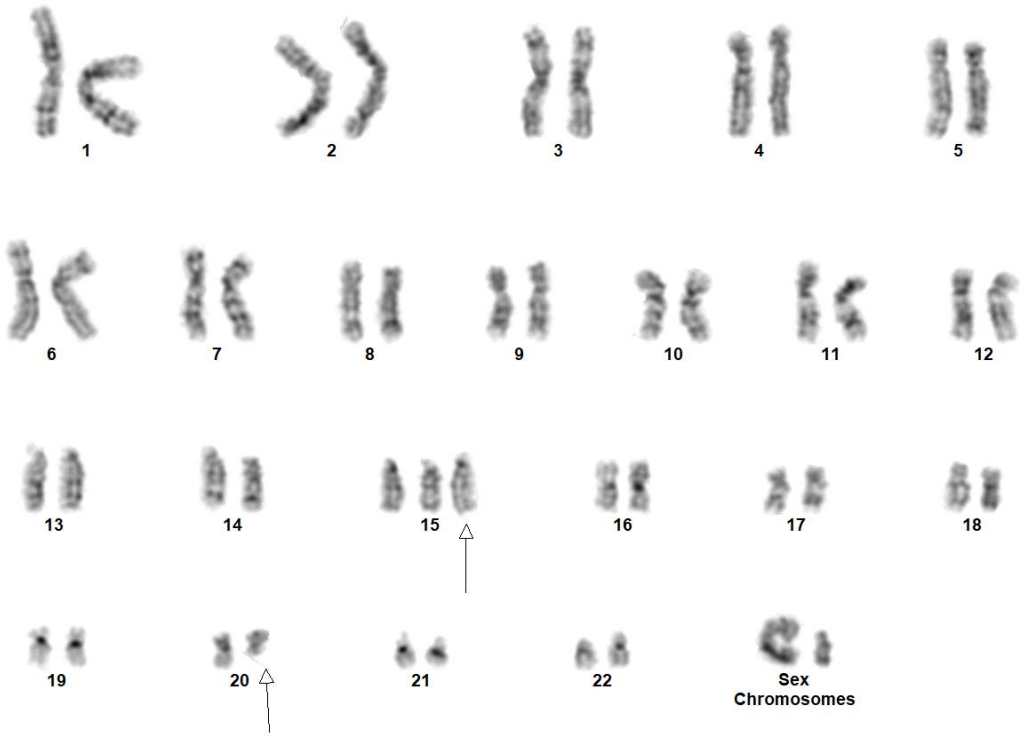

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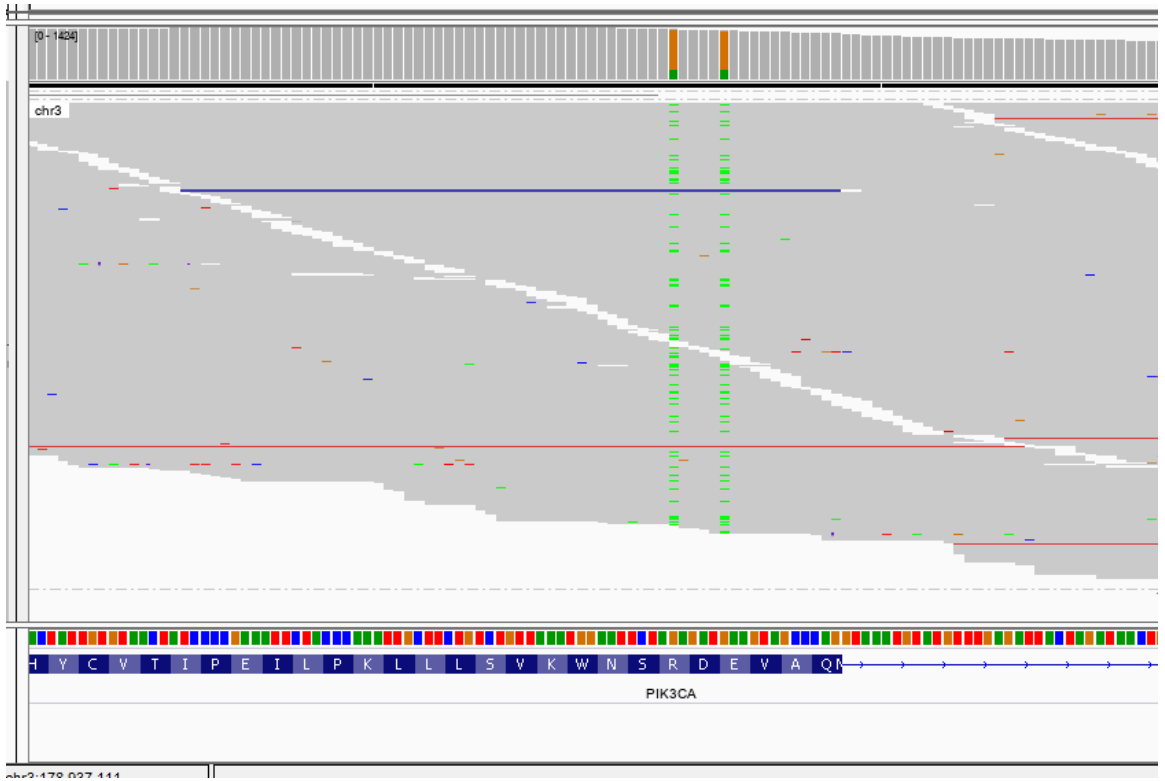
- **Next Generation Sequencing (NGS)**: Emerging technologies that detect genomic alterations useful for diagnostic, prognostic and targeted therapy selection.



High Grade B-Cell Lymphoma (HGBL) Case Study RF

- RF 75 y/o male p/w severe back pain and N/V
  - CT imaging revealed retroperitoneal adenopathy and osseous lesions
  - RP lymph node bx c/w high grade b cell lymphoma with MYC and BCL2 gene rearrangements
  - ECOG 1
  - No other significant PMH
  - International Prognostic Index (IPI)score 4

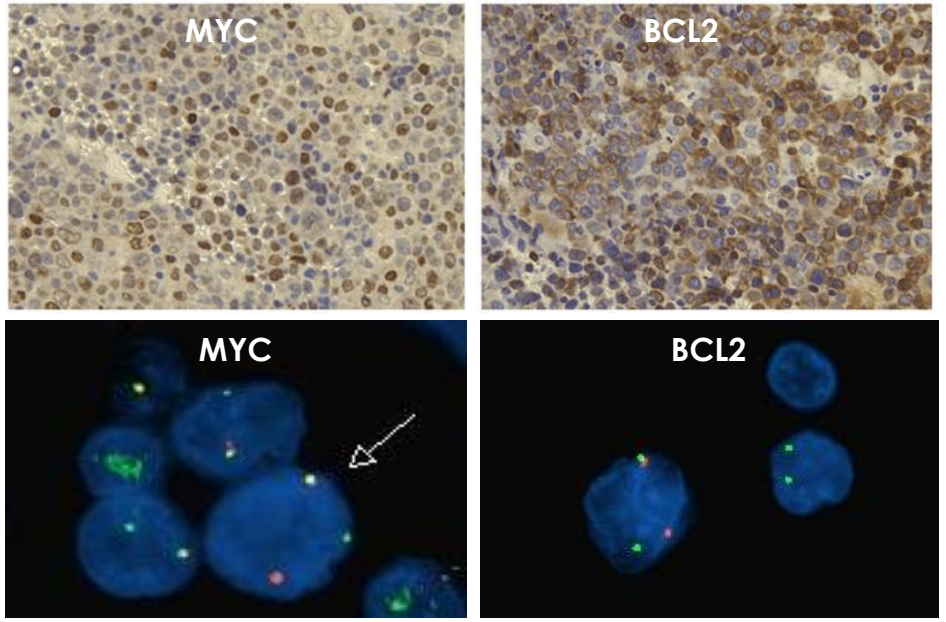
  - Diagnostic considerations at time of diagnosis for large cell lymphoma?

Laboratory testing considerations for DLBCL

- Flow cytometry analysis
- Histological evaluation

- Immunostains
  - B cell antigens
  - CD10, BCL6, Mum1 (COO)
  - MYC and BCL2 "double expressor" DLBCL

- Cytogenetics
  - FISH for MYC, BCL2 "double/triple hit" DLBCL
  - Karyotype



High-Grade B-Cell Lymphoma With MYC and BCL2 and/or BCL6 Rearrangements

- "Double-hit" or "Triple-hit" DLBCL by FISH
  - WHO HGBL-DH/TH
  - Identifies rearrangements between MYC, BCL2 and/or BCL6
  - Up to 8% of de novo DLBCL
  - Aggressive disease
  - Intensive therapies

- "Double Expressor" DLBCL by IHC
  - WHO HGBL-NOS
  - Increased protein expression of MYC and BCL2
  - 20% of DLBCL
  - Poor prognosis relative to negative expression

Swerdlow et al., WHO Classification of Tumours of Haematopoietic and Lymphoid Tissues (Revised 4th Ed), 2017

Treatment for DLBCL

- Chemoimmunotherapy with rituximab, cyclophosphamide, doxorubicin, vincristine, and prednisone (R-CHOP) cures an average of 70% of standard diffuse large b-cell lymphoma (DLBCL) cases

- Cures based on different factors
  - Age
  - IPI score
  - Molecular cell of origin subtype
  - Presence/absence of chromosomal rearrangements

- HGBL accounts for 5-7% of DLBCLs
  - Poor prognosis
  - Long-term survivors are rare

(Riedell & Smith, 2018)

Standard or Intensive Regimen for HGBL?

- R-CHOP vs rituximab, etoposide, prednisone, vincristine, cyclophosphamide, and doxorubicin (R-EPOCH) or other similar intensive regimens?

- Majority of data available is retrospective
  - Study with 129 pts with DHL at MD Anderson
    - R-EPOCH superior event free survival (EFS)
  - Study with 311 pts with DHL treated with various regimens including R-CHOP & other intensive regimens like R-EPOCH
    - Inferior median progression free survival (PFS) in R-CHOP, no overall survival (OS) benefit R-EPOCH

- Data remains limited so treatment choices vary depending on institution, treating clinician and assessment of pts ability to tolerate more intensive regimen

(Riedell & Smith, 2018)
Petrich, Gandhi, Jovanovic, et al., 2014)

RF Case Study Continued

- Started treatment with R-CHOP
- PET imaging s/p 3 cycles c/w partial response to therapy (PR)
- Therapy switched to R-EPOCH with IT MTX for CNS prophylaxis
  - End of treatment imaging c/f progressive disease (PD)
- Bx c/w HGBL (gains of both MYC & BCL2)
- Started lenolidamide and rituxan for second line salvage
  - PET s/p 4 cycles with PD
- Referred for CAR T-cell therapy and received tisagenlecleucal (tisa-cel) infusion
- Now 12 months out and remains in complete remission (CR)

Nursing Considerations for HGBL

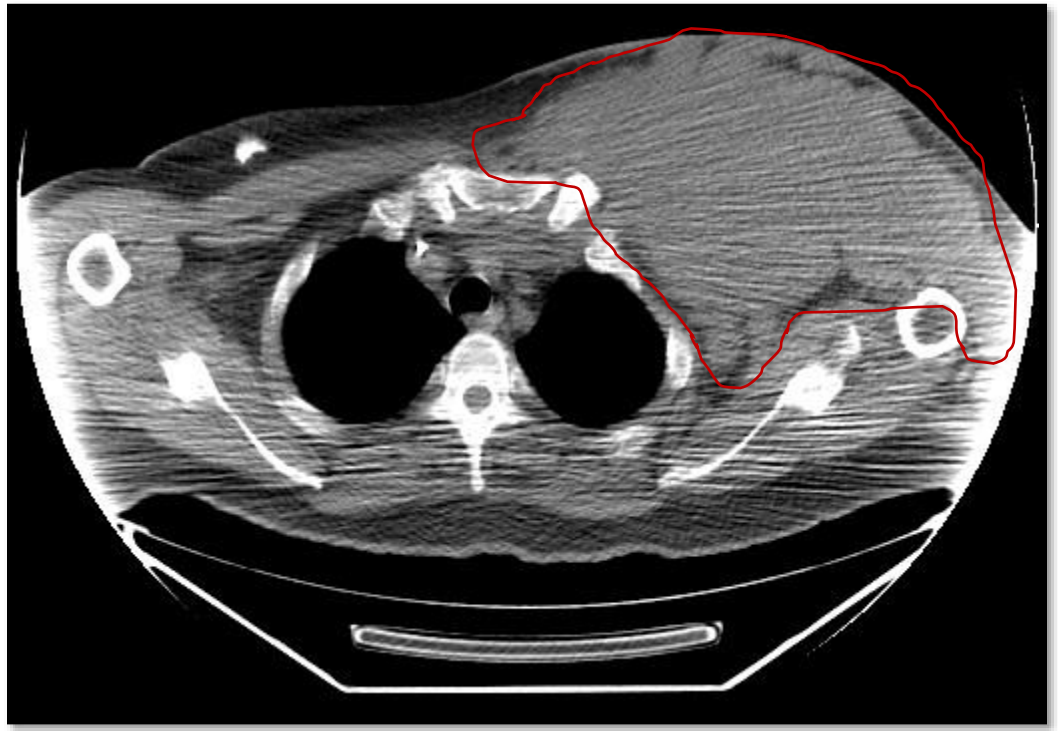
- R-CHOP vs. R-EPOCH or other intensive regimen?
  - Inpatient vs. outpatient administration
  - CNS prophylaxis
  - Frequency of lab monitoring and follow up
  - Management of treatment side effects
  - Patient education

High grade B-cell Lymphoma: A diagnostic challenge

- 52 year-old male
- Developed left shoulder pain, initially thought to be a musculoskeletal injury, but symptoms progressively worsened
- Admitted to an outside institution for evaluation
  - Clinical exam/imaging: Mass in the left chest wall/shoulder
  - Core biopsy: High-grade B-cell lymphoma, MYC focal, BCL2 negative, Ki67 >90%
  - Staging bone marrow: Negative for lymphoma
  - PET scan: Disease localized to the left chest
- Received 4 cycles of R-CHOP

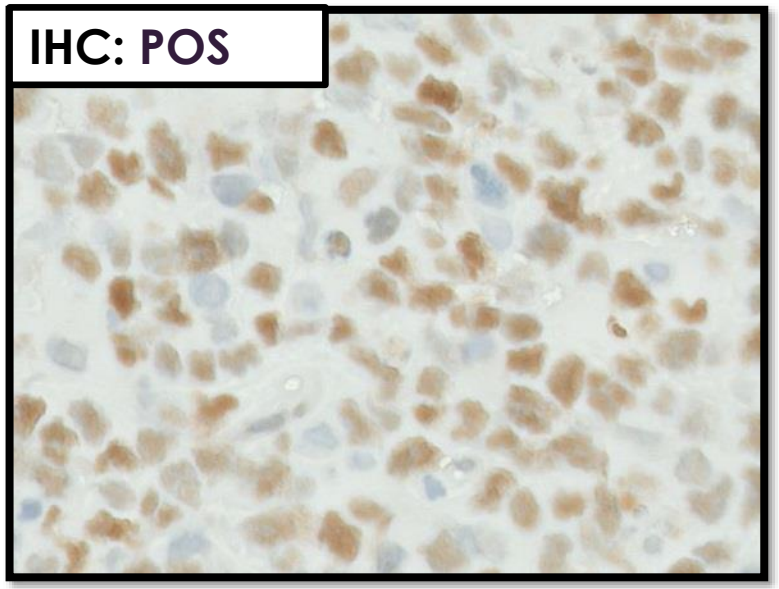
Clinical history, FR

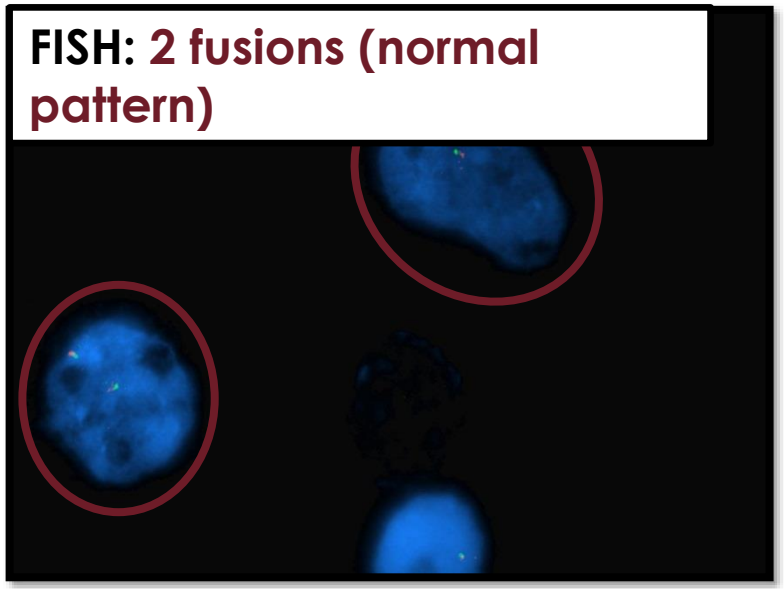
- on R-CHOP, the patient's disease progressively worsened with:
  - Enlargement of the primary mass
  - Progressive lymphadenopathy
  - Overlying skin ulcers/nodules
  - A new left distal femur lesion
- The patient transferred care to University of Chicago
- Underwent debridement of the enlarged left shoulder/chest wall mass



Chest CT after 4 cycles of R-CHOP showing enlargement of primary mass

MYC discrepancy (IHC+, FISH-)

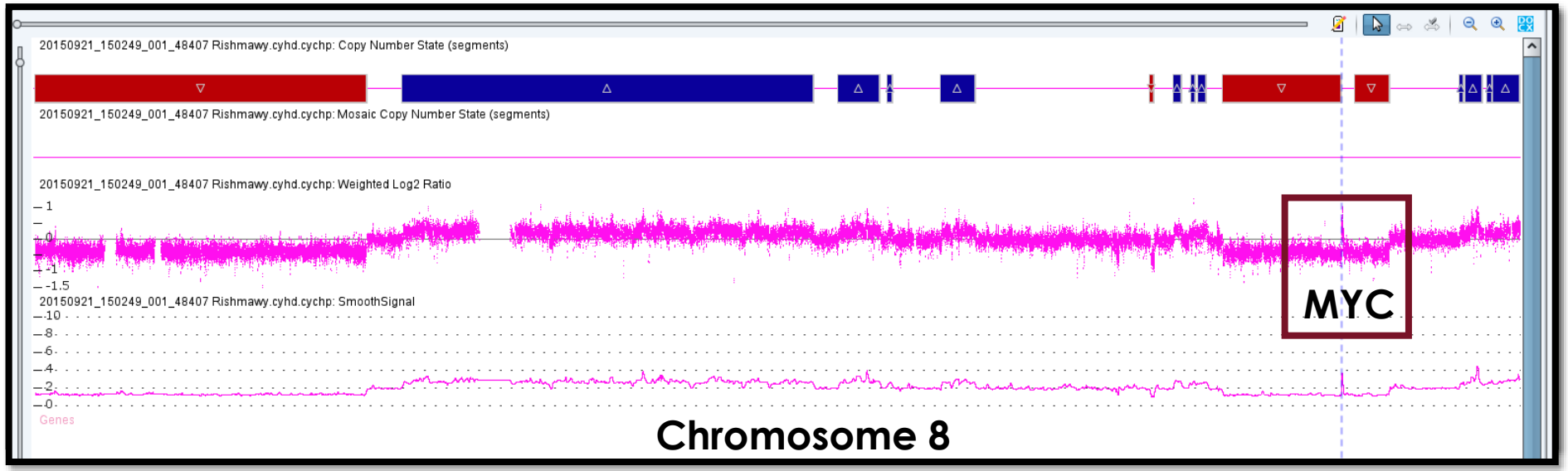




- BCL2 positive (IHC, FISH)
- High expression MYC
- FISH negative for MYC rearrangement
- **SNP microarray to clarify**

Sukhanova M. et al., Reviews & Reports. 24(6):272–276.

SNP array results: High Grade B-ell Lymphoma NOS



- Numerous complex chromosomal abnormalities
- Negative for 11q gain/loss characteristic of Burkitt-like lymphoma
- **1 MB gain of 18q22.22 including BCL2**
- **180 Kb 2 copy gain of MYC at 8q24.1**

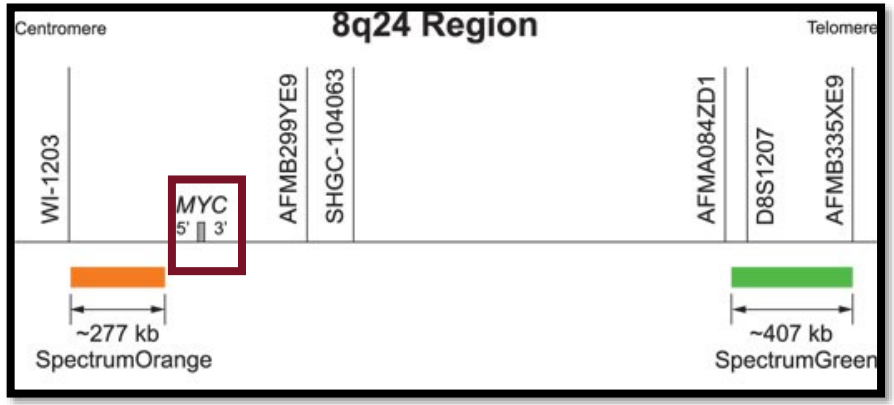
Sukhanova M. et al., Reviews & Reports. 24(6):272–276.

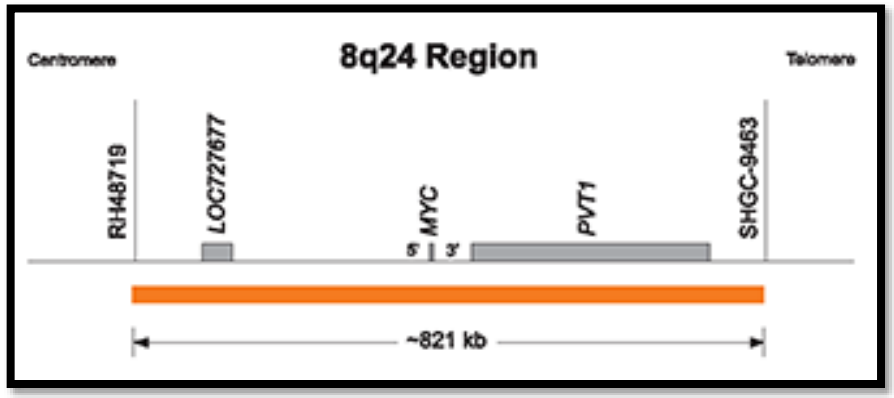
FISH sensitivity: not all probes are equal

- *Dramatic and aggressive clinical course progressing through chemotherapy*

- **Identification of MYC deregulation**
  - **MYC IHC+** → protein overexpression
  - **MYC FISH** with two fusions (normal pattern)
  - **SNP CMA** confirmed MYC gene amplification as the result of a chromothripsis event on chromosome 8

MYC gain not detected by commercial FISH probes





Sukhanova M. et al., Reviews & Reports. 24(6):272–276.

IHC sensitivity: not all antibody stains are equal

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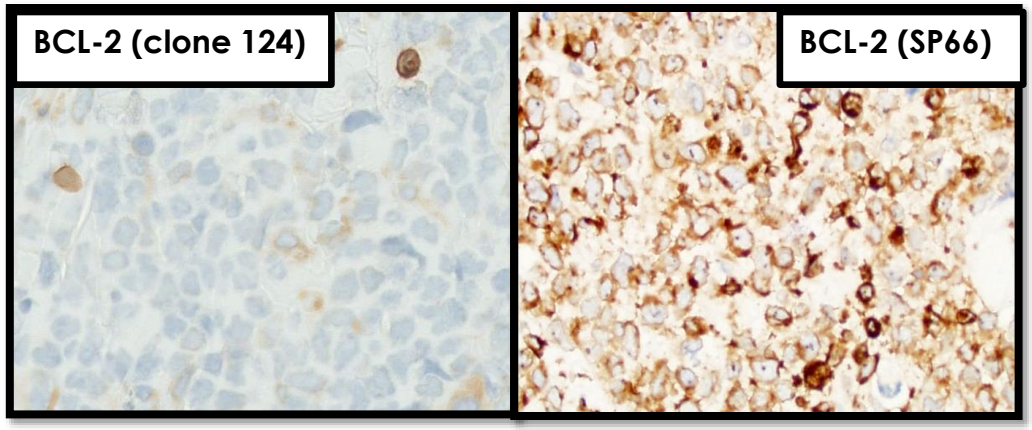
- **Identification of BCL2 deregulation**
  - **BCL2 IHC** (clone 124) was negative
  - **BCL2 FISH** showed gains and rearrangement of the BCL2 locus
  - **BCL2 IHC** (SP66 clone) was was subsequently positive
  - **SNP CMA** confirmed the FISH findings

Differential detection likely due to genetic complexity of BCL2 locus



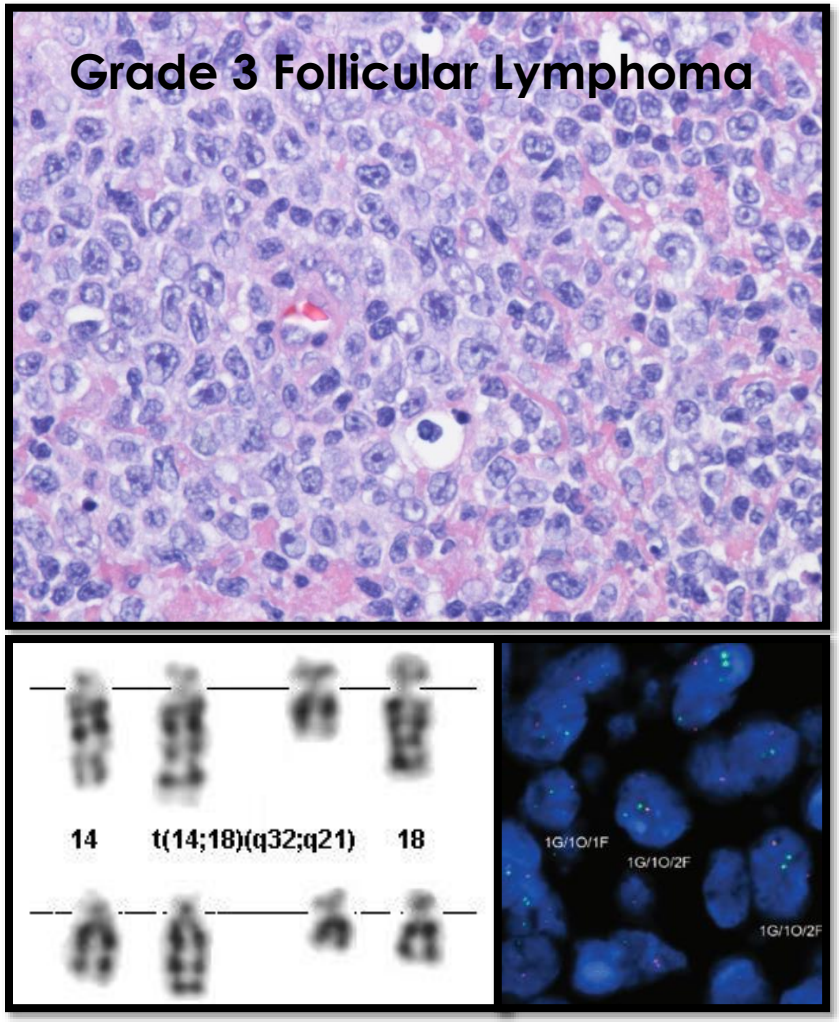
Sukhanova M. et al., Reviews & Reports. 24(6):272–276.

Follicular Lymphoma

- Follicular Lymphoma (FL) is the most common type of indolent B-cell lymphoma
  - In 2019 pts had a 10 year overall survival probability of over 70%
- Diagnosis and staging primarily made on histology and clinical parameters
- Cytogenetics and FISH can add prognostic value
- Molecular not standard of care
- Histologic transformation to more aggressive lymphomas and relapses remain a challenge

Follicular Lymphoma

- Hallmark t(14;18) in 85-90%
- BCL6 rearrangements in 5-15%
- Complex karyotypes are associated w/ higher histological grade
- Transformation (tFL) to DLBCL with inferior survival
  - TP53 mutations in 25-30% (resistance to rCHOP)
  - Bi-allelic is common by LOH or deletion
  - Immune evasion: *B2M* (antigen presentation) and *CD58 (*T-cell and NK cell activation)
  - Epigenetic regulation: *KMT2D, CREBBP, EZH2*
  - CDKN2A/B: 45-70% of tFL; may be predict response to therapy



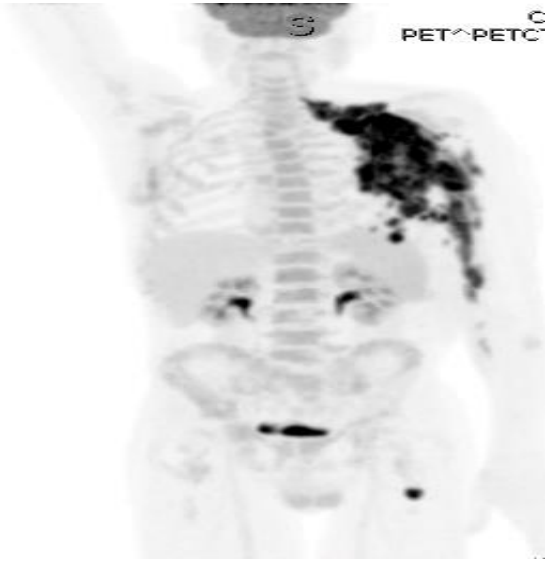
Brad S. Kahl, and David T. Yang Blood 2016;127:2055-2063.

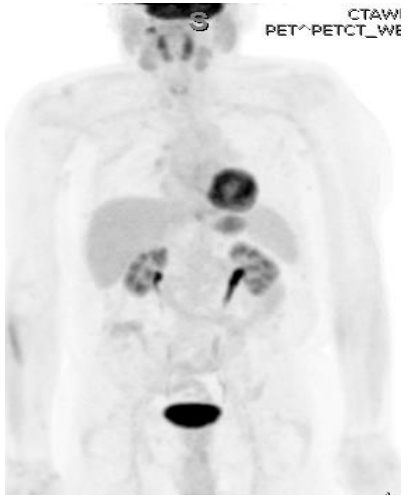
FL Case Study RM

- 66 y/o M with no significant PMH. Initially p/w acute onset left flank & groin pain treated w/ NSAIDs
  - Of note he also reported having lumps under both axilla present for 2 years prior
- Developed worsening pain. CT findings revealed large chest mass, adrenal masses & axillary LAD
  - Right axillary LN biopsy c/w grade 1 FL
  - FISH positive for IGH, MYC and BCL2 rearrangements
  - Transformed FL with double-hit genetic features
- Referred to oncologist for additional w/u & management
  - PET/CT w/ diffuse disease & skeletal involvement
  - Bone marrow with >95% involvement
  - Karyotype abnormal: add(8)(q24), add(14)(q32), trisomy 12, del(13q)
  - CSF + for lymphomatous involvement

RM case study continued

- Transformed FL w/ aggressive features, Stage IVB
- Resistant to several therapies (DA-R-EPOCH, R-DHAP, R-GEMOX)
- Treated with CAR T-cell therapy on clinical protocol & achieved remission



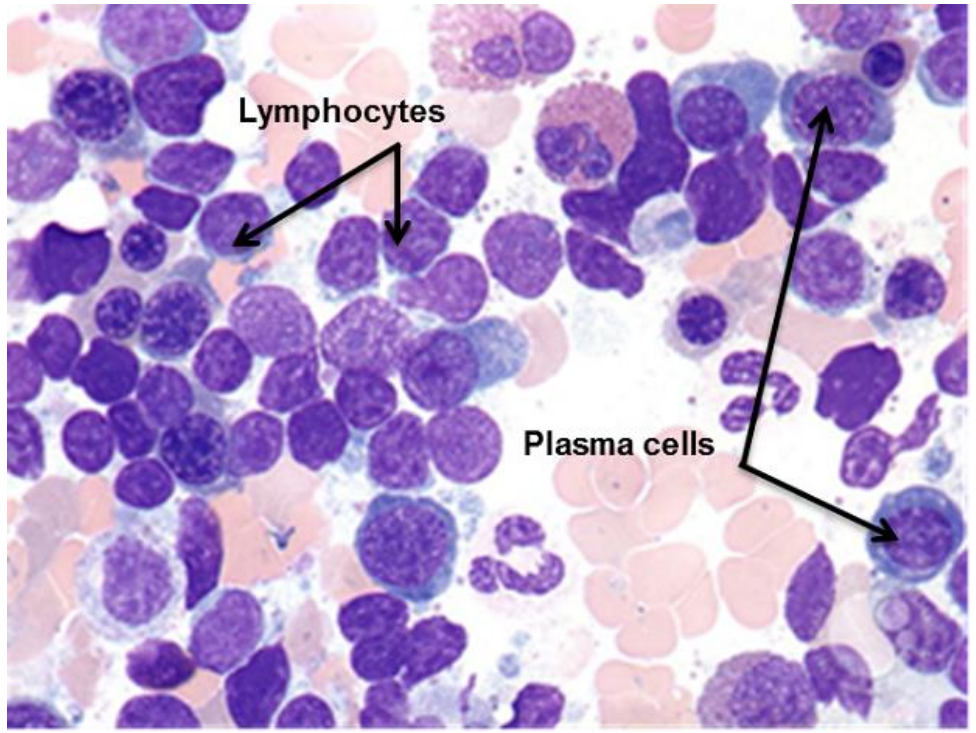


RM case study continued

- Developed new small left forearm lesion w/FDG avidity
- FNA bx c/w recurrent B-cell lymphoma
- Underwent radiation therapy w/ good clinical response
- Within a few months developed new L axillary LAD
- Biopsy c/w relapsed (DLBCL)
  - Double expressor of MYC and BCL2 w/ 100% proliferation
  - FISH confirms MYC, BCL2 rearrangements c/w relapsed double-hit lymphoma
- Started therapy on check point inhibitor clinical trial & again achieved a CR, which is ongoing

Mutations are also important

- 61 yo male with relapsed LPL/WM
- Extranodal lesions: MCL vs. LPL?
- Cyto with Del(6q), loss of 17p
- MYD p.L265P mutation
  - Activating mutation
  - 95% of LPL
- Response to ibrutinib
- CXCR4 mutations (30-40% LPL) diminish response



J Clin Oncol. 2017 Mar 20;35(9):994-1001

Chronic Lymphocytic Leukemia (CLL)

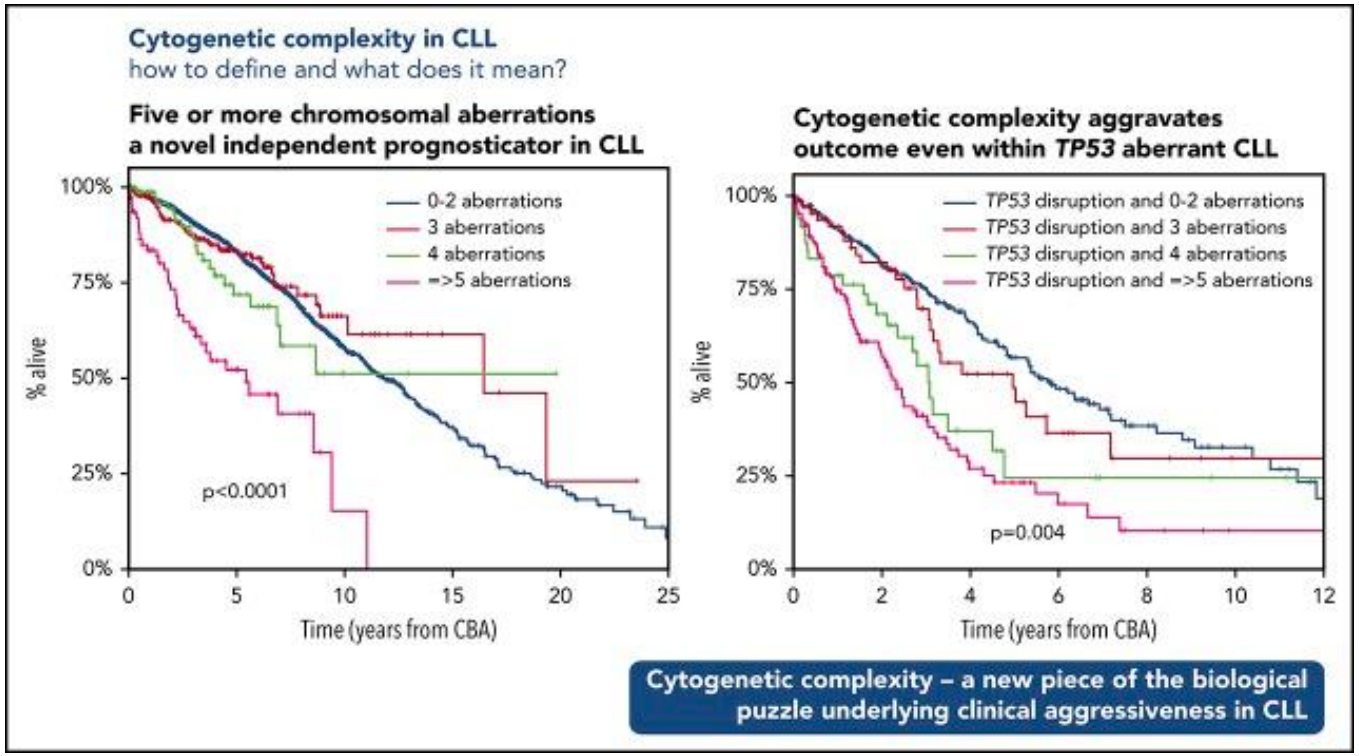
- CLL is a lymphoproliferative disorder characterized by clonal proliferation and progressive accumulation of morphologically mature, CD5+ B lymphocytes in the blood, bone marrow, and lymphatic tissues
- Staging and Prognosis:
  - Rai Staging most common in U.S. (low, intermediate, high risk)
- Clinical course varies, observation appropriate for many pts
- DNA sequencing, cytogenetics, serum markers helpful for prognosis

Chronic Lymphocytic Leukemia (CLL)

- 80-90% will have abnormalities detected by commonly used CLL panels:
  - Deletion 13q (30-65%)
  - Trisomy 12 (15-20%)
  - Deletion ATM (4-30%)
  - Deletion TP53/17p (3-10%)
  - 3-15% will have mutations: *NOTCH1, SF3B1, TP53, ATM, BIRC3*, *POT1* and *MYD88*
- IGHV, ZAP-70
- 2-8% will develop DLBCL
- <1% will develop Hodgkin Lymphoma

Complex karyotype in CLL

- Low mitotic index precludes routine chromosome analysis
- B cell mitogens allow labs to routinely karyotype CLL cases
- >5 cytogenetic abnormalities independently predicts poor outcome



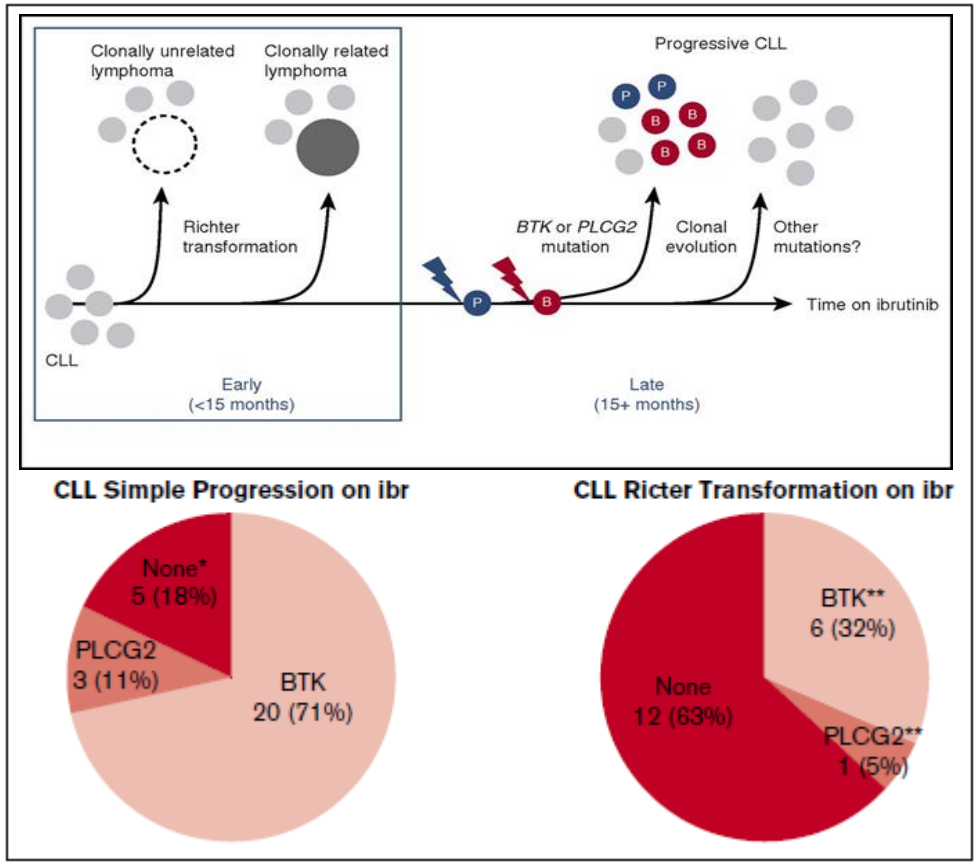
Panagiotis B. et al., Blood 2019 Mar 14;133(11):1205-1216

CLL case study LM

- LM 72 y/o male with CLL, diagnosed in 2011
  - Karyotype normal
  - CLL FISH panel shows deletion 13q
- Treated on clinical protocol, s/p 6 cycles of Fludarabine and Rituxan in 12/2011 followed by a course of Revlimid 10 mg/day per day.
  - Tolerated well
- In Aug 2014, he developed abdominal fullness with a palpable lower abdominal mass, early satiety, and weight loss. CT c/w PD.
  - FISH shows progression: del(13q), ATM loss, del(17p)
- Started ibrutinib and tolerated well for ~ 5 years, but again developed weight loss, increased LAD & WBC count c/w PD
- OncoPlus shows two BTK mutations
  - c.1441T>A
  - c.1442G>C
  - Both result in common p.c481S

CLL Progression: High incidence of BTK mutations

- CLL progression and Richter Transformation
- Common in heavily pretreated patients and high risk CLL
- Impact of p.C481S
- Significance of 2 independent mutations arising in subclones
- Salvage therapies: PI3K inhibitors, BCL2 targeting agents



Kadri, Lee, Fitzpatrick et al., Bood Adv 2017 May 2;1(12):715-727

LM CLL case study continued

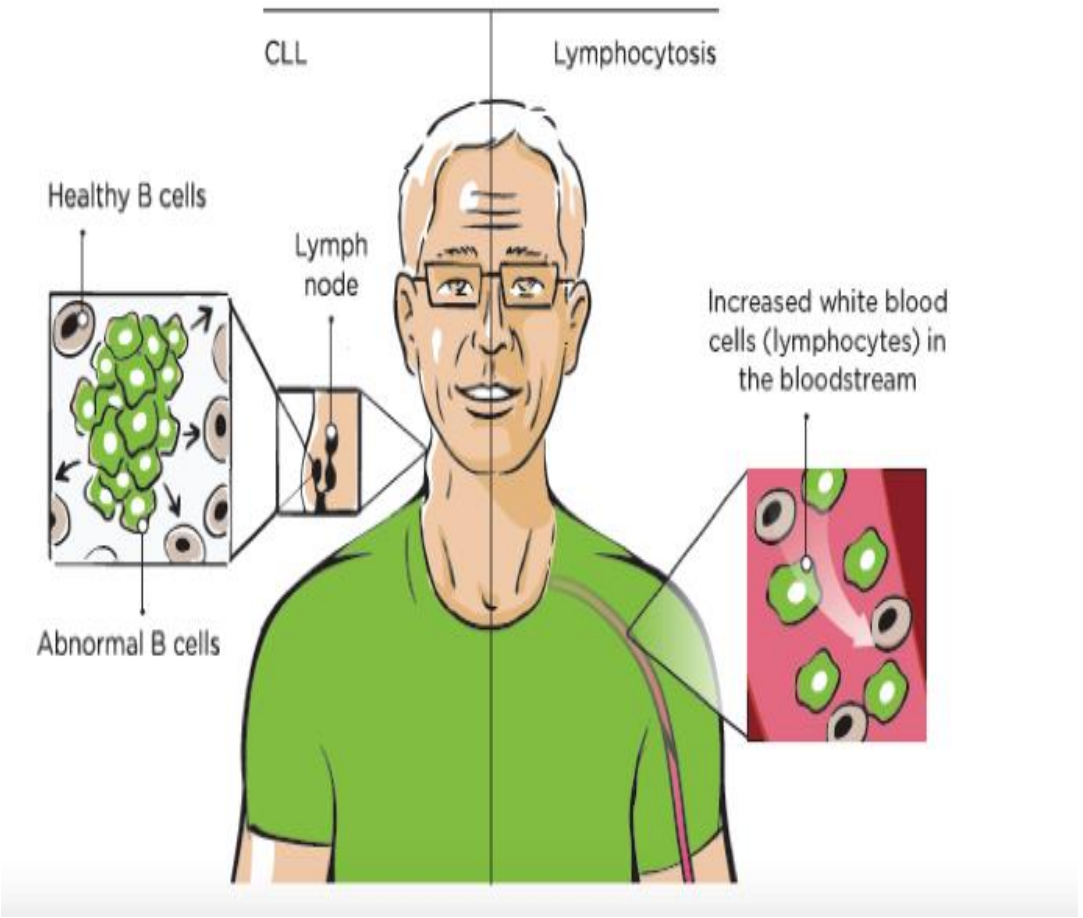
- Therapy switched to novel agent venetoclax due to ibrutinib resistance
- Attempted to add acalabrutinib for combination therapy, but was stopped after 3 weeks 2/2 myalgias, arthralgias, and fatigue.
- Tolerated venetoclax well with decrease in adenopathy on CT imaging and improved blood counts
- Repeat flow cytometry after >6 months of therapy with no evidence of minimal residual disease, he continues on venetoclax

Nursing considerations

- Targeted therapies and novel agents have changed the treatment landscape in CLL
- Nurses are a crucial aspect of patient management with these therapies

Nursing considerations with BTK inhibitors

- Ibrutinib is a BTK inhibitor
- Common adverse reactions with ibrutinib
  - Cytopenias
  - Diarrhea
  - Fatigue
  - Musculoskeletal pain
  - Bruising
- Patient education
  - Consider drug diary
  - Supportive care
  - Drug Interactions
- Reassurance over potential lymphocytosis
- Frequency of lab monitoring



Nursing considerations with novel agents

- Venetoclax is a BCL-2 inhibitor
- Common adverse reactions:
  - Cytopenias
  - Diarrhea
  - Nausea
  - Upper respiratory infections
  - Fatigue

- Inpatient or outpatient administration?
  - Lower tumor burden often managed as an outpatient
- Ramp up dosing generally over 5 weeks
- TLS monitoring during ramp up
  - Frequent labs
  - Consider IV fluids
  - Allopurinol or rasburicase
- Patient education

Conclusions

- Accurate pathologic diagnosis is crucial to ensure appropriate therapy and treatment course for pts
- Studies that can provide useful data to aid in clinical diagnosis:
  - IHC
  - Flow cytometry
  - Cytogenetics
  - Molecular profile
- Close collaboration between the clinical team and the laboratory can improve testing and patient care