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Imagine A Cure: A World Without WM

Genomics/Science of WM

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What is Waldenström Macroglobulinemia?

- WM is a rare plasma cell cancer with ~1,400 cases diagnosed each year
- IgM-MGUS is precursor condition, conferring a 46-fold higher relative risk for developing WM
- B-cell lymphoproliferative pathophysiology
- Diagnosis of WM made by increased serum IgM and lymphoplasmacytic cell invasion of bone marrow (and/or organs) in conjunction with clinical symptoms

Groves, et al. Cancer 1998;82. Stone, et al. Haematologica 2011;95. Koshiol, et al. Arch Intern Med 2008;168. Owens, et al. Semin Oncol 2003;30.

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Lymphoplasmacytic lymphoma

- A unique lymphoid malignancy
- Monoclonal paraprotein is essential for diagnosis. WM if paraprotein is IgM. Malignant cell and paraprotein cause clinical disease.
- *MYD88 L265P* is present in > 85% of WM cases. Help in diagnosis and may mediate a role in ibrutinib response
- FcγRIIIA (CD 16) and *WHIM-like CXCR4* mutations and their implications

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Diagnostic Criteria

- Bone marrow infiltration with small lymphocytes, plasmacytoid cells, and plasma cells
- Clonal cells are $CD19^+$, $CD20^+$, $sIgM^+$
  - CD5, CD10, CD23 can be expressed in some cases, and doesn't exclude WM
- IgM monoclonal gammopathy of any concentration

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http://www.nccn.org

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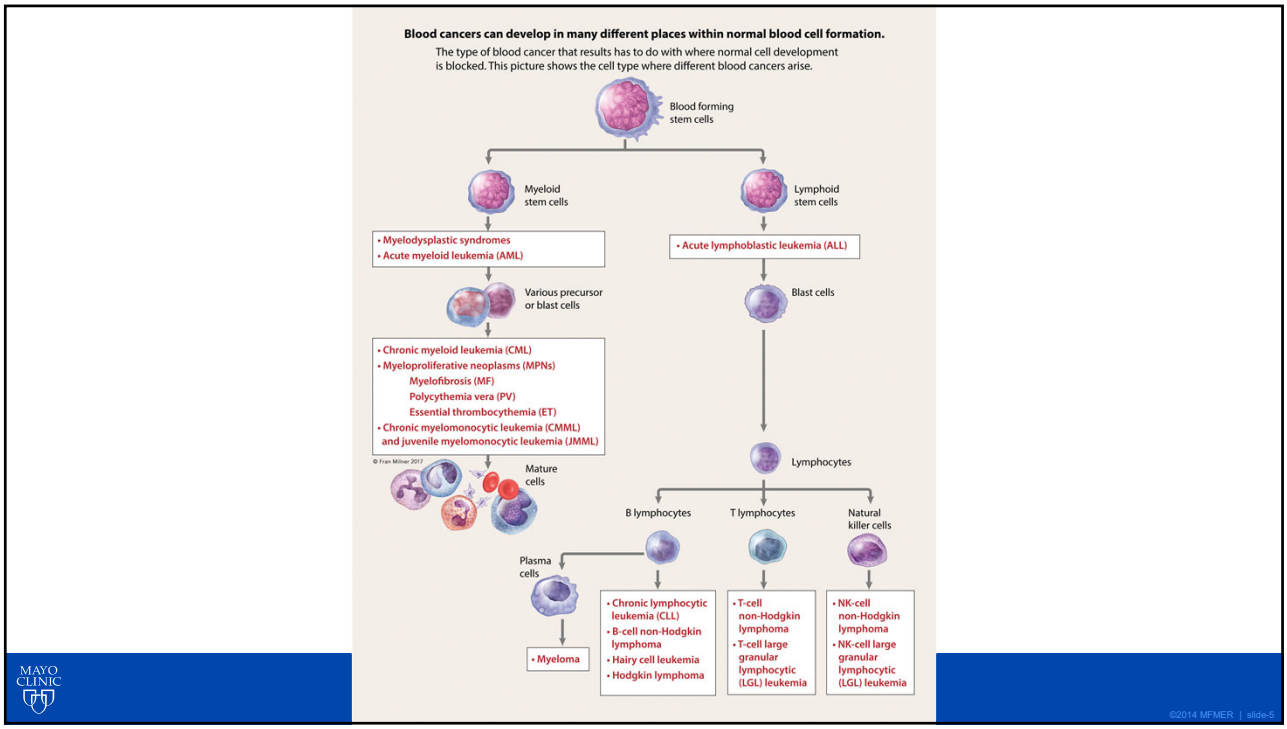
Blood cancers can develop in many different places within normal blood cell formation.
The type of blood cancer that results has to do with where normal cell development is blocked. This picture shows the cell type where different blood cancers arise.
Blood forming stem cells
Myeloid stem cells
Lymphoid stem cells
• Myelodysplastic syndromes
• Acute myeloid leukemia (AML)
• Acute lymphoblastic leukemia (ALL)
Various precursor or blast cells
Blast cells
• Chronic myeloid leukemia (CML)
• Myeloproliferative neoplasms (MPNs)
Myelofibrosis (MF)
Polycythemia vera (PV)
Essential thrombocythemia (ET)
• Chronic myelomonocytic leukemia (CMML) and juvenile myelomonocytic leukemia (JMML)
Lymphocytes
Mature cells
B lymphocytes
T lymphocytes
Natural killer cells
Plasma cells
• Chronic lymphocytic leukemia (CLL)
• B-cell non-Hodgkin lymphoma
• Hairy cell leukemia
• Hodgkin lymphoma
• T-cell non-Hodgkin lymphoma
• T-cell large granular lymphocytic (LGL) leukemia
• NK-cell non-Hodgkin lymphoma
• NK-cell large granular lymphocytic (LGL) leukemia
• Myeloma
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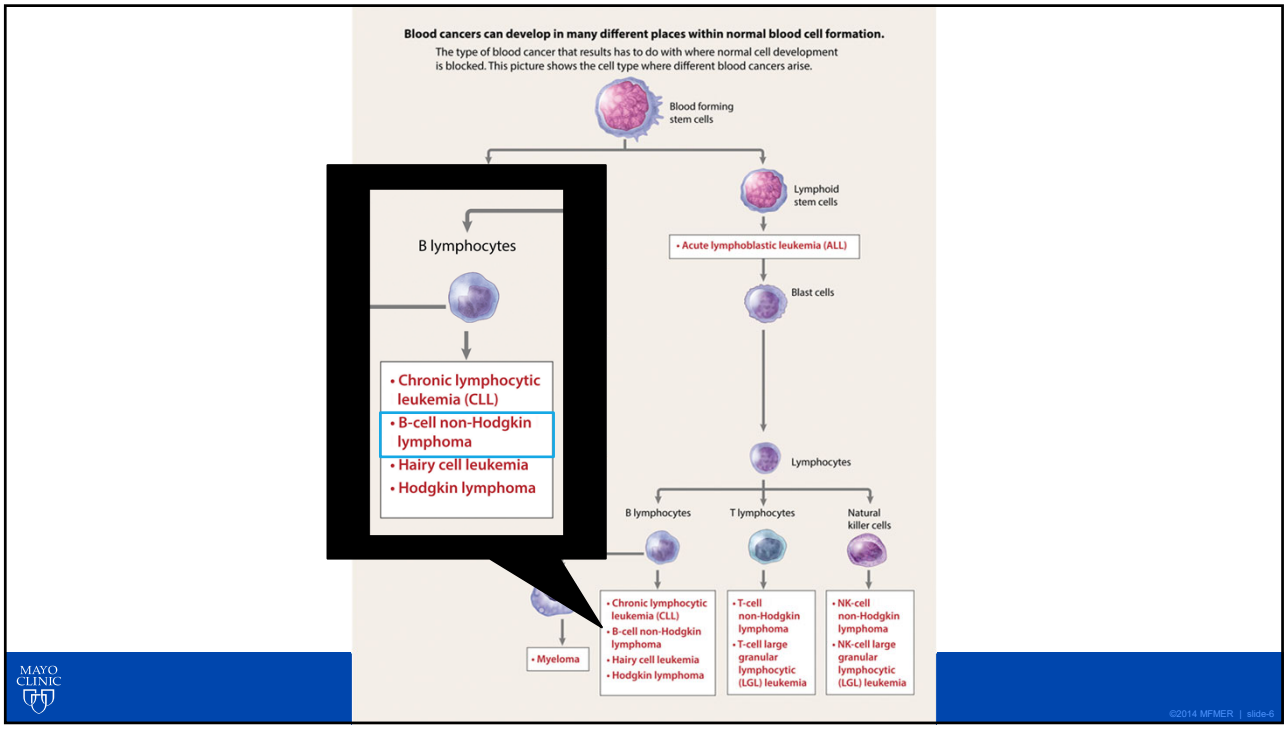
Blood cancers can develop in many different places within normal blood cell formation.
The type of blood cancer that results has to do with where normal cell development is blocked. This picture shows the cell type where different blood cancers arise.
Blood forming stem cells
B lymphocytes
• Chronic lymphocytic leukemia (CLL)
• B-cell non-Hodgkin lymphoma
• Hairy cell leukemia
• Hodgkin lymphoma
Lymphoid stem cells
• Acute lymphoblastic leukemia (ALL)
Blast cells
Lymphocytes
B lymphocytes
T lymphocytes
Natural killer cells
Plasma cells
• Chronic lymphocytic leukemia (CLL)
• B-cell non-Hodgkin lymphoma
• Hairy cell leukemia
• Hodgkin lymphoma
• T-cell non-Hodgkin lymphoma
• T-cell large granular lymphocytic (LGL) leukemia
• NK-cell non-Hodgkin lymphoma
• NK-cell large granular lymphocytic (LGL) leukemia
• Myeloma
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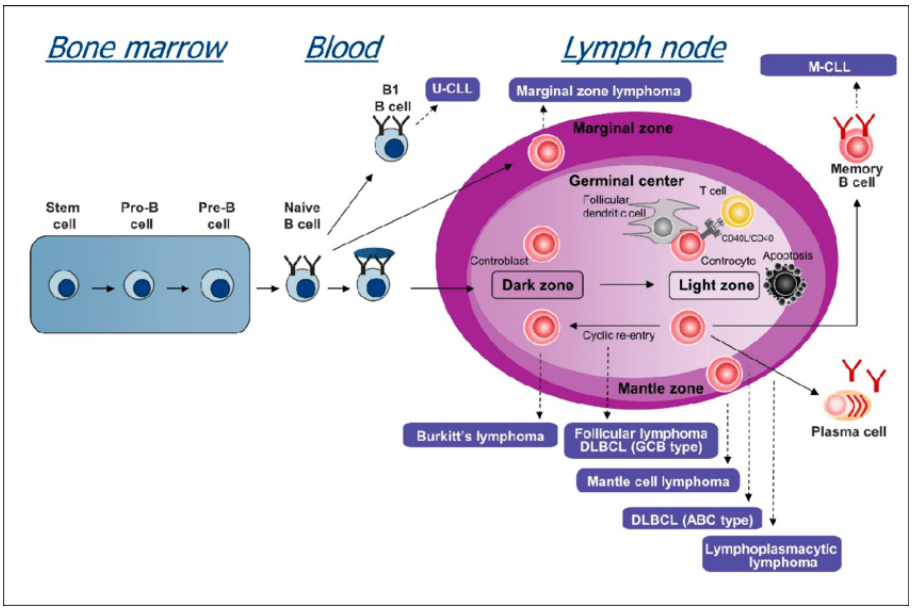

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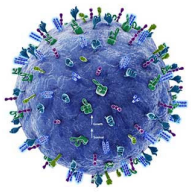
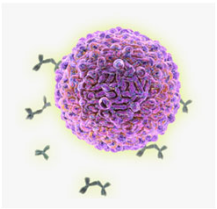
WHO Classification of Waldenstrom's Macroglobulinemia: IgM-Secreting Lymphoplasmacytic Lymphoma



B-Lymphocyte Cells
CD19+, CD20+, CD38-



Plasma Cells
CD19-, CD20-, CD38+

Lymphoplasmacytic Cells
CD19+, CD20+, CD38+/-

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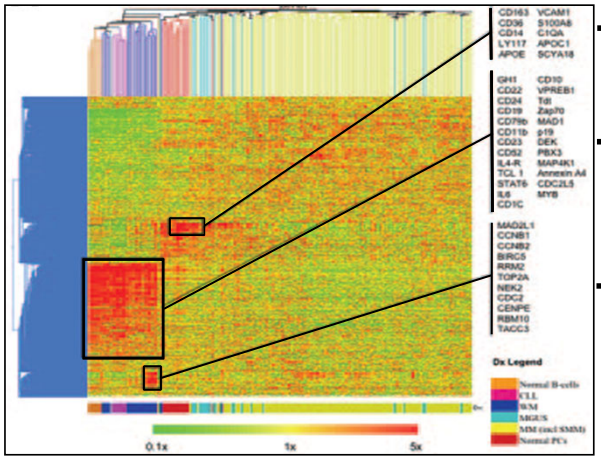
WM Genomics

Gene Expression Profiling (GEP) of WM



→ Normal plasma cells

→ WM & Chronic lymphocytic leukemia

→ Multiple myeloma

WM shares a similar GEP to CLL

Chng and Bergsagel. Blood 2006;108.

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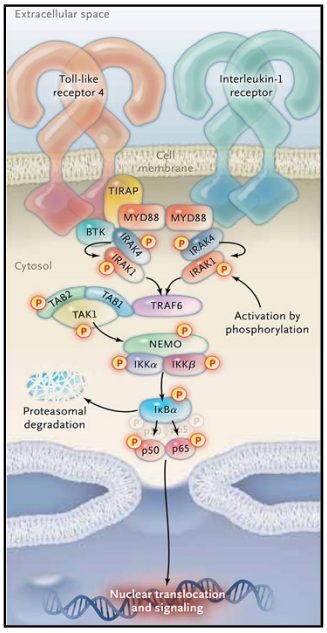
MYD88 and CXCR4

- MYD88 (Myeloid differentiation Primary Response Gene 88)
  - Mediates NF-kB and JAK/STAT activation → increased WM cell growth and survival
  - Overexpressed in ~90% WM patient cells
- CXCR4 Expressed in high levels by WM cells
  - Mediates interaction of WM cells with their microenvironment
  - Activation of downstream cell survival pathways (PI3K/mTOR/AKT)



Treon SP, et al. NEJM 2012;367.

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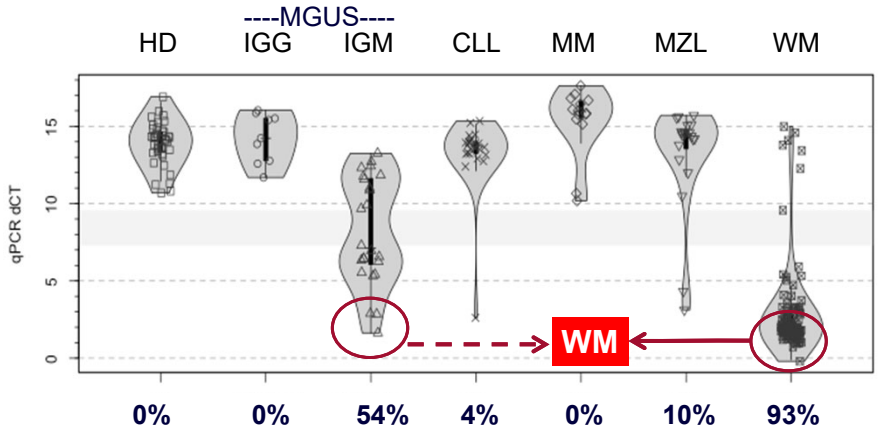
MYD88 L265P by AS-PCR Can Help Distinguish WM From Overlapping Entities



Xu et al. *Blood.* 2013.

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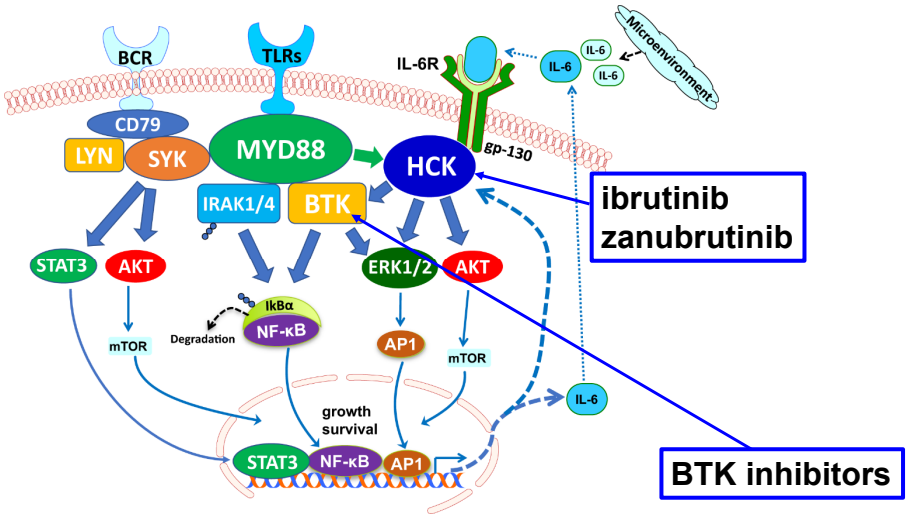
Pro-Survival Signaling by Mutated MYD88 in Waldenstrom's Macroglobulinemia



Yang et al *Blood* 2013;122(7):1222-1232. Yang et al *Blood* 2016;127(25):3237-52. Munshi, Yang et al *BCJ* 2020.

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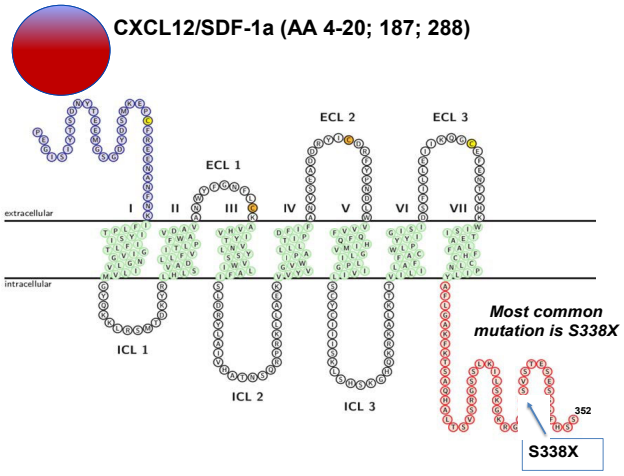
WHIM-like CXCR4 Mutations in Waldenstrom's Macroglobulinemia



Hunter et al. *Blood*. 2013; Rocarro et al. *Blood*. 2014: Xu et al. *Br J Haematol*. 2016; Adapted from Kahler et al. *AIMS Biophysics*. 2016.

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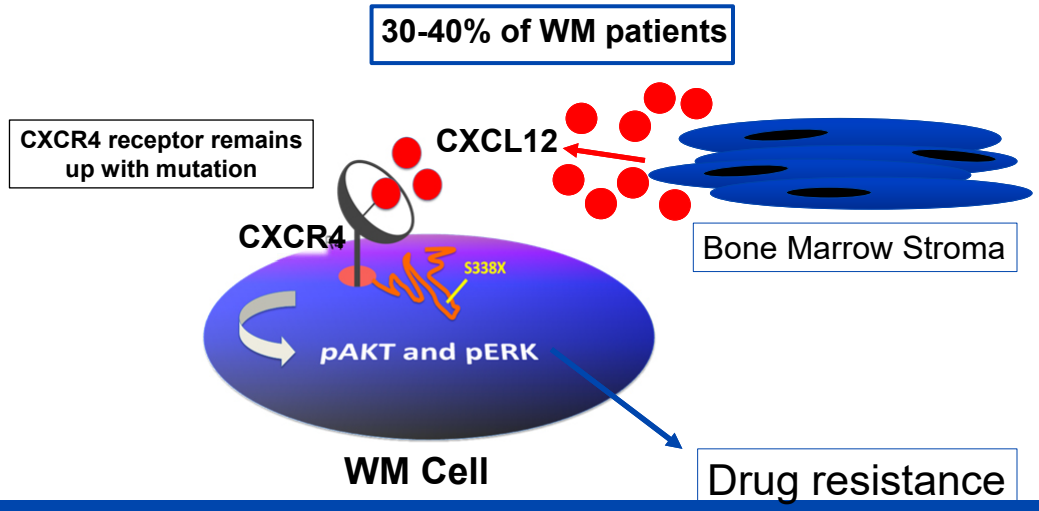
Mutated CXCR4 Permits Ongoing Pro-Survival Signaling by CXCL12



Cao et al. *Br J Haematol*. 2015;168(5):701-707; Roccarro et al. *Blood*. 2014;123(26):4120-4131.

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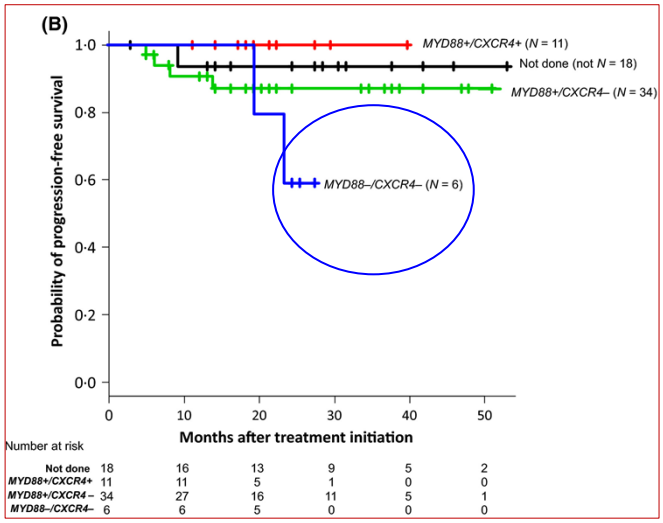
Bendamustine Plus Rituximab in Newly Diagnosed WM Patients – French Innovative Leukaemia Organization (FILO)



Laribi et al. *BJH.* 2018.

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Ibrutinib Activity in Previously Treated WM: Update of the Pivotal Trial (median f/u 59 mos)

	All Patients	MYD88^{MUT} CXCR4^{WT}	MYD88^{MUT} CXCR4^{MUT}	MYD88^{WT} CXCR4^{WT}	*P* Value
N	63	36	22	4	N/A
Overall response rate, n (%)	90.5%	100%	86.4%	50%	<.01
Major response rate, n (%)	**79.4%**	**97.2%**	**68.2%**	**0%**	**<.0001**
Categorical responses					
Minor responses, n (%)	11.1%	2.8%	18.2%	50%	<.01
Partial responses, n (%)	49.2%	50%	59.1%	0%	.03
Very good partial responses, n (%)	**30.2%**	**47.2%**	**9.1%**	**0%**	**<.01**
Median time to response (months)					
Minor response (≥ minor response)	0.9	0.9	0.9	0.9	.38
Major response (≥ partial response)	**1.8**	**1.8**	**4.7**	**N/A**	**.02**

***One patient had MYD88 mutation, but no CXCR4 determination and had SD.**

Updated from Treon et al. *N Engl J Med.* 2015.

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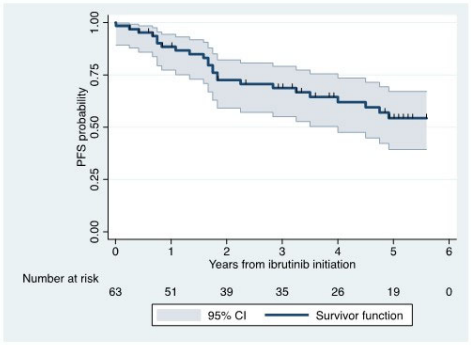
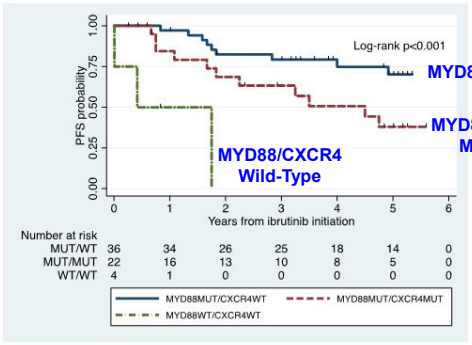
Ibrutinib in Previously Treated WM: Updated PFS

All patients | **MYD88 and CXCR4 Mutation Status**

A.



B.



5 year PFS: 54%
5 year OS: 87%

Updated from Treon et al. *N Engl J Med.* 2015.

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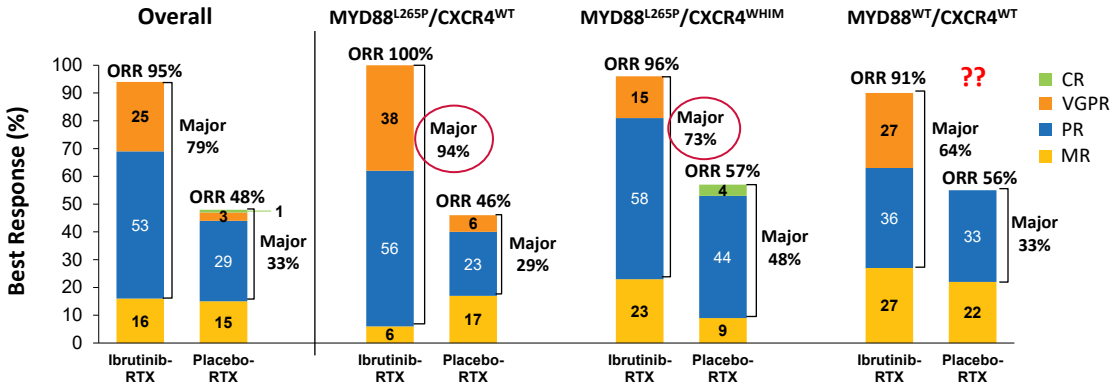
Responses in iNNOVATE Study *(Ibrutinib+Rituximab vs. Placebo+Rituximab)*: Update



[a]Following modified 6th IWWM Response Criteria (NCCN 2014); required 2 consecutive assessments.

Buske et al. ASH 2018. Abstract 149 (oral presentation).

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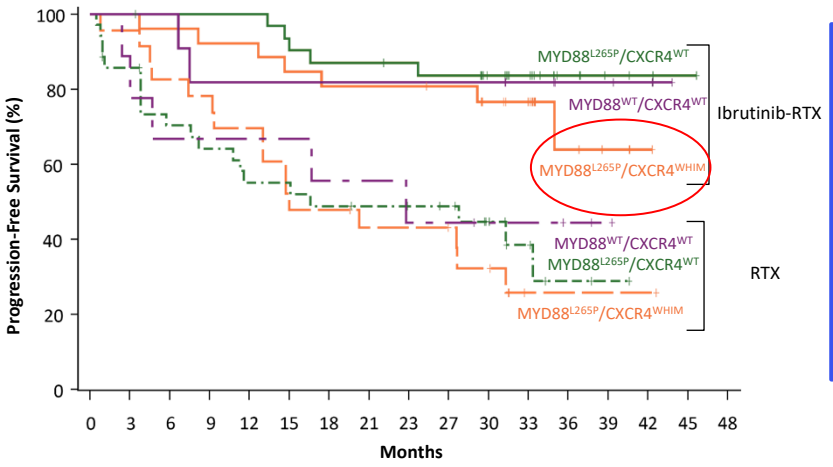
Progression-Free Survival Benefit: Impact of MYD88/CXCR4 Genotype



- Improved PFS with ibrutinib
- 36-month PFS rates
  - MYD88^{L265P}/CXCR4^{WT} 84% vs 29%
  - MYD88^{L265P}/CXCR4^{WHIM} 64% vs 26%
  - MYD88^{WT}/CXCR4^{WT} 82% vs 44%

Buske et al. ASH 2018. Abstract 149 (oral presentation).

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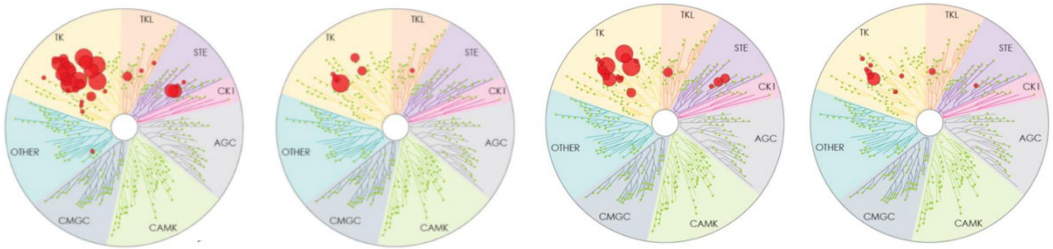
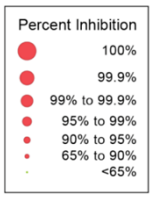
Covalent BTK Inhibitors in WM



Ibrutinib | Acalabrutinib | Zanubrutinib | Tirabrutinib



	IC$_{50}$/EC$_{50}$ (nM)				
	acalabrutinib	ibrutinib	spebrutinib	zanubrutinib	tirabrutinib
BTK	5.1 ± 1.0	1.5 ± 0.2	2.3 ± 0.5	0.5 ± 0.0	5.6 ± 1.0
TEC	126 ± 11	10 ± 12	16 ± 4	44 ± 19	77 ± 7
ITK	>1000	4.9 ± 1.2	24 ± 2	50 ± 5	>1000
TXK	368 ± 141	2.0 ± 0.3	9.1 ± 2.7	2.2 ± 0.6	116 ± 35
BMX	46 ± 12	0.8 ± 0.1	1.6 ± 0.4	1.4 ± 0.4	4.3 ± 0.4
EGFR	>1000	5.3 ± 1.3	199 ± 35	21 ± 1	>1000
ERBB2	~1000	6.4 ± 1.8	>1000	88 ± 26	>1000
ERBB4	16 ± 5	3.4 ± 1.4	49 ± 12	6.9 ± 0.6	991 ± 274
BLK	>1000	0.1 ± 0.0	131 ± 27	2.5 ± 0.4	1133 ± 767
JAK3	>1000	32 ± 15	5.4 ± 1.1	1377 ± 218	>1000
hPBMC	2.9 ± 0.2	0.6 ± 0.0	7.4 ± 0.7	0.9 ± 0.3	6.2 ± 1.9
hWB	9.2 ± 4.4	5.8 ± 3.0	140 ± 85	2.4 ± 0.4	not assessed

BLK, B lymphocyte kinase; BMX, bone marrow tyrosine kinase gene in chromosome X; ERBB2, erb-b2 receptor tyrosine kinase; ERBB4, erb-b4 receptor tyrosine kinase; ITK, interleukin-2-inducible T-cell kinase; JAK3, Janus kinase 3; TEC, tyrosine kinase expressed in hepatocellular carcinoma; TXK, T and X cell expressed kinase.

Kaptein et al. ASH 2018. Abstract 1871.

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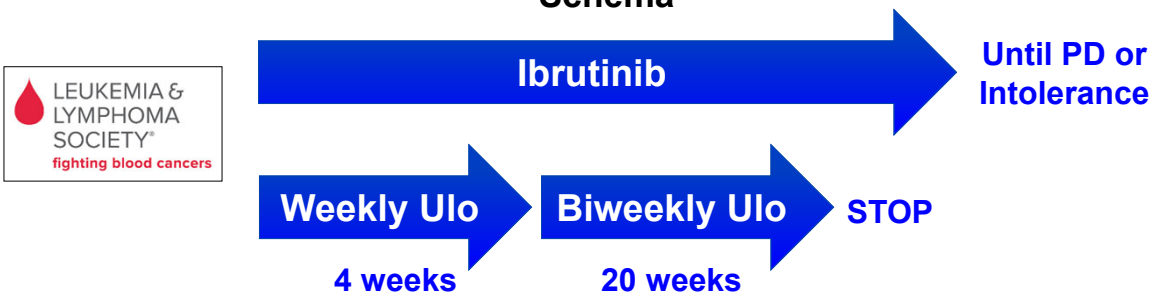
Phase I/II Trial of Ulocuplumab and Ibrutinib in CXCR4-Mutated Patients With Symptomatic WM

Schema



Dose Level	Ibrutinib	Ulocuplumab Cycle 1	Ulocuplumab Cycles 2-6
Level 1 –Starting dose	420 mg PO DQ	400 mg weekly	800 mg every other week
Level 2	420 mg PO DQ	800 mg weekly	1200 mg every other week
Level 3	420 mg PO DQ	800 mg weekly	1600 mg every other week

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ClinicalTrials.gov Identifier: NCT03225716

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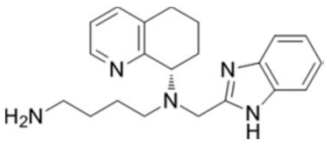
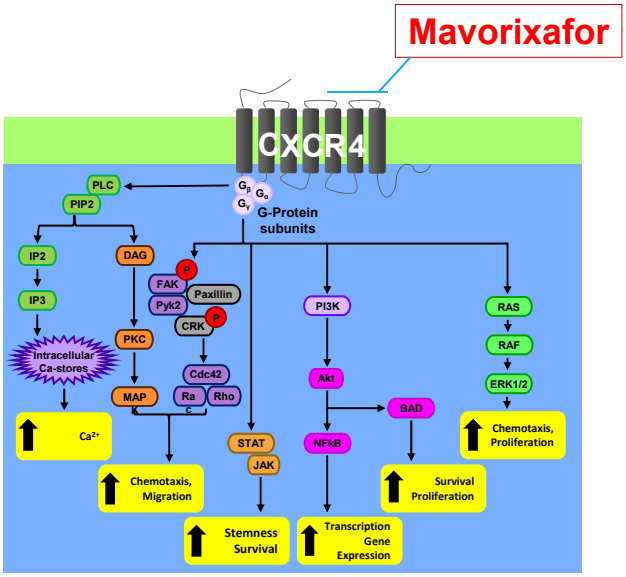
Mavorixafor in Combination With Ibrutinib in CXCR4-Mutated WM



- Non-competitive, allosteric, small molecule antagonist of CXCR4
- Orally Bioavailable; mean $t_{1/2}$ of ~23 hours
- High volume of distribution



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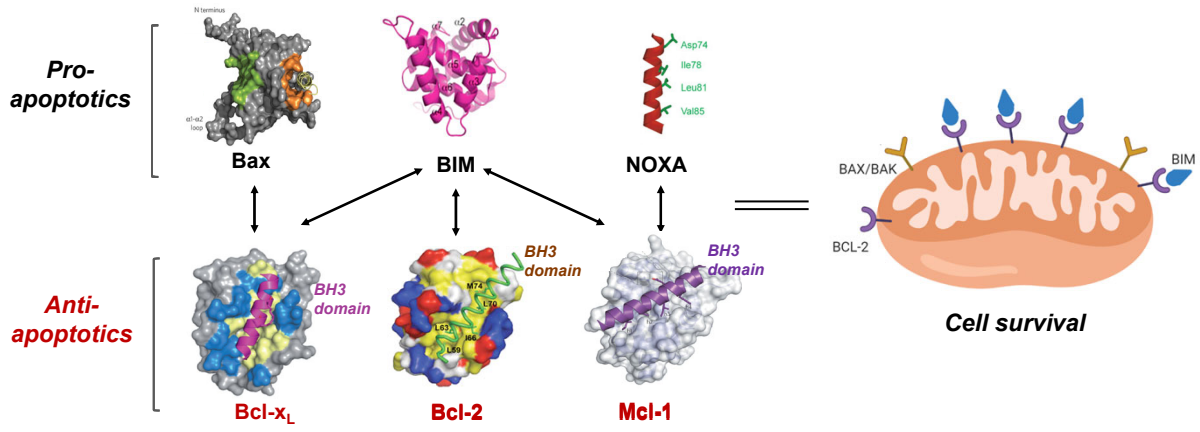
Bcl-2 family proteins: Guardians of the mitochondria



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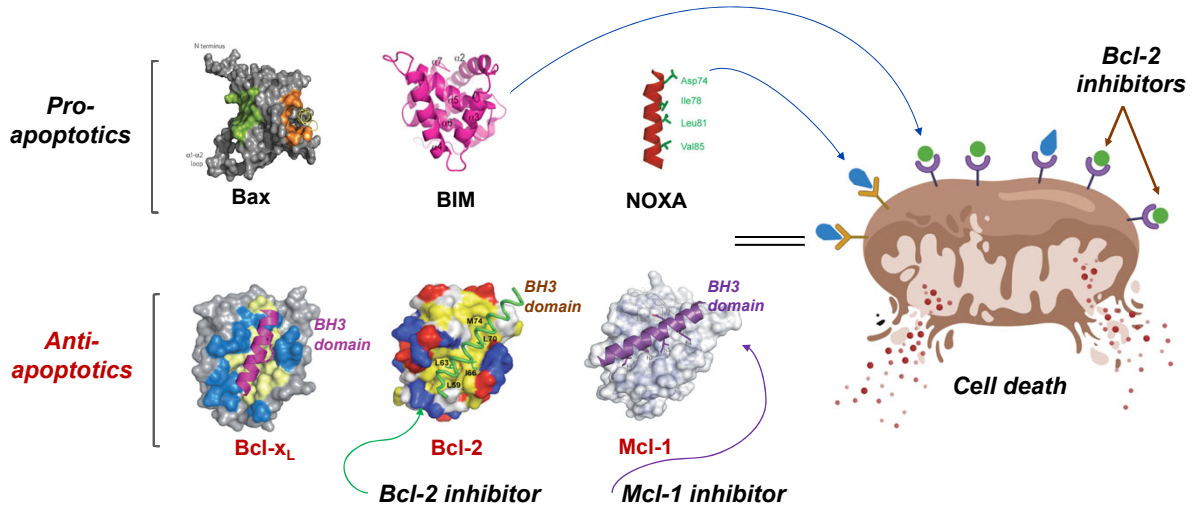
Bcl-2 family proteins: Guardians of the mitochondria



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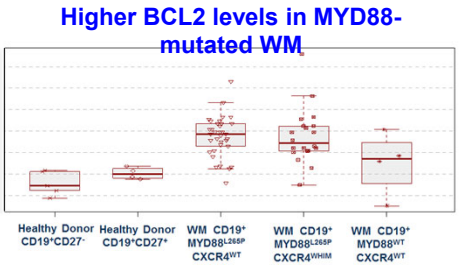
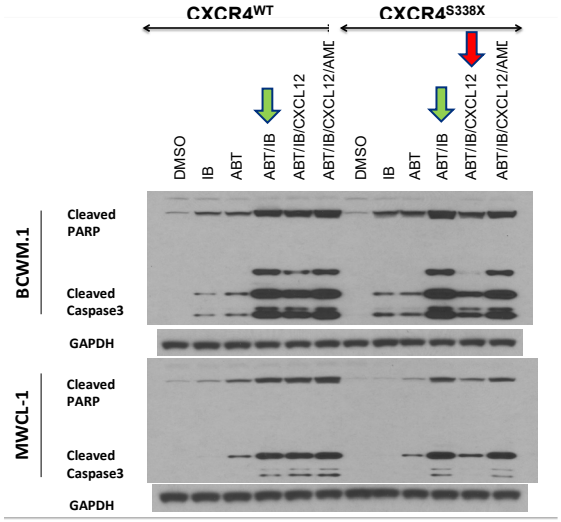
Venetoclax (ABT-199) Augments Ibrutinib-Induced Apoptosis





Cao et al. *BJH*. 2015;168(5):701–707.

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Phase II Study of Venetoclax in Previously Treated WM

	All patients	Prior BTK inhibitor		CXCR4 mutations	
Response	N = 31	No (n = 15)	Yes (n = 16)	No (n = 14)	Yes (n = 17)
Overall (≥ minor)	27 (90%)	14 (93%)	13 (81%)	13 (93%)	14 (82%)
Major (≥ partial)	25 (83%)	13 (87%)	12 (75%)	12 (86%)	13 (76%)
Very good partial	6 (20%)	5 (33%)	1 (6%)	4 (29%)	2 (12%)
Partial	19 (63%)	8 (54%)	11 (69%)	8 (54%)	11 (69%)
Minor	2 (7%)	1 (6%)	1 (6%)	1 (6%)	1 (6%)
Time to response	1.9 months	1.1 months	3.8 months	1.3 months	2.1 months

BM involvement
At baseline, median 40% (4-95%).
At best response, median 3% (0-50%).

Castillo et al. 17th IMW 2019.

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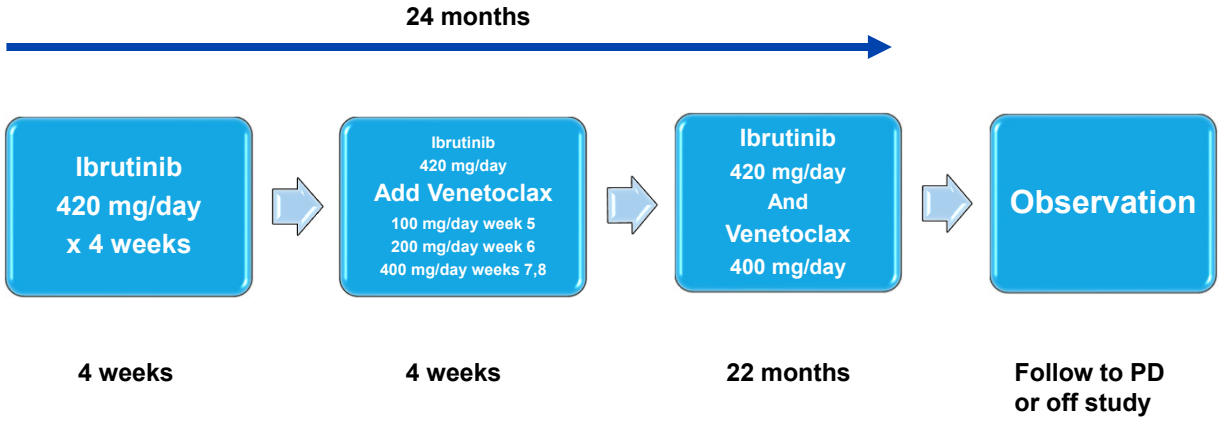
Ibrutinib and Venetoclax in Treatment-Naive WM

Ongoing Clinical Trial at Dana Farber Cancer Institute



Personal communication

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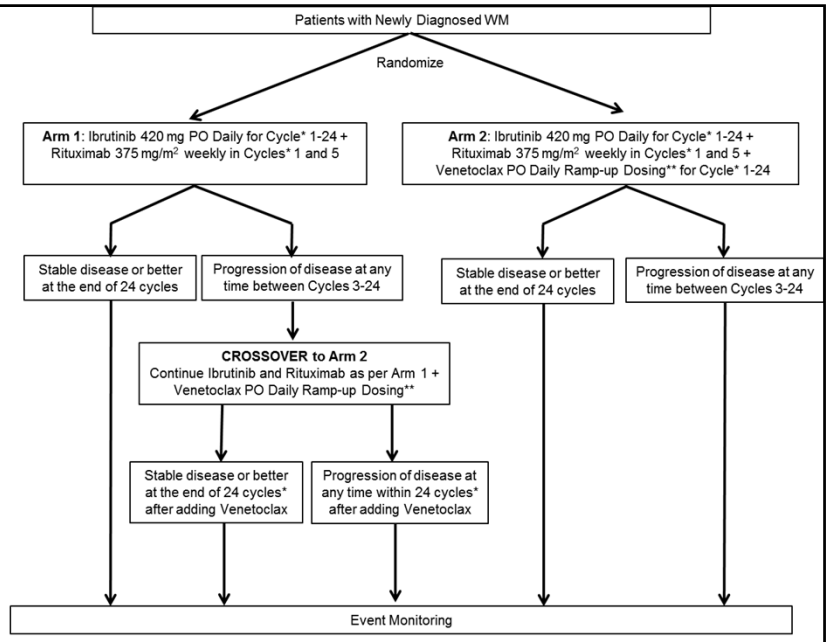
Ibrutinib and Venetoclax in Treatment-Naive WM

NCT0480602



*Cycle=28 Days
**Venetoclax Ramp-up dosing:
Cycle 1 Day 1-7: Venetoclax 200 mg PO daily, Cycle 1 Day 8-14: Venetoclax 400 mg PO daily, Cycle 1 Day 15-28
Cycle 2 onwards: Venetoclax 800 mg PO daily (or 400 mg PO daily, if 800 mg not tolerated)

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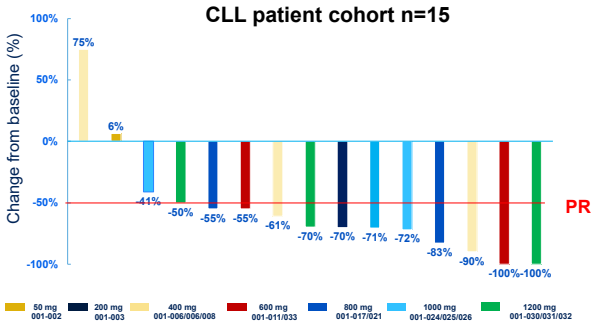
Phase I clinical trial of APG-2575 (NCT03537482)

Baseline characteristics of patients

Age, yr	
Median (range)	70.0 (39–89)
≥ 70, no. (%)	19 (52.8)
Gender, no. (%)	
Male	26 (72.2)
Female	10 (27.8)
Type of cancer, no. (%)[b]	
CLL/SLL	15 (41.7)
MM	6 (16.7)
Follicular lymphoma	5 (13.9)
WM	5 (13.9)
Acute myeloid leukemia	1 (2.8)
Mantle-cell lymphoma	1 (2.8)
Diffuse large B-cell lymphoma	1 (2.8)
Myelodysplastic syndrome	1 (2.8)
Hairy-cell leukemia	1 (2.8)
Median (range) no. of previous therapies	2.0 (1–13)

36 patients



- APG-2575 (lisaftoclax) was well tolerated up to 1,200 mg/day
- MTD not yet reached
- No significant new or unmanageable safety findings

Ailawadhi et al. ASCO 2020.

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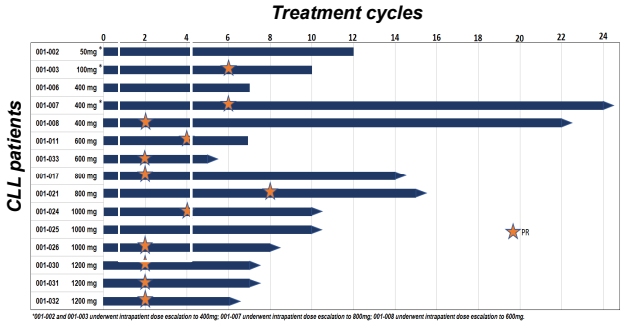
Phase I clinical trial of APG-2575 (NCT03537482)

Cycles to PR (partial remission)



*001-002 and 001-003 underwent intrapatient dose escalation to 400mg; 001-007 underwent intrapatient dose escalation to 800mg; 001-008 underwent intrapatient dose escalation to 600mg.

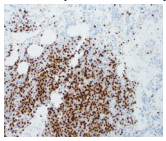
- Most CLL patients achieved PR within 2 - 4 cycles

CLL Patient 008 (APG-2575 400 mg)

Bone marrow

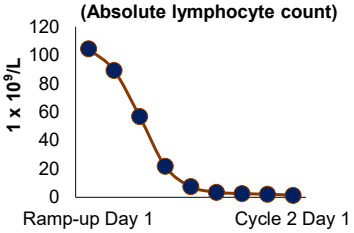
Pre-tx (PAX5 stain)

Pre-tx (PAX5 stain)



Blood

(Absolute lymphocyte count)



Ailawadhi et al. ASCO 2020.

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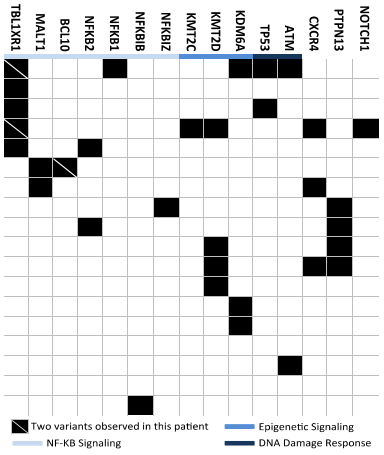
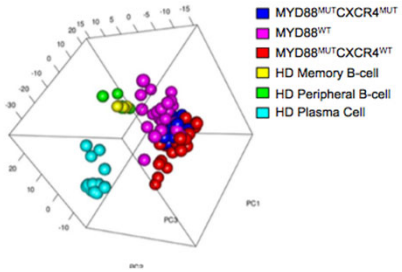
New Driver Mutations Identified in MYD88 WT WM





Principal component analysis of top 500 high variance genes.

Hunter et al. *Blood Adv.* 2018.

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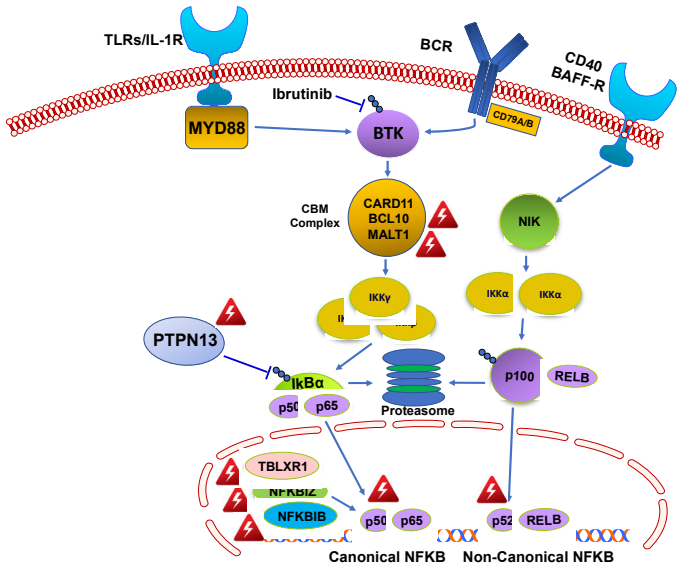
NFKB Signaling Cascades in MYD88 WT WM



Hunter et al. *Blood Adv.* 2018.

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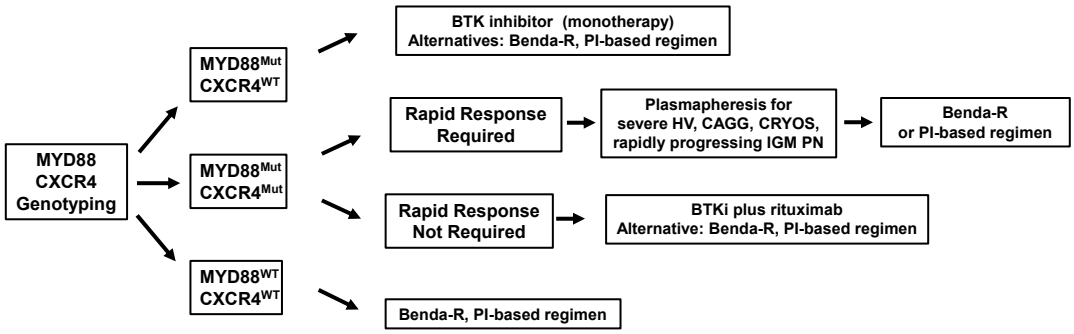
Genomic-Based Treatment Approach to Symptomatic Treatment-Naive WM



- Rituximab should be held for serum IgM ≥4,000 mg/dL
- Benda-R for bulky adenopathy or extramedullary disease
- PI-based regimen for symptomatic amyloidosis, and possible ASCT as consolidation
- Rituximab alone, or with ibrutinib if MYD88^{Mut} or bendamustine for IgM PN depending on severity and pace of progression
- Maintenance rituximab may be considered in patients responding to rituximab-based regimens

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Treon et al. *JCO*. 2020.

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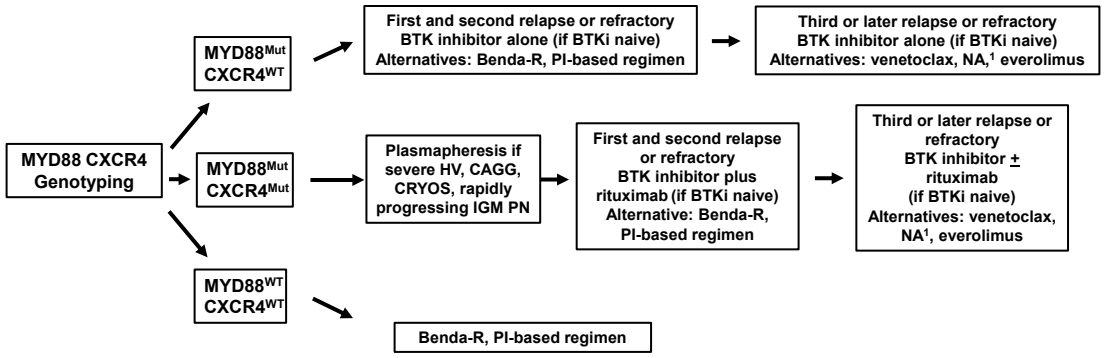
Genomic-Based Treatment Approach to Symptomatic Relapsed or Refractory WM



- Nucleoside analogs (NA) should be avoided in younger patients, and candidates for ASCT[1]
- ASCT may be considered in patients with multiple relapses, and chemo-sensitive disease

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Treon et al. *JCO*. 2020.

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Development of biological tools
(*In vitro and in vivo models of WM)*

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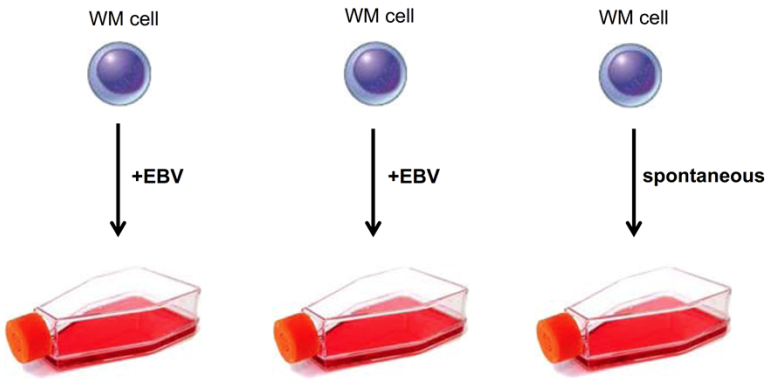
Establishment and characterization of first molecularly validated Waldenstrom macroglobulinemia (WM) cell line



BCWM-1
- MYD88 mutation
- normal karyotype
- no verification

MWCL-1
- MYD88 mutation
- rearranged karyotype resembles patient
- identical IGVH seq

RPCI-WM1
- MYD88 mutation
- rearranged karyotype resembles patient
- identical IGVH seq

Drexler et al, Leuk Lymph 2013.

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Creation of Waldenstrom macroglobulinemia digital avatars using machine-learning and systems biology algorithms exposes novel and clinically relevant therapeutic opportunities

Aneel Paulus MD[1,2], Sharoon Akhtar MPhil[1], Shumail M. Paulus MBBS[1], Yamima Bashir MBBS[1], Hassan Yousaf MBBS[1], Davite Cogen[1], Shireen Vali PhD[3], Ansu Kumar[4], Neeraj K. Singh[4], Shweta Kapoor[4], Taher Abbasi[3], Vivek Roy MD[2], Sikander Ailawadhi MD[2] and Asher Chanan-Khan MD[1,2]

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2. Division of Hematology and Oncology, Mayo Clinic, Jacksonville, FL USA.
3. Cellworks Group, San Jose, CA, USA
4. Cellworks Research India Limited, Whitefield, India

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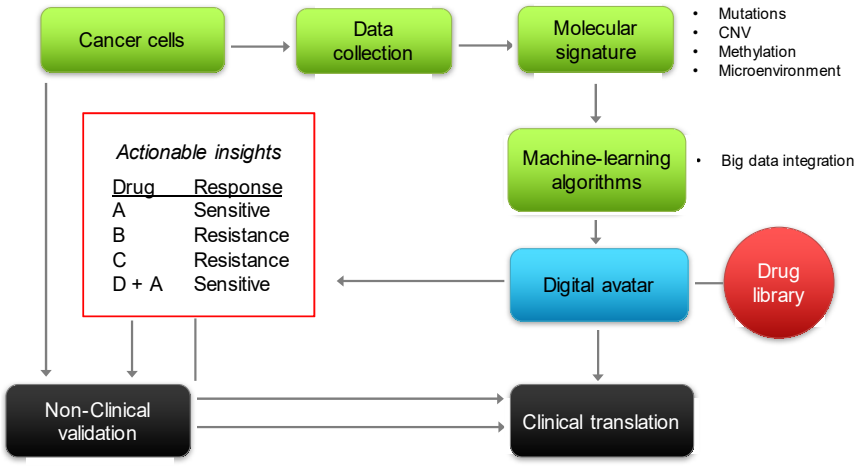
Predictive Modeling Workflow



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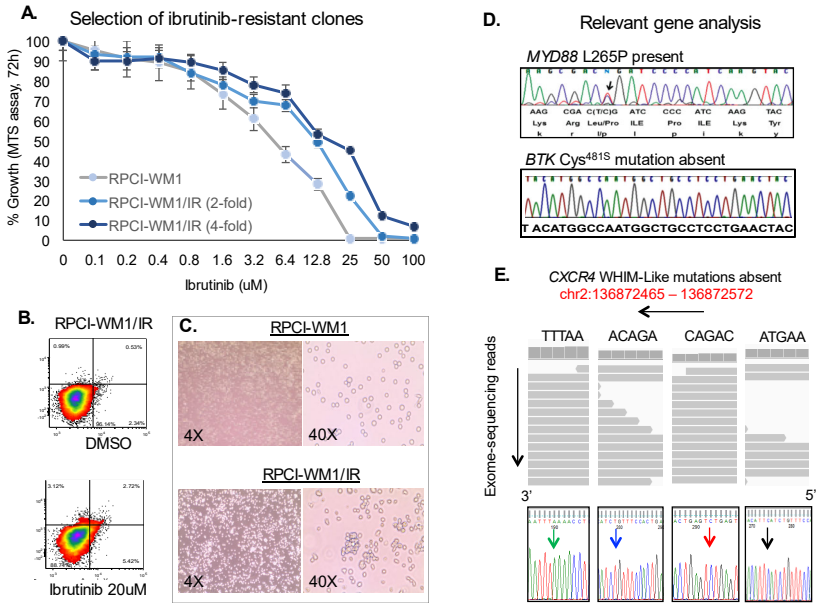
Ibrutinib-Resistant WM Cells as a Biological Validation Tool



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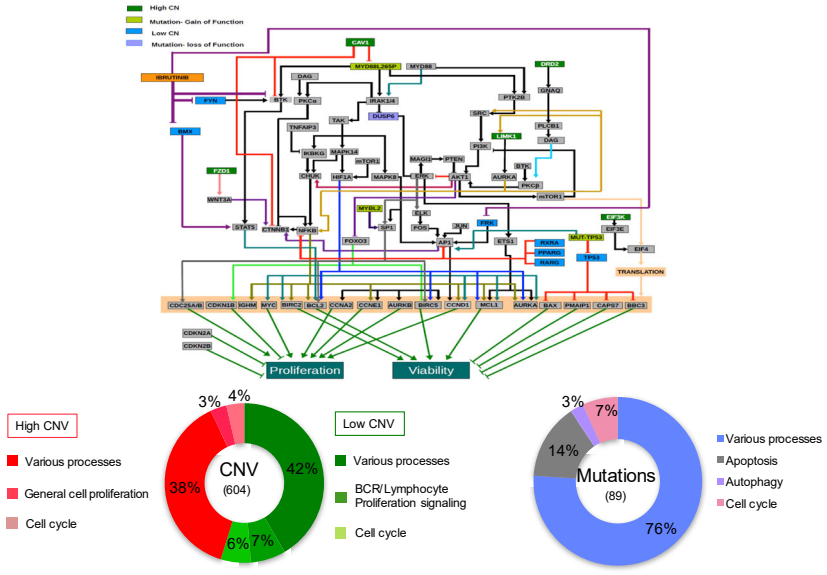
Digital Avatar of Ibrutinib-Resistant RPCI-WM1/IR WM Cells



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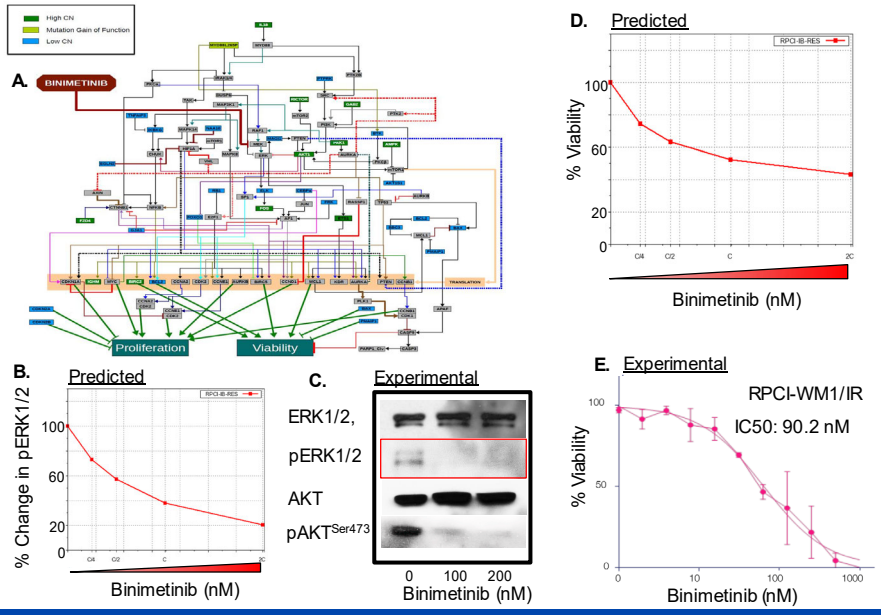
In silico modeling predicts MEK1/2 as a targetable driver of RPCI-WM1/IR cells



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Key Takeaways

- Better understanding of WM biology is shaping our work in drug development and new treatments.
- WM treatment should no longer be a "*one size fits all*" concept.
- Newer studies are focusing on how to change the natural history of WM and get deeper responses / fixed-duration therapy with non-traditional drug approaches.
- Better and more targeted regimens can lead to development of more effective and tolerable agents/regimens for WM treatment.

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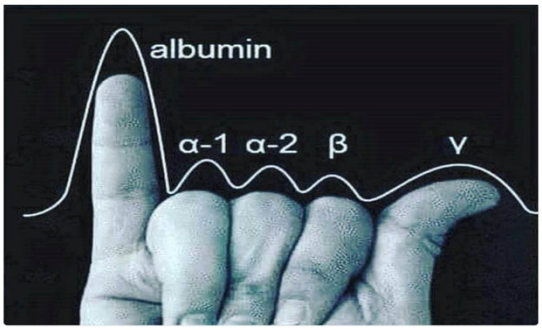

Questions & discussion

ailawadhi.sikander@mayo.edu

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