Mechanisms and Management of Coagulopathy in Acute Promyelocytic Leukemia

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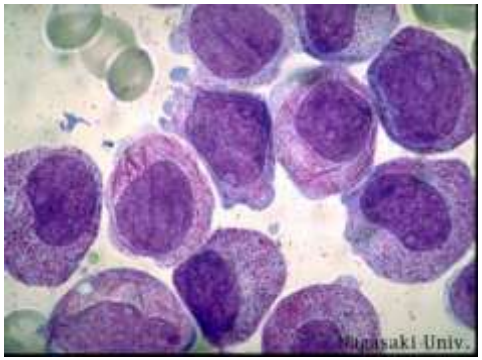
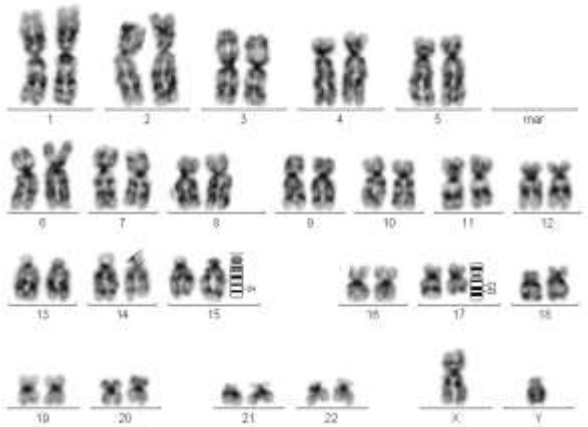
Acute promyelocytic leukemia

Subtype of AML: AML-M3
5 - 15% of all AML
Distinctive:

- **morphology**
- **pancytopenia**
- ***clinical features - coagulopathy***
- **younger age**
- **response to retinoic acid**
- **good prognosis**





Reciprocal translocation
t(15;17)(q22;q21) : *PML-RARα (>95%)*
3 isoforms
Chimeric oncoprotein PML-RARA
(11 other variants) Acta Haematol 2016;136:1–15
Rapidly fatal
Conventional therapy ATRA + chemotherapy
(repeated cycles + maintenance therapy x 2 years

bcr 1	**63.8%**	**50%**
bcr 2	**6.9%**	**5%**
bcr 3	**29.2%**	**45%**

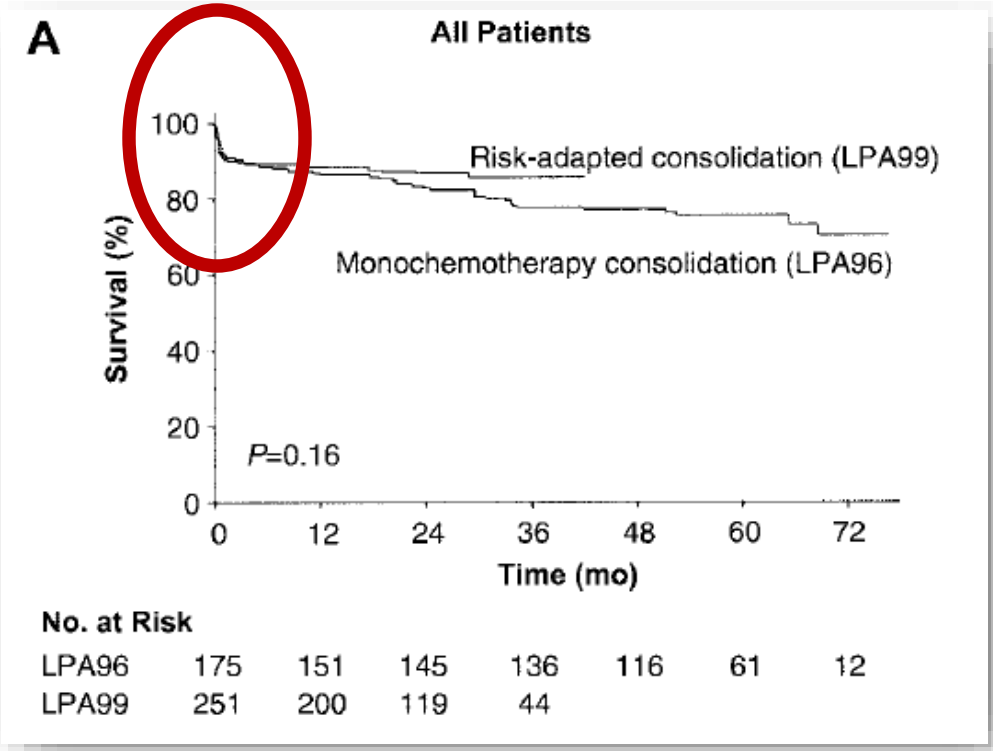
N=72

Mathews V et al. Blood 2006.

Treatment of APML

ATRA + Chemotherapy

- 2 to 3 cycles consolidation
- Two years of maintenance therapy



Risk stratification:

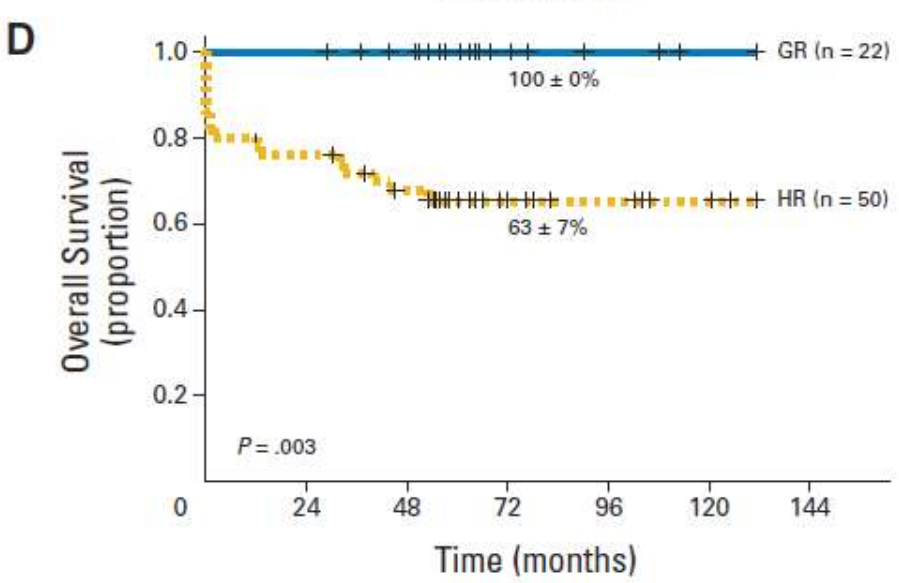
WBC : > 10,000/mm3

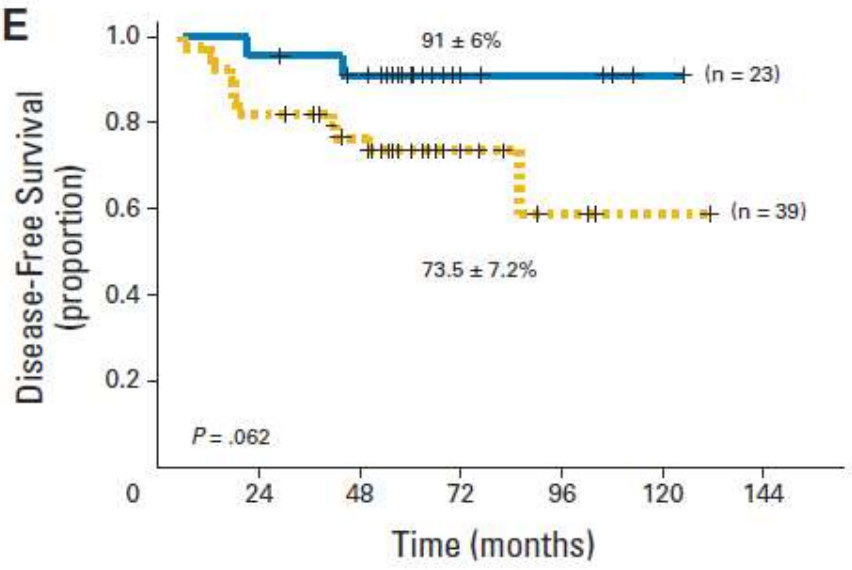
Platelet count : < 40,000/mm3

Sanz et al. Blood 2004

Single agent ATO based regimen:

Median follow up 58 months (5 yr KM estimate±1SE)





Long Term Follow up Data:

Multicenter study - IAPLSG04

7 center's India

RCT : 6 vs. 12 months maintenance

N = 159

5 yr OS	**75%**
5 yr EFS	**69%**

Good Risk Group Relapses = 2

(Only one received an anthracycline in induction)

High Risk Group Relapses = 11

5 year Kaplan-Meier estimate of OS:

LR = 100±0.0%

HR = 63±7.6%

JCO 2010

What we have learnt with this experience

- Very well tolerated regimen
- Cytopenia post induction ~6% (same as GIMMEMA data)
  Post induction out patient treatment
- Most side effects self limiting, no major long term side effects (hepatotoxicity different from GIMMEMA data?).
- Correlation with efficacy and hepatotoxicity (MTHFR A1298C)
- Low risk subset for whom this is apparently adequate
- High risk group increased relapse risk. Inadequate!
- No evidence of exacerbation of coagulopathy
- Neither FLT3-ITD or CTG variations alters prognosis
- No significant long term retention of ATO (JCO).
- No second malignancy to date
- Fertility preserved
- **Cost 1/4th conventional**

Blood 2006
Leukemia 2007
Haematologica 2007
JCO 2010

The NEW ENGLAND JOURNAL of MEDICINE

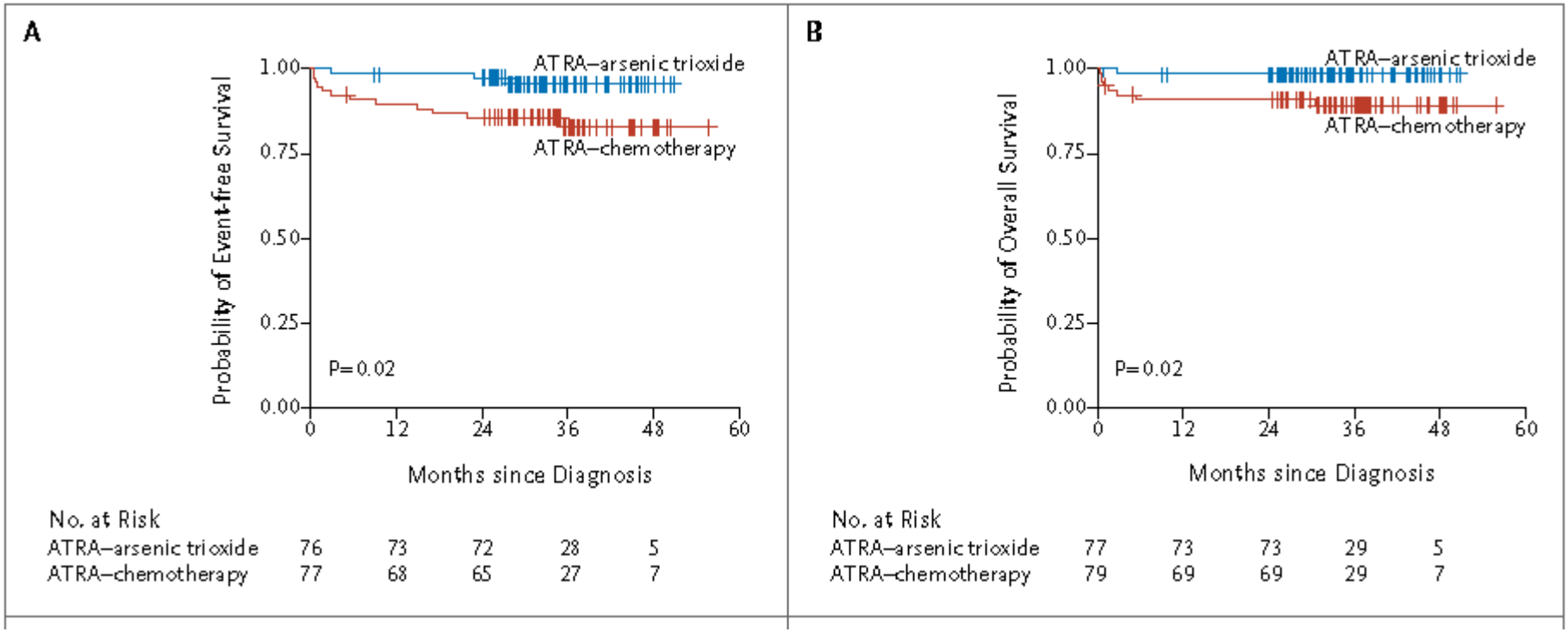
ESTABLISHED IN 1812 JULY 11, 2013 VOL. 369 NO. 2

Retinoic Acid and Arsenic Trioxide for Acute Promyelocytic Leukemia

F. Lo-Coco, G. Avvisati, M. Vignetti, C. Thiede, S.M. Orlando, S. Iacobelli, F. Ferrara, P. Fazi, L. Cicconi, E. Di Bona, G. Specchia, S. Sica, M. Divona, A. Levis, W. Fiedler, E. Cerqui, M. Breccia, G. Fioritoni, H.R. Salih, M. Cazzola, L. Melillo, A.M. Carella, C.H. Brandts, E. Morra, M. von Lilienfeld-Toal, B. Hertenstein, M. Wattad, M. Lübbert, M. Hänel, N. Schmitz, H. Link, M.G. Kropp, A. Rambaldi, G. La Nasa, M. Luppi, F. Ciceri, O. Finizio, A. Venditti, F. Fabbiano, K. Döhner, M. Sauer, A. Ganser, S. Amadori, F. Mandelli, H. Döhner, G. Ehninger, R.F. Schlenk, and U. Platzbecker for Gruppo Italiano Malattie Ematologiche dell'Adulto, the German–Austrian Acute Myeloid Leukemia Study Group, and Study Alliance Leukemia

Phase III prospective trial
Non-inferiority design
Low and Intermediate Risk APL

Outcome estimates: Median follow up 34 months



AK Burnett et al (51)	**RCT: all risk groups. High risk group also received GO**		
ATO+ATRA	119	94%	4 year EFS 91%
Conventional	116	89%	4 year EFS 70%

NEJM 2013

Study	Induction therapy	n	Early Death (ED) n (%)	Early Hemorrhagic Death (EHD) / ED (%)	CR (%)
Pre-ATRA era					
Bernard J *et al* (1973)(38)	Prednisone, Anthracyline, 6-Mercaptopurine and Methotrexate	80	11(13.75)	11/11(100)	23.75
Cordonnier *et al* (1985)(39)	Anthracycline + Cytosine	57	7(12.2)	6/7(85.7)	53
Kantarjian *et al* (1986)(40)	Anthracyclines	60	16(26)	16/16 (100)	53
Cunningham *et al* (1989)(41)	Anthracyclines + Cytosine	57	12 (21)	8/ 12(66.6)	72
ATRA-era					
Fenaux *et al*, 1993 (European APL 91)(42)	ATRA Vs Anthracyline + Cytosine	101	5 (9)	3/5(60)	91 (ATRA arm) 81 (chemo arm)
Tallman *et al* (1997)(43)	ATRA Vs Anthracyline + Cytosine	346	43(12)	22/43(51)	72 (ATRA arm) 69 (chemo arm)
Fenaux *et al* 1999 (European APL)(44)	ATRA vs ATRA + Anthracycline	413	31(7)	10/31(32.2)	95
Mandelli *et al* (1997)(45)	ATRA + Anthracyline	253	11(5)	8/11(72.7)	95
Lengfelder *et al* 2000 (German AML Cooperative Group)(46)	ATRA followed by Thioguanine + Anthracycline + Cytosine	51	4(8)	3/4 (75)	92
PETHEMA group LPA 99 (Sanz *et al* 2004)(47)	ATRA + Anthracycline	426	39 (9.1)	25/39 (64.1)	90
Yanada *et al* (2007)(19)	ATRA + Anthracycline+ Cytosine	279	9(3.2)	8/9 (88.8)	95
Liu *et al* (2010)(48)	ATRA ± ATO	340	50(14.7)	45/50 (90)	84.7
ATO alone/ ATO + ATRA					
Niu *et al*(1999)(49)	ATO	11	1(9.09)	1/1(100)	72.7
Shen *et al* (2004)(50)	ATRA + ATO	61	4(6.5)	2/4 (50)	90
Estey *et al* (2006)(51)	ATRA + ATO	25	4(16)	2/4 (50)	89
Mathews *et al* (2006)(52)	ATO	72	7(9.7)	7/7 (100)	86.1
Ravandi *et al* (2009)(53)	ATRA + ATO ± gemtuzumab ozogamicin	82	7(8.5)	3/7 (42.8)	92
Iland *et al* (2012)(54)	ATRA + Anthracycline + ATO	129	4(3.2)	2/4 (50)	93.2
Lo-Coco et al. NEJM 2013(55)	ATO + ATRA	77	0 (0)	0 (0)	100

Leukemia (2011) 25, 1128–1134

www.nature.com/leu

ORIGINAL ARTICLE

Continuing high early death rate in acute promyelocytic leukemia: a population-based report from the Swedish Adult Acute Leukemia Registry

S Lehmann[1,9], A Ravn[1], L Carlsson[1], P Antunovic[2,9], S Deneberg[1], L Möllgård[1,9], Å Rangert Derolf[3,9], D Stockelberg[4,9], U Tidefelt[5,9], A Wahlin[6,9], L Wennström[4,9], M Höglund[7,9] and G Juliusson[8,9]

[1]Hematology Centre, Karolinska University Hospital, Huddinge, Stockholm and Regional Tumor Registry, Stockholm, Sweden; [2]Department of Hematology and Regional Tumor Registry, Linköping University Hospital, Linköping, Sweden; [3]Center of Hematology and Regional Tumor Registry, Karolinska University Hospital, Solna, Stockholm, Sweden; [4]Department of Medicine and Regional Tumor Registry, Sahlgrenska University Hospital, Göteborg, Sweden; [5]Department of Medicine, Örebro University Hospital, Örebro, Sweden; [6]Department of Radiation Sciences, University of Umeå and Regional Tumor Registry, Norrland University Hospital, Umeå, Sweden; [7]Department of Hematology and Regional Tumor Registry, Academic Hospital, Uppsala, Sweden; [8]Department of Hematology and Regional Tumor Registry, Skåne University Hospital and Lund University, Lund, Sweden and [9]Swedish Acute Myeloid Leukemia Group, Sweden

Registry data
1997-2006
N = 105 (2.7%)
Early death = 30 (29%)
Non early deaths relapses = 16%
Survival 62%

Table 3 Causes of death

Cause of death	*ED patients* (%)
Bleeding total	12 (41)
CNS bleeding	11 (38)
Pulmonary bleeding	1 (3.4)
Cardiac or respiratory failure	5 (17)
Sepsis	3 (10)
Multiorgan failure	2 (6.9)
Suspected DS	1 (3.4)
Cerebral infarction	1 (3.4)
Cerebral leukostasis	1 (3.4)
Unknown	3 (10)

Abbreviations: CNS, central nervous system; DS, differentiation syndrome; ED, early death.

blood

Prepublished online June 8, 2011;
doi:10.1182/blood-2011-04-346437

Early death rate in acute promyelocytic leukemia remains high despite all-trans retinoic acid

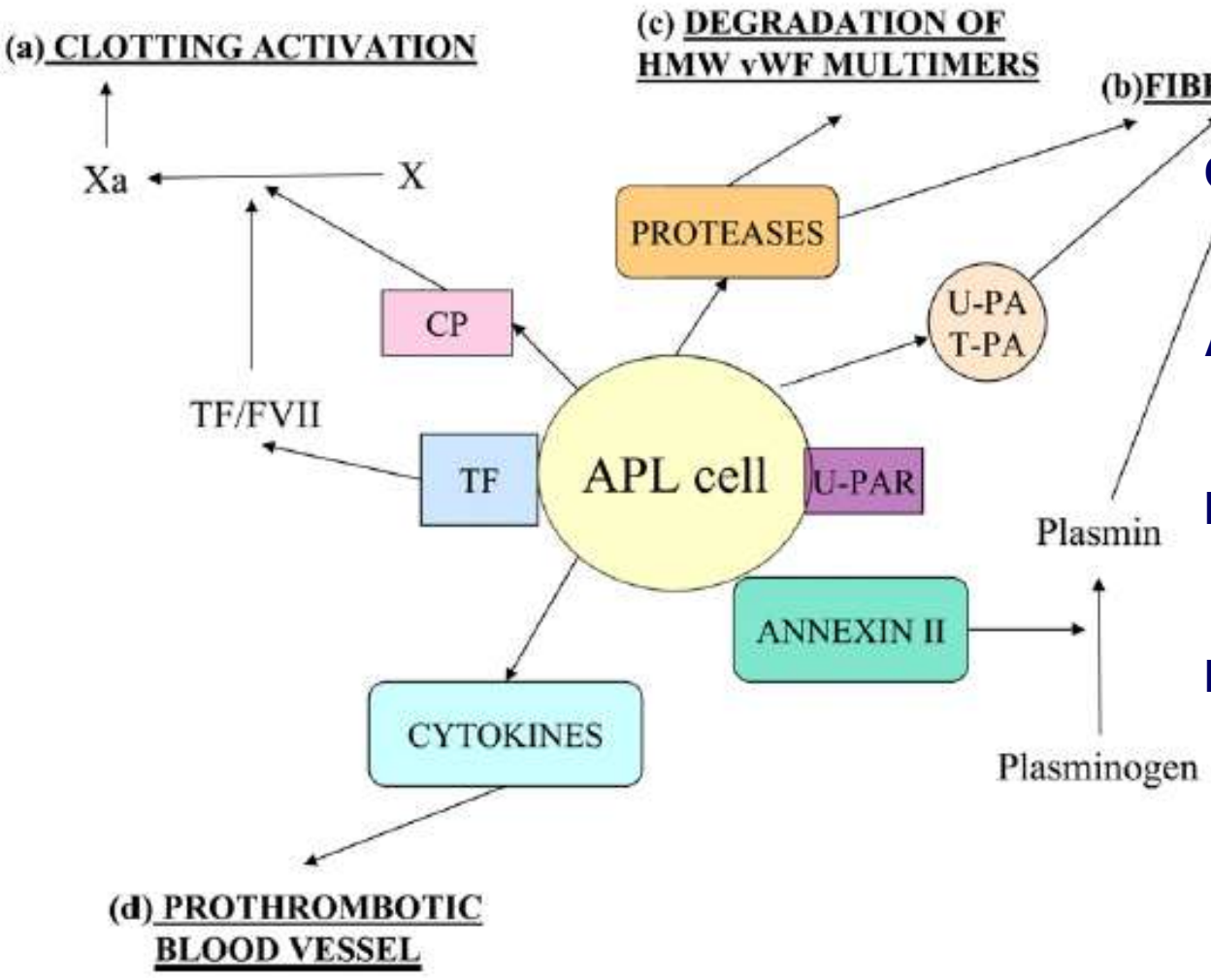
Jae H. Park, Baozhen Qiao, Katherine S. Panageas, Maria J. Schymura, Joseph G. Jurcic, Todd L. Rosenblat, Jessica K. Altman, Dan Douer, Jacob M. Rowe and Martin S. Tallman

N = 1400
1992 – 2007
SEER database
Early death rate
= 17.3%
3 yr OS
= 54.6 – 70%
>55 = 44.6%

Do Early Events Excluding Patients with APL From Trial Enrollment Modify Treatment Result Evaluation? Real-Life Management of 100 Patients Referred to the University Hospital Saint-Louis Between 2000 and 2010.
Jean-Baptiste Micol*,1, Emmanuel Raffoux, MD*,2 et al.
ASH Abstract 2010

Non enrolled = 29%
Early death 21% (vs. 3%)
5 yr EFS 62% vs. 84%

APL cell interactions with the haemostatic system



Chemotherapy induced increased ROS activity

↓

Activation of NF-Kb pathway

↓

Increased TF expression

↓

Potential paracrine effect of ROS on mononuclear cells and endothelial cells

Blood Reviews (2006) 20, 289–297
Journal of Immunology 2000

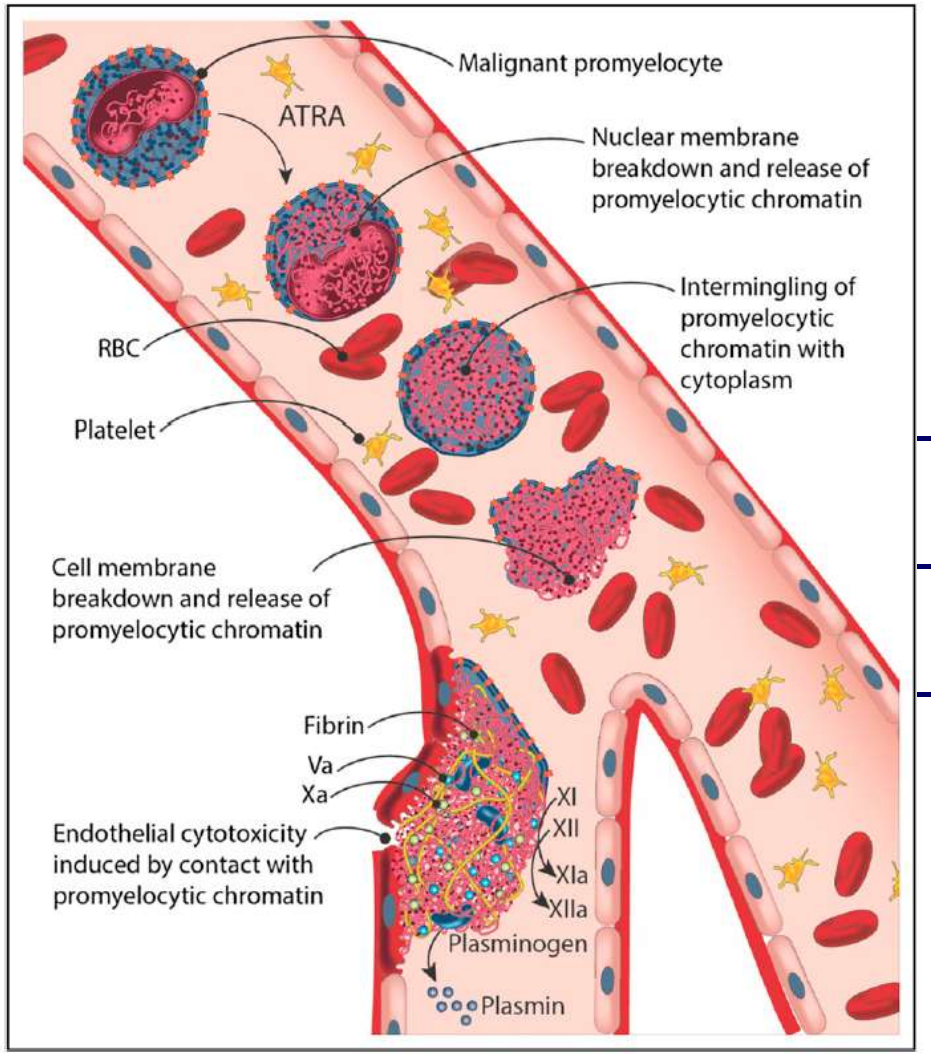


Illustration of mechanism and effects of ETosis in malignant promyelocytes following exposure to ATRA. Malignant promyelocytes on exposure to ATRA undergo nuclear and granule membrane breakdown. Subsequently, there is intermingling of chromatin and cytoplasmic contents within the cell. Following this, there is bulging, further weakening, and final breakdown of the cell membrane with release of promyelocytic chromatin, which forms a net-like structure and binds to other cells and endothelial cells. The surface of the extracellular chromatin, along with the surface membrane of the cell from which it arose, concentrates procoagulant factors and fibrin. The extracellular chromatin and cf-DNA also facilitate increased generation of plasmin and activate the intrinsic coagulation cascade. Promyelocytic extracellular chromatin also damages endothelial cells with which they come into contact, leading to a procoagulant phenotype, and provides additional surface area for clot formation and fibrin deposition. Ensuing endothelial cytotoxicity probably also leads to loss of endothelial cell integrity. RBC, red blood cell. Professional illustration by Somersault18:24.

THROMBOSIS AND HEMOSTASIS

Comment on Cao et al, page 1855

APL: Oh! What a tangled web we weave

Vikram Mathews CHRISTIAN MEDICAL COLLEGE, VELLORE

In this issue of *Blood*, Cao et al[1] report on a novel mechanism of coagulopathy in acute promyelocytic leukemia (APL) induced by treatment with all-*trans*-retinoic acid (ATRA).

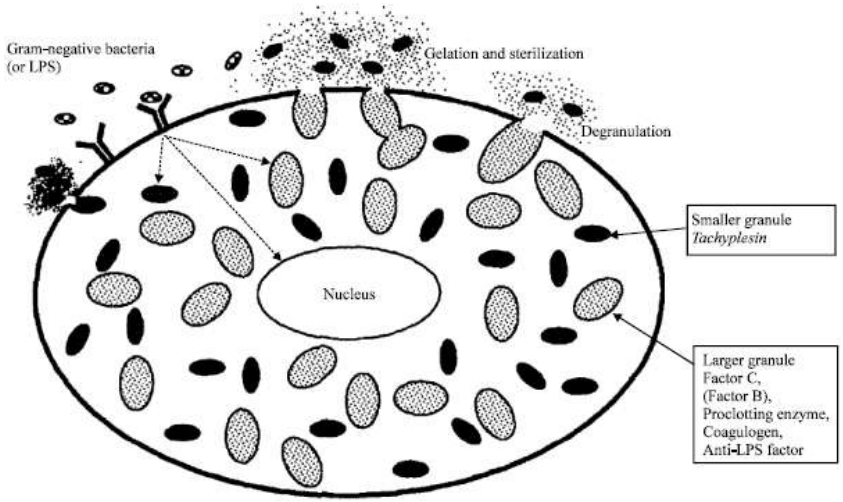
- **ETOSIS – novel cell death pathway neither apoptosis or necrosis**
- **Initially described in neutrophils as a mechanism of bacterial kill**
- **Neutrophil extracellular traps (NET) antimicrobial peptides (AMPs) enzymes (such as bactericidal/permeability increasing protein [BPI], elastases, and cathepsin G)**
- **Bacteria trapped in NET and killed**
- **Localizes effect of toxic enzymes**
- **Similar effect with coagulation proteins – ATRA enhances it**

Primitive coagulation and infection

Marine arthropods
Family Limulidae, suborder Xiphosurida

Blue Blood

Limulus Amebocyte Lysate (LAL) is a lyophilized preparation made from the amebocytes of the **horseshoe crab**. The LAL will clot to form a gel in the presence of endotoxin from gram-negative microorganisms.



Distinctive features APL:

- Exacerbation of coagulopathy with chemotherapy
- Relative preservation of physiological anti-coagulant levels
- Disproportionate decrease in fibrinogen relative to D-dimer
- Prominence of fibrinolysis / fibrinogenolysis over coagulation
- Disproportionate high incidence of IC bleed
- Bleeding often disproportionate to laboratory parameter abnormalities

Clinical Features:

- Up to 76% have some clinical evidence of bleeding at presentation[1]
- Majority limited to muco-cutaneous
- Currently in clinical trial settings EHD <10%
- Multi-organ failure due to micro-vasculature thrombosis as in sepsis related DIC is unusual
- Thrombosis at presentation in up to 10% at presentation[2,3]
- Post-mortem incidence of thrombosis ~25%[4]

1. Avvisati G et al. LAP 0389 data. Blood. 2002;100(9):3141-6
2. Escudier SM et al. Leuk Lymphoma. 1996;20(5-6):435-9
3. De Stefano V et al. J Thromb Haemost. 2005;3(9):1985-92
4. Polliack A. Am J Clin Pathol. 1971;56(2):155-61

Predictors of bleeding and thrombosis

- None of the conventional bleeding parameters either alone or in combination are diagnostic or predictive of bleeding
- Predictors of bleeding:
  - High WBC count at diagnosis[1,2,3]
  - Poor performance status[3]
  - Elevated serum creatinine[2]
  - Low fibrinogen[1]
- Predictors of thrombosis[4]
  - High WBC count at diagnosis
  - bcr3 isoform
  - CD2 IPT
  - FLT3-ITD mutation

1.Yanada M et al. Eur J Haematol. 2007;78(3):213-9
2. Tallman MS et al. Leuk Res. 2005;29(3):347-51
3. Mantha S et al. Blood 2017. 129(13) 1763
4. Breccia M et al. Leukemia. 2007;21(1):79-83

Global Clot Formation assays

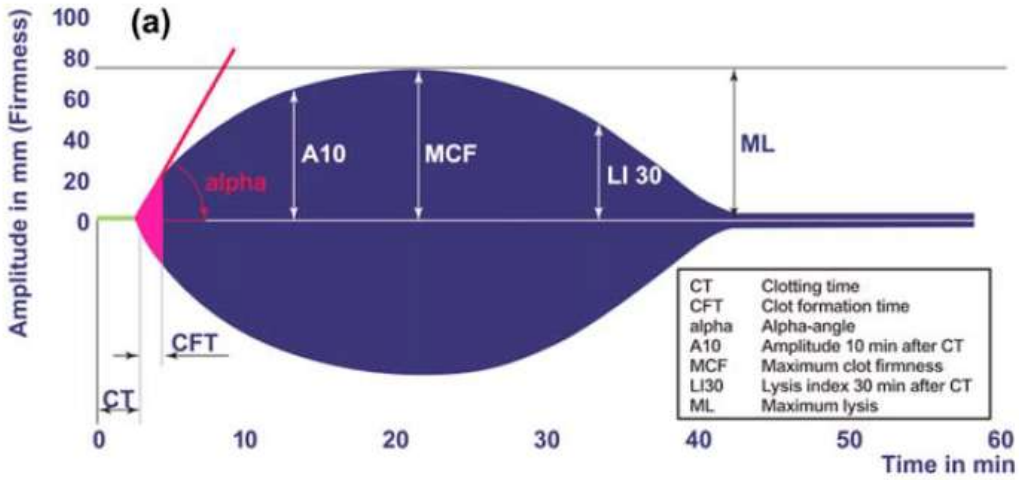
Viscoelastic Methods:

- Thromboelastogram (TEG)
- Rotational thromboelastometry (ROTEM)
- Sonoclot

Setting usually used: (POC)

1. Cardiac surgery
2. Trauma
3. Post partum hemorrhage
4. Liver transplantation

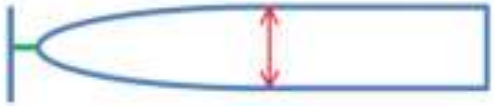
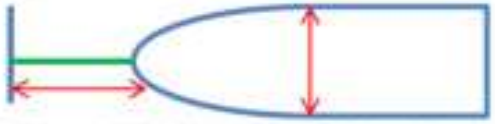
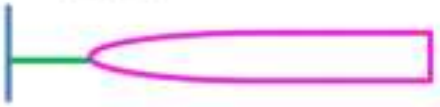
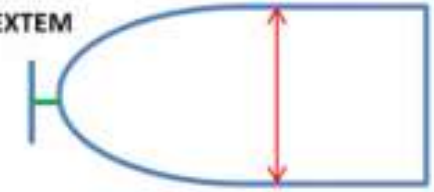
ROTEM in APL



- Complex interplay of different parameters
- Additional functional defects in platelets?
- Impact of micro-particles

Whiting, D. and DiNardo, J. A. (2014), TEG and ROTEM: Technology and clinical applications. Am. J. Hematol., 89: 228–232.

Early ROTEM test as part of initial blood work done as soon as possible

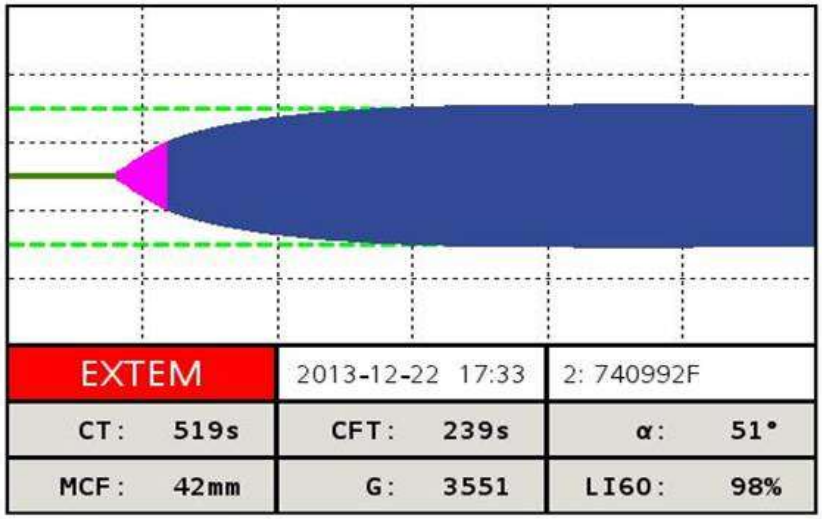
Clinical Decision	Interpretation
Consider administering fibrinogen (*cryoprecipitate or fibrinogen concentrate*)	EXTEM  A10<40mm or MCF<50mm plus FIBTEM  A10<10mm or MCF<12mm; A10<7mm or MCF<9mm
Consider administering plasma (or prothrombin complex concentrate) (*caution ! low platelet and low fibrinogen also prolong CT*)	EXTEM  CT≥ 80 sec AND A10 ≥ 40mm/MCF ≥ 50mm plus FIBTEM  normal A10 or MCF
Consider administering platelets	EXTEM  A10<40mm or MCF <50 mm plus FIBTEM  normal A10 or MCF
Consider administering antifibrinolytic drugs	EXTEM  Any evidence of hyperfibrinolysis FIBTEM  Any evidence of hyperfibrinolysis
Consider withholding transfusions	EXTEM  Abnormal high A10/MCF

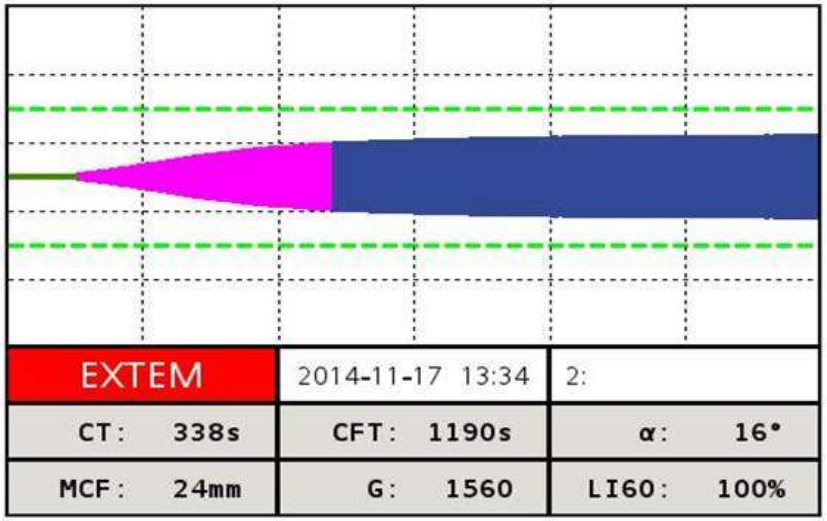
Inaba K, Rizoli S, Veigas PV, Callum J, Davenport R, Hess J, Maegele M; Viscoelastic Testing in Trauma Consensus Panel. 2014 Consensus conference on viscoelastic test-based transfusion guidelines for early trauma resuscitation: Report of the panel. J Trauma Acute Care Surg. 2015 Jun;78(6):1220-9.

Preliminary experience in APL

- On going study
- Preliminary data reported at ASH
- ROTEM assay done at diagnosis prior to any replacement therapy and repeated on day 4
- Extended panel of coagulation parameters also run (TAT complex, Protein C,S, AT-III, PIC complex, tPAIC complex, D-dimer and thrombomodulin)
- Conventional monitoring and replacement therapy
- No intervention based on these parameters

Illustration of discordance between conventional coagulation parameters and ROTEM values at diagnosis in two cases





	UPN 258	UPN 288
WBC (x 10^9/L)	31.5	5.9
Platelet (x 10^9/L)	26	29
PT	24.1	21.3
Sanz risk group	High risk	Intermediate risk
aPTT	17.1	30.7
Fibrinogen	44.2	101.2
Outcome (day)	Alive at 514 days	Died on day 3 – alveolar hemorrhage

Preliminary experience in APL

- N = 50 (40 newly diagnosed, 10 relapsed)
- None of the conventional coagulation parameters at diagnosis were associated with major bleeding, thrombosis or death (univariate)
- ROTEM parameter of maximum clot firmness (MCF) as a continuous variable was significantly associated with death (p=0.012)
- MCF ≤30mm was an independent poor prognostic variable (hazard ratio of 11.89; 95% CI of 1.43 - 98.75, p 0.022)
- 4/6 major bleeding events and 4/5 thrombotic events in MCF ≤30mm

Preliminary experience in APL

Variables	MCF ≤30 (n = 27) N (%) / Median (range) / Mean±SD	MCF > 30 (n=21) N (%) / Median (range) / Mean±SD	p value
Age (years)	40(12-68)	27(9-48)	0.006
Males	12(44.4)	12(57.1)	0.561
Relapsed cases	1(3.7)	9(42.9)	0.001
WBC count ($x10^9$/L)	24(1.1-180.7)	7.11(11.69)	0.001
WBC >10 x $x10^9$/L	17(62.96)	2(9.52)	0.003
Platelet count ($x10^9$/L)	17(4-89)	45(10-339)	0.001
PT in seconds	14.5(11-87.5)	11.8(10.1-14.8)	0.004
aPTT in seconds	28(23.2-77.9)	30.2(22-53.6)	0.104
Fibrinogen in mg%	134.8(24.7-393)	172.2(14.7-549.5)	0.066
Bone marrow blasts(%)	84.68(11.15)	66.38(23.98)	0.002
Major bleeding	4(14.8)	1(4.8)	0.368
Thrombosis	4(14.8)	1(4.8)	0.369
Death	12(44.4)	1(4.8)	0.003

RELAPSED PATIENTS ARE DIFFERENT?

RESEARCH ARTICLE

Comparison of Newly Diagnosed and Relapsed Patients with Acute Promyelocytic Leukemia Treated with Arsenic Trioxide: Insight into Mechanisms of Resistance

Ezhilarasi Chendamarai[1], Saravanan Ganesan[1], Ansu Abu Alex[1], Vandana Kamath[2], Sukesh C. Nair[2], Arun Jose Nellickal[3], Nancy Beryl Janet[1], Vivi Srivastava[4], Kavitha M. Lakshmi[1], Auro Viswabandya[1], Aby Abraham[1], Mohammed Aiyaz[5], Nandita Mullapudi[5], Raja Mugasimangalam[5], Rose Ann Padua[6], Christine Chomienne[6], Mammen Chandy[1], Alok Srivastava[1], Biju George[1], Poonkuzhali Balasubramanian[1], Vikram Mathews[1]*

1 Department of Haematology, Christian Medical College, Vellore, India, 2 Department of Transfusion Medicine and Immunohaematology, Christian Medical College, Vellore, India, 3 Department of Biochemistry, Christian Medical College, Vellore, India, 4 Cytogenetics Unit, Christian Medical College, Vellore, India, 5 Genotypic Technology, Bengaluru, India, 6 UMR 1131 Institut d'Hématologie, Hôpital Saint Louis, I avenue Claude Vellefaux, 75010 Paris, France

PLOS One 2015

- **Possibility of lead time bias**
- **Significantly less bleeding / thrombotic events**
- **Significantly less blood product requirement**

Management

- Early diagnosis and start of treatment with ATRA[1]. When in doubt / Awaiting confirmation?
- Tertiary centre effect (trauma care available)[2]
- Monitor CBC / PT / APTT / Fibrinogen daily at least in first 10-14 days. If bleeding – more frequent as required to target parameters.
- Target platelet count 30 x 10^9/Lt (absence of bleeding), > 50 x 10^9/Lt if bleeding
- Target normal PT / APTT
- Target Fibrinogen > 140 mg%
- Usually coagulopathy resolves in 14 days
- Fluid overload!!

Saravanan

OPEN

Leukemia (2016) **30**, 2169–2178

www.nature.com/leu

ORIGINAL ARTICLE

Rationale and efficacy of proteasome inhibitor combined with arsenic trioxide in the treatment of acute promyelocytic leukemia

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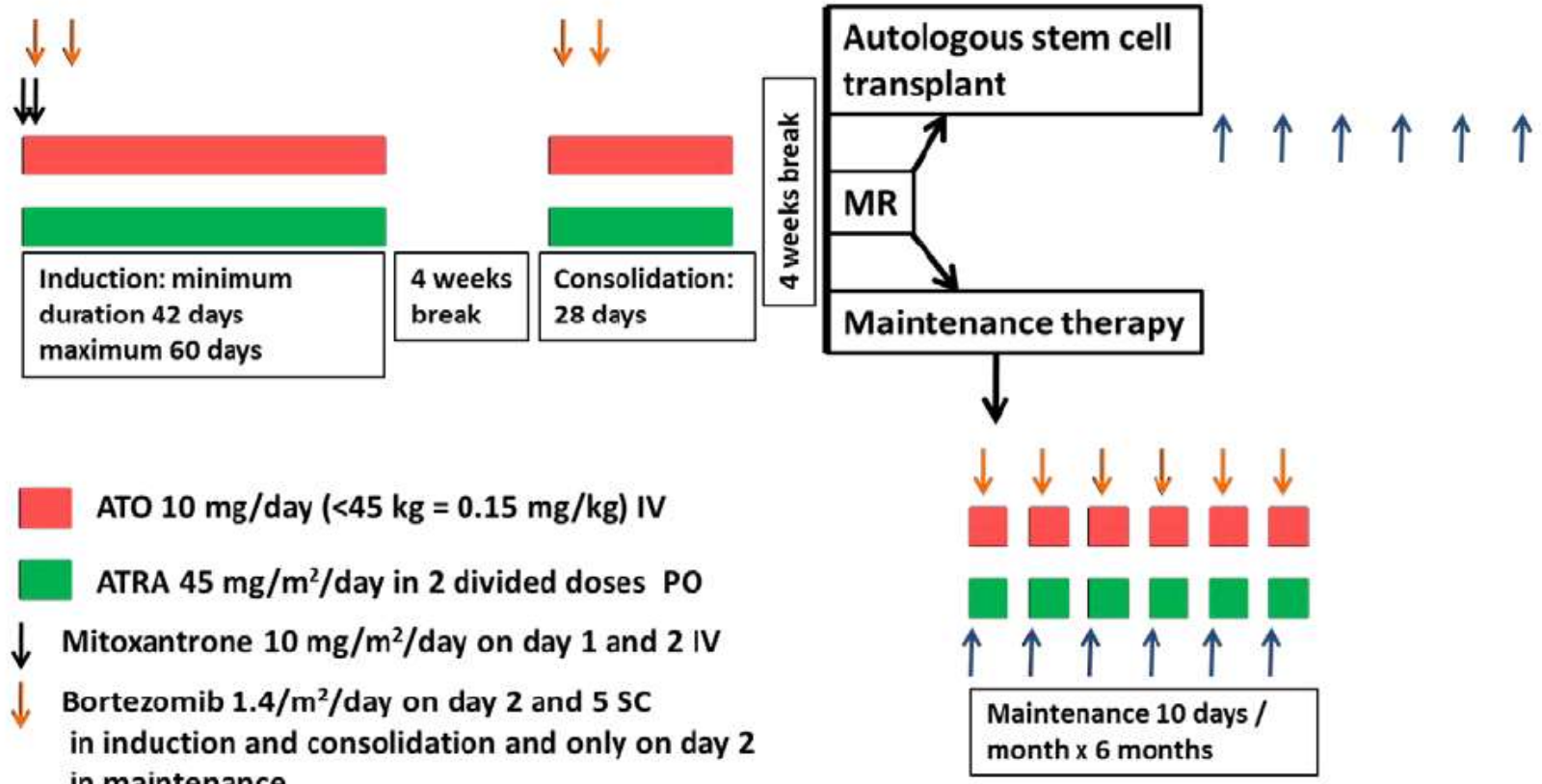
- Demonstrates prominence of the NF-kB pathway in driving EM-DR to ATO in APL

- Establishes the potential to drug this target (pathway)

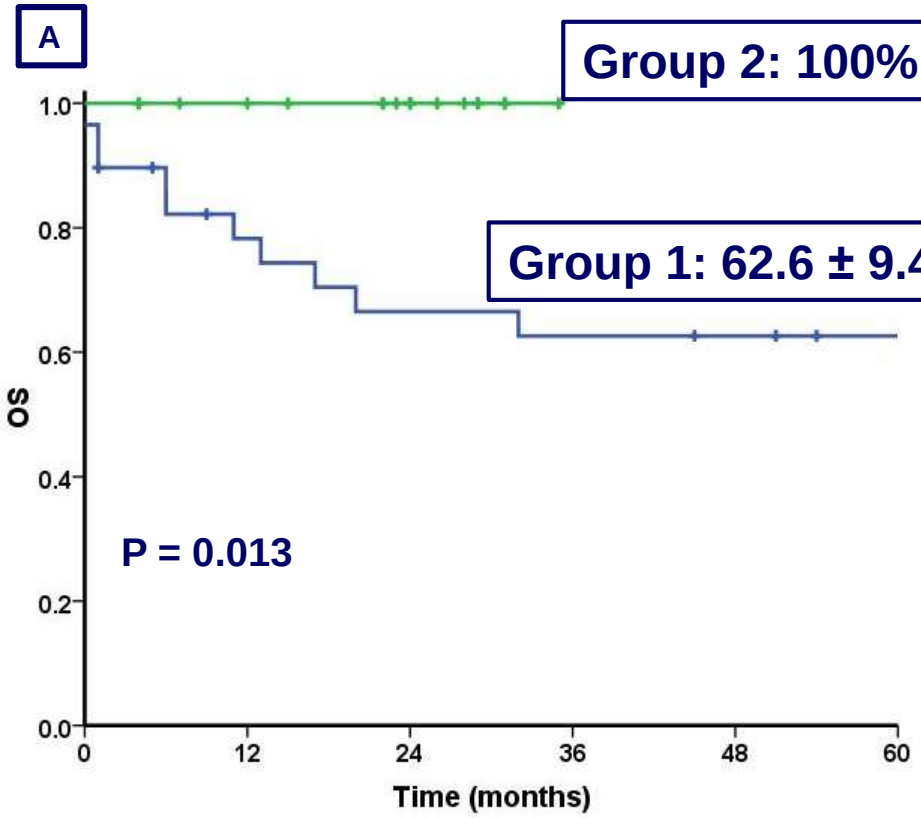
- Highlights potential of bortezomib an FDA approved drug to be re-purposed for this leukemia – and that it can be combined with ATO

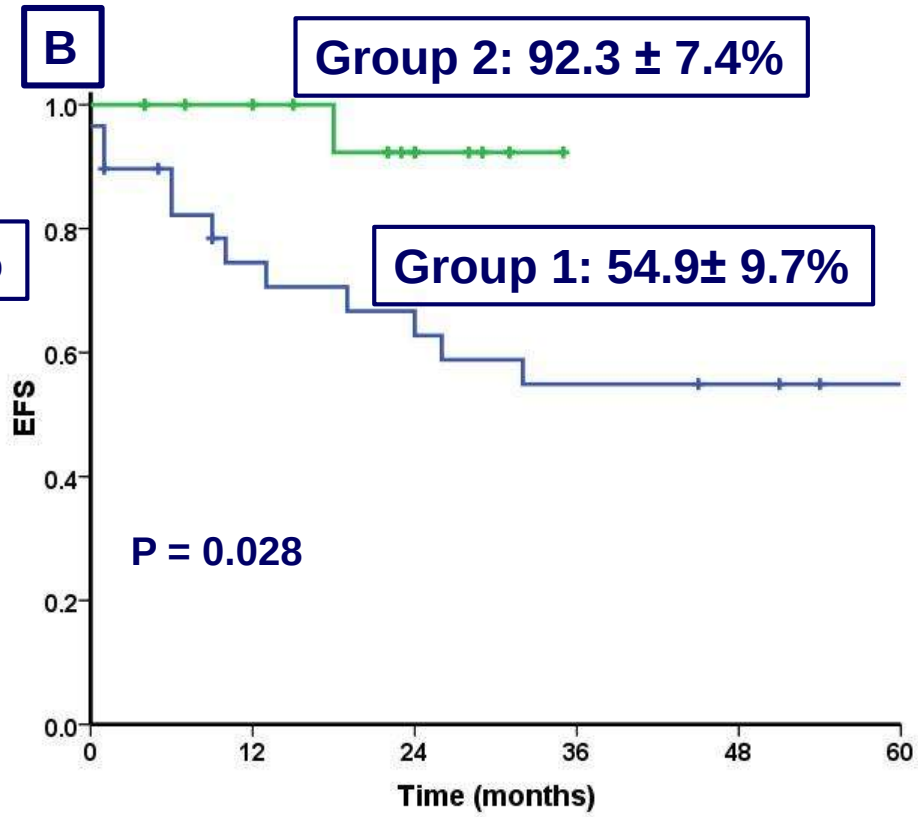
Phase II Clinical Trial: schedule



No prophylaxis for DS
Approach to CNS disease

A. Overall survival B. Event free survival.
Comparison between historical Group 1 and Group II patients enrolled on Phase II Study with additional bortezomib





ASH 2016 ab

Conclusion:

- Significant reduction in coagulopathy
- Significant reduction in consumption of blood bank products.
- Early data suggests reduction in TF, Annexin II, and reduction in Etosis (provisional)
- Potential to reduce incidence of differentiation syndrome (hypothesis)

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