

# **Preemptive and Maintenance Strategies to Prevent Relapse after Allogeneic SCT**

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## Disclosures

Celgene : research grant



# Outline

Relapse

Thoughts on the 'ideal' agent

Focus on myeloid leukemias

Discuss maintenance of remission

Epigenetic manipulation of GVL / GVHD

Most importantly: join us for the protocol development discussion!!

# **CML could be considered the gold standard**

- Reliable marker of disease persistence or recurrence
- Interventions are effective (DLI, TKIs)

# Why prevent relapse ?

1- For obvious reasons !

2- Treatment of relapse post transplant is suboptimal  
(for most diseases)

# **(Some) Strategies for Preventing Relapse**

## 1- Pre-emptive treatment of relapse

- minimal residual disease-based (PCR, flowcytometry etc)

- based on pre-transplant high-risk parameters or on dynamic approaches, such as presence of MRD after HSCT etc

## 2- Maintenance of remission

## 3- Improve conditioning regimen

## 4- Improve the graft

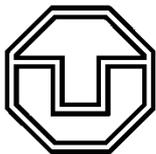
## 5- Select patients.

# **MRD Based Preemptive 5–Aza Treatment in patients with MDS or AML after allogeneic HSCT – Results of the “RELAZA” Trial**

**U. Platzbecker, M. Wermke, J. Radke, A. Kiani, F. Seltmann, C. Röllig, M. v. Bonin, B. Mohr, U. Oelschlägel, J. Schetelig, G. Ehninger, M. Bornhäuser and C. Thiede**

**University Hospital “Carl Gustav Carus”**

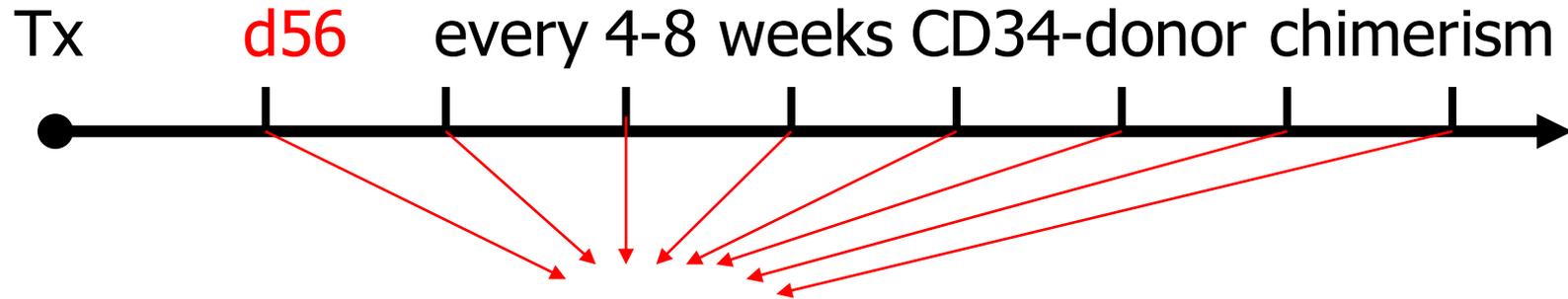
**Technical University of Dresden, Dresden, Germany**



**TECHNISCHE  
UNIVERSITÄT  
DRESDEN**



# Study design - Therapy



- Pt eligibility criteria for treatment:
  - CD34<sup>+</sup>-DC dropped < 80%
  - Still in CR
- **AZA 75 mg/m<sup>2</sup>/day s.c. days 1–7, q28d x 4**

# **(Some) Strategies for Preventing Relapse**

- 1- Pre-emptive treatment of relapse
  - minimal residual disease-based (PCR, flowcytometry etc)
  - based on pre-transplant high-risk parameters or on dynamic approaches, such as presence of MRD after HSCT etc
  
- 2- Maintenance of remission
  
- 3- Improve conditioning regimen
  
- 4- Improve the graft
  
- 5- Select patients.

# Candidate agents

- FLT3 inhibitors
- Histone deacetylase inhibitors
- Hypomethylating agents
- Lenalidomide and others in class

# Candidate agents

- Monoclonal antibodies (lymphoid > myeloid)
- Moxetumomab pasudotox (anti CD22)
- Immunostimulatory mAb:
  - *anti-CTLA-4, anti-PD1, anti-PDL1 (antagonistic)*
  - *anti-4-1BB, anti-OX40 (agonistic)*
- Cells – educated or not
- Tumor vaccines
- Etc

# The maintenance agent

1- Active against the disease.

2- Not too toxic.

3- Not myelotoxic (or with tolerable myelotoxicity).

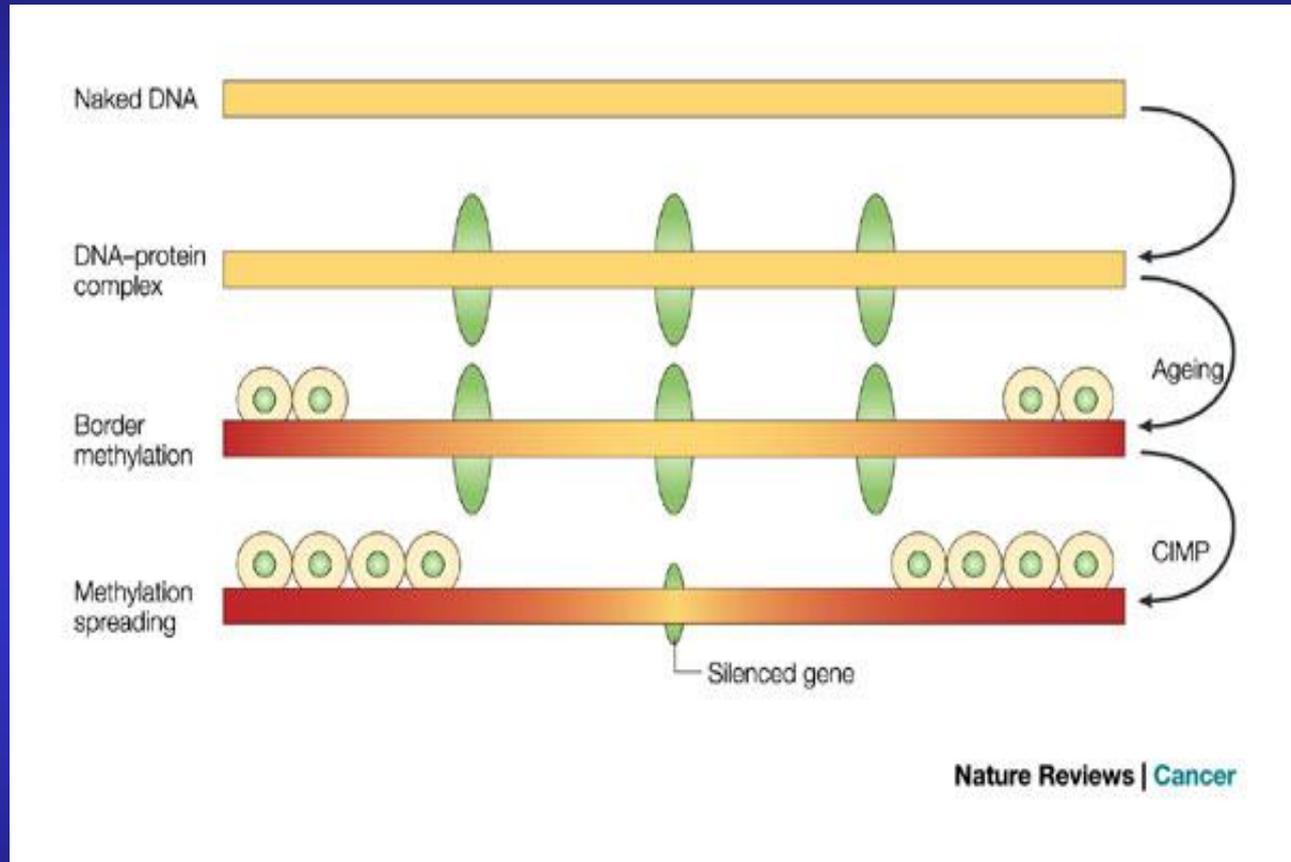
4- Can be given early after transplant.

5- Influence donor cells favorably.

6- Increase immunogenicity of malignant cells.

# CpG Island Methylator Phenotype in Cancer

DNA  
methyltransferase  
[DNMT]



Nature Reviews Cancer 4, 988-993 (2004)

Aberrant methylation of promoter-associated CpG islands is an epigenetic oncogenic mechanism.

# Low dose azacitidine to treat relapsed AML / MDS after allogeneic transplant

-Relapsed AML / MDS after allogeneic HSCT:

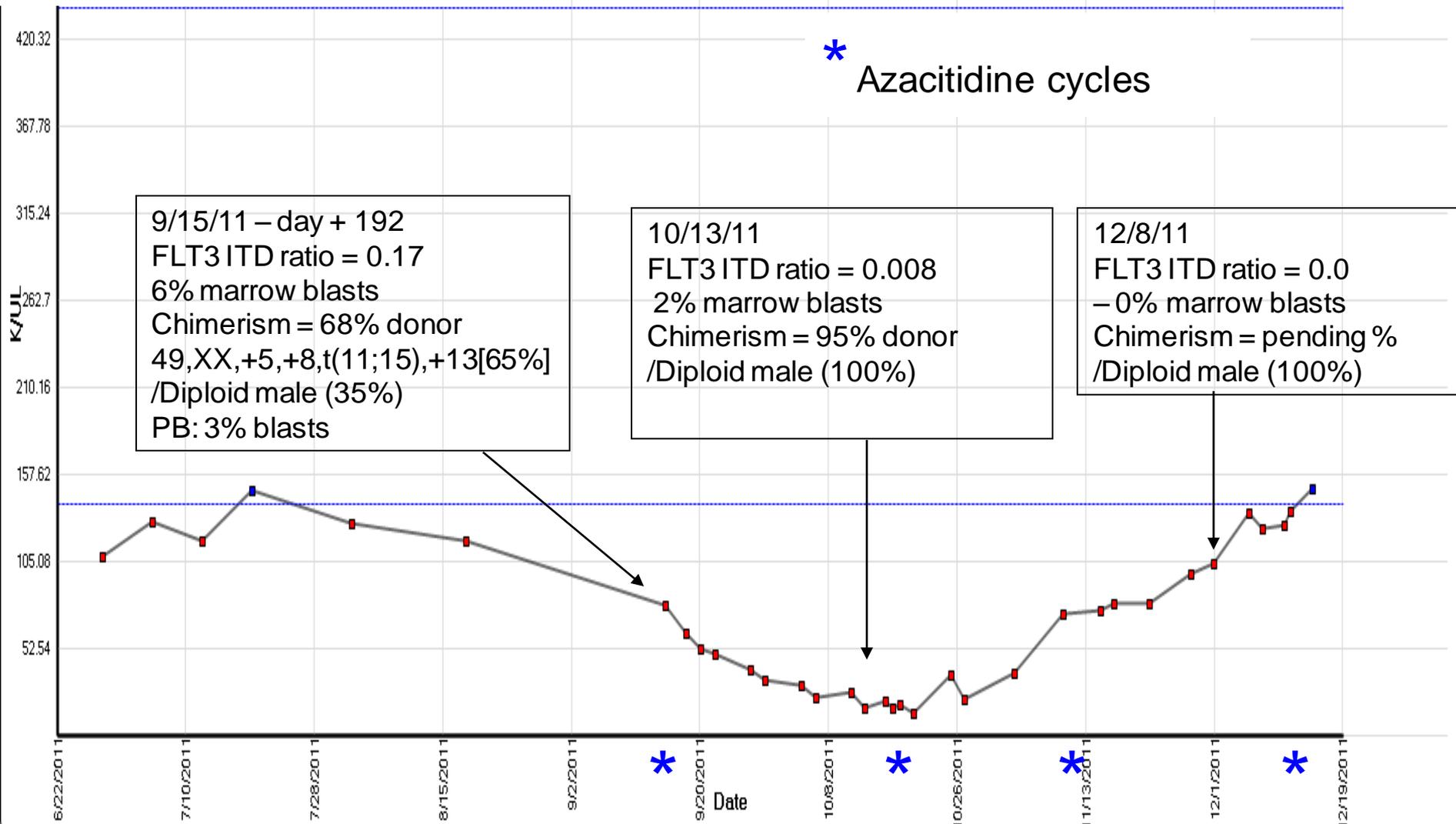
- doses of 16 – 40 mg/m<sup>2</sup> for 5 days in 28-30-day cycles induced complete remission and reversion to full donor chimerism in 20-25% of patients treated (n=19)

# Platelet count

24 year old

HLA identical sib transplant in CR1

Bu Flu conditioning



\* Azacitidine cycles

9/15/11 – day + 192  
FLT3 ITD ratio = 0.17  
6% marrow blasts  
Chimerism = 68% donor  
49,XX,+5,+8,t(11;15),+13[65%]  
/Diploid male (35%)  
PB: 3% blasts

10/13/11  
FLT3 ITD ratio = 0.008  
2% marrow blasts  
Chimerism = 95% donor  
/Diploid male (100%)

12/8/11  
FLT3 ITD ratio = 0.0  
– 0% marrow blasts  
Chimerism = pending %  
/Diploid male (100%)

Immunosuppression withdrawal ←

# Ideal maintenance agent

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## Trial design

- 1- Dose may not be the same as in other scenarios!
- 2- Phase III trial mandatory given multiples biases, confounding variables etc

# Hypomethylating Agents

- induce phenotypic modification of leukemic cells, including reduction of CD13 and CD33 expression,
- increase antigenic density of surface determinants of mature myeloid cells such as CD16 and CD11c,
- increase expression of MHC-class I molecules, HLA-DR and beta-2-microglobulin.

**Pinto A. Blood 1984; 64: 922-929.**

**Pinto A. Lancet 1984; 2: 867-868.**

**Coral S. J Immunother. 1999; 22:16-24**

# Hypomethylating Agent dose

Classic idea : Allogeneic stem cell transplant context  
(with BuCy): - decitabine 400 mg / m<sup>2</sup>, 600 mg / m<sup>2</sup> and  
800 mg / m<sup>2</sup>

**de Lima. Cancer. 2003 Mar 1;97(5):1242-7.**

Phase 1 study of low-dose prolonged exposure schedules of  
decitabine in hematopoietic malignancies.

5-20 mg/m<sup>2</sup> 5 days/week x 2 weeks  
15 mg/m<sup>2</sup> best - 30 times < MTD

**Issa JP et al. Blood. 2004 Mar 1;103(5):1635-40.**

Duration of exposure - longer may be better.

Dose – is low better, same or worse ??

## **Hypothesis**

Low dose 5-Azacitidine will decrease the relapse rate after allogeneic transplantation.

## **Study Aim**

To determine the safest dose and schedule combination of azacitidine given after allogeneic transplant.

# Patients in complete remission after HSCT

Serum creatinine <1.6 mg/dL and  
Serum bilirubin <1.6 mg/dL and  
SGPT < 3 X upper limit of normal and  
Platelet count greater than  
15,000/mm<sup>3</sup>  
ANC > 1,000/mm<sup>3</sup>  
No active bleeding  
No uncontrolled acute GVHD  
No acute GVHD grade III or IV  
No life-threatening infection.



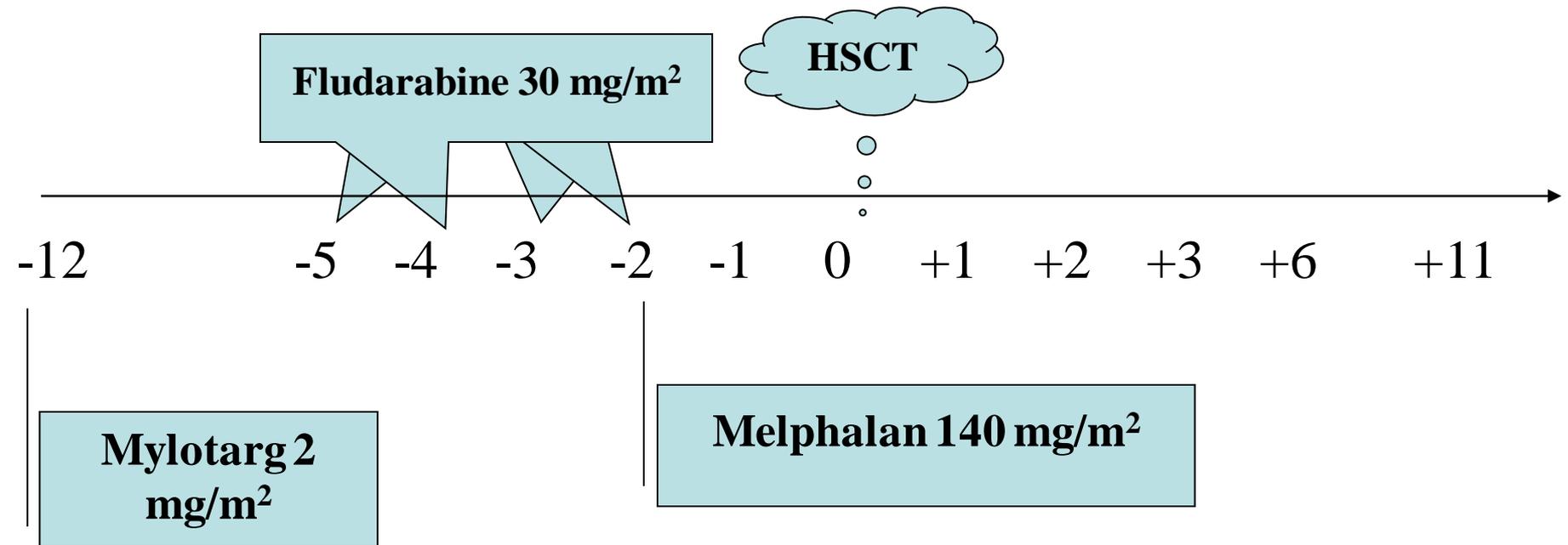
Assigned to  
5-azacitidine

Dose

AND

Schedule

**Treatment Plan** : 5-azacitidine was given in up to four monthly cycles; each patient is assigned to a dose and a schedule (for example, study started with 8 mg/m<sup>2</sup> x 1 cycle)



CD33 positivity by flow cytometry in > 20% of leukemia cells

**GVHD prophylaxis: tacrolimus from day -2 (levels at 5-15 ng/mL) and mini-methotrexate 5 mg/m<sup>2</sup> on days +1, +3, +6 and +11**

**MUD: Rabbit ATG 0.5 mg/Kg day -3, 1.25 mg/Kg days -2 and -1**

# Protocol 2005-0417

## Patient characteristics

Median age = 60 ( range, 24 – 67 )

Median comorbidity score : 3 (range, 0-8)

Chemotherapy regimens prior to HSCT (median = 2)

AML from MDS : 73%      MDS : 5%      AML : 22%

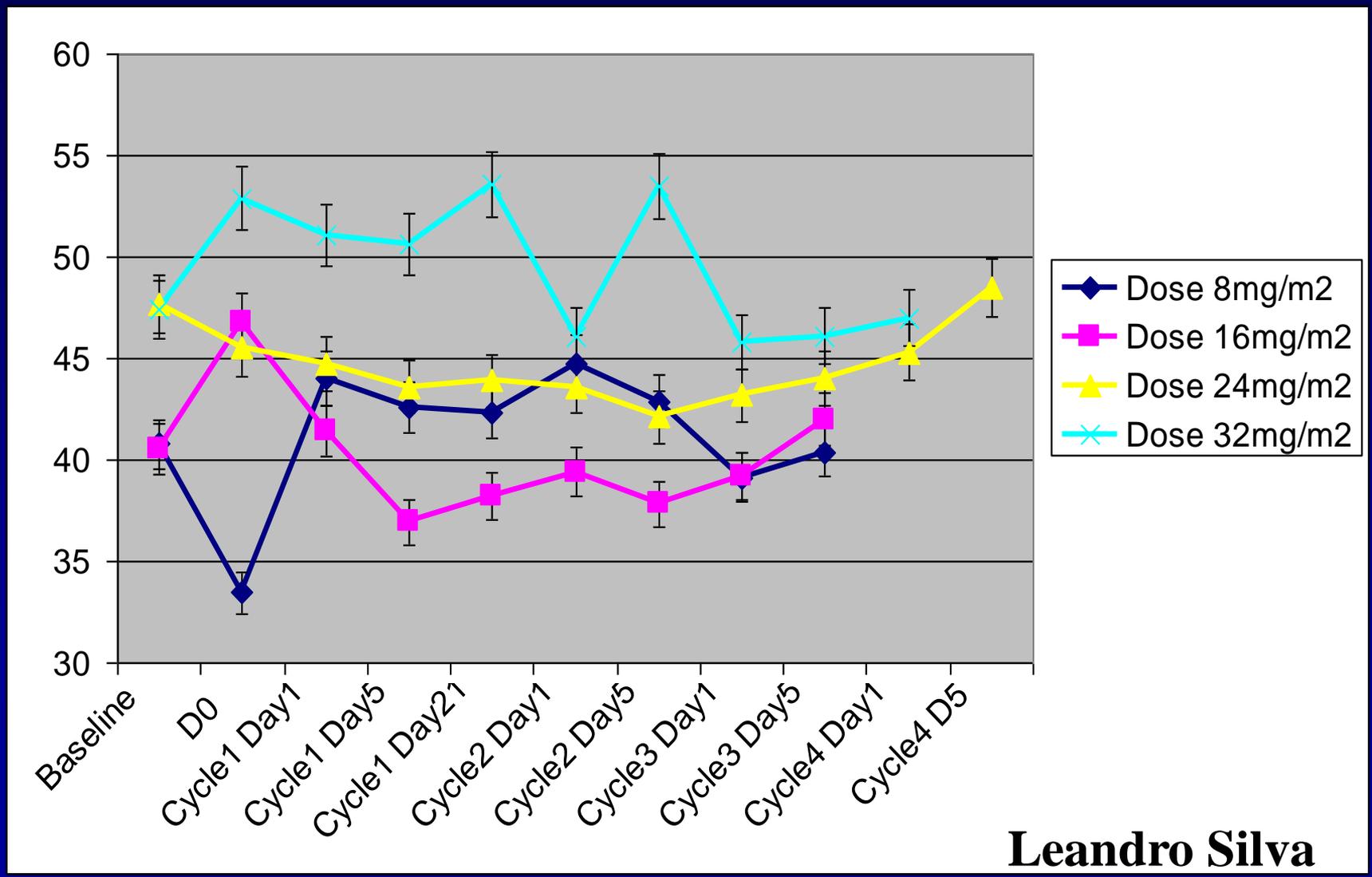
Median bone marrow blasts at transplant : 10% (0-86%)

CR at HSCT : 20%

# Global DNA methylation (LINE assay (bisulfite pyrosequencing)) :

No dose was found to significantly affect global methylation

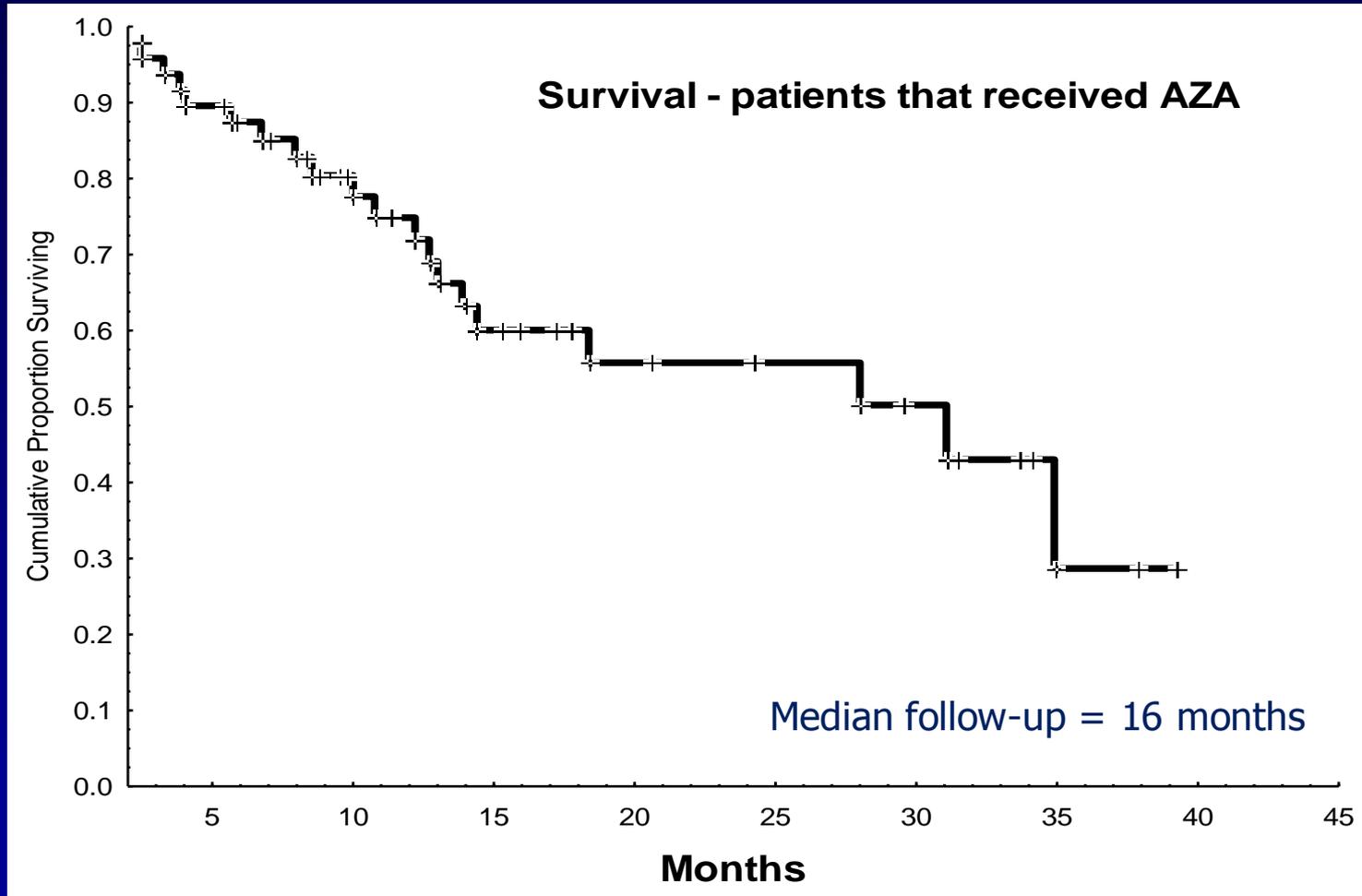
**Guillermo Garcia-Manero's laboratory**



**Leandro Silva**

# Azacitidine maintenance

– MTD : 32 mg/m<sup>2</sup>



50% unrelated donor HSCT -  
96 cycles delivered - safe.

# Fitted Bayesian logistic regression model :

less cGVHD with longer exposure, independent of the dose

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| Variable         | Mean            | SD    | Posterior 95% Credible Interval |        | Probability of a Beneficial Effect |
|------------------|-----------------|-------|---------------------------------|--------|------------------------------------|
|                  |                 |       | 2.50%                           | 97.50% |                                    |
| Intercept        | 0.582           | 0.779 | -0.887                          | 2.111  | -                                  |
| Azacitidine dose | -<br>0.014<br>5 | 0.036 | -0.083                          | 0.057  | 0.658                              |
| Number of cycles | -<br>0.439      | 0.311 | -1.073                          | 0.159  | 0.928                              |

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# Protocol 2005-0417

- Azacitidine was well tolerated
- Approximately 60% of the patients (heavily pre-treated, refractory etc) were able to receive at least one cycle
- At least 4 cycles at 32 mg/m<sup>2</sup> could be delivered.
- Randomized protocol 2008-0503 is ongoing :  
32 mg/m<sup>2</sup> daily X 5 days, every 30 days, for 1 year, versus no maintenance.

# The dose issue

Lenalidomide maintenance after nonmyeloablative allogeneic stem cell transplantation in multiple myeloma is not feasible: results of the HOVON 76 Trial. Kneppers E, et al. Blood. 2011

nonmyeloablative allo-SCT+ Lenalidomide (10 mg 21/28 days)

N=35 - 30 started with lenalidomide

N=14 (47%) maintenance stopped due to aGVHD

N=13 (43%) stopped treatment because of GVHD

N=5 (17%) due to other adverse events

N=5 (17%) because of progression.

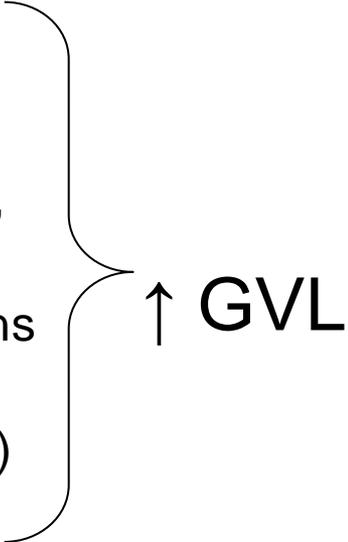
- increased the frequency of HLA-DR(+) T cells and regulatory T cells

# Ideal maintenance agent

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- 2- Not too toxic.
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# Hypomethylating Agents – Potential Effects

- Increased expression of tumor-associated antigens ie CTA (Roman-Gomez, 2007) Tatjana Stankovic et al. Goodyear et al.
  - Increased expression of KIR ligands on hematopoietic cells (Liu, 2009)
  - Recovery of reduced expression of HLA class I, II and III antigens on tumor cells (Campoli & Ferrone, 2008) (Pinto et al – 1984)
  - Increased expression of known Minor antigens (Hambach, 2009)
  - Affect microRNA function - inhibition of oncogenes
- 
- Increased FoxP3 expression and T<sub>reg</sub> generation (Polansky, 2008) (Choi et al. 2010) (Sanchez-Abarca et al. 2010)  
John DiPersio et al.



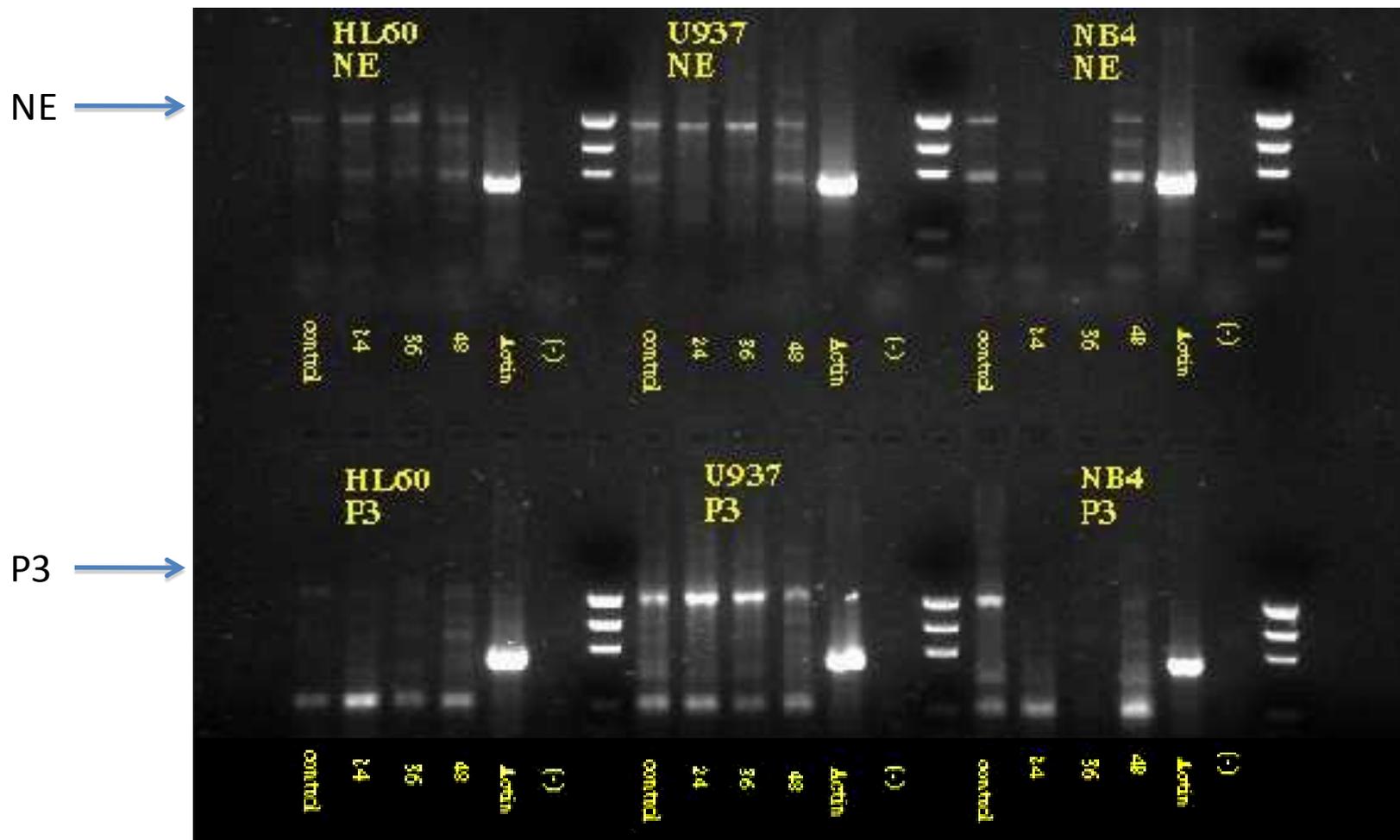
↑ GVL

? GVL

Tolerance

Without affecting relapse ?

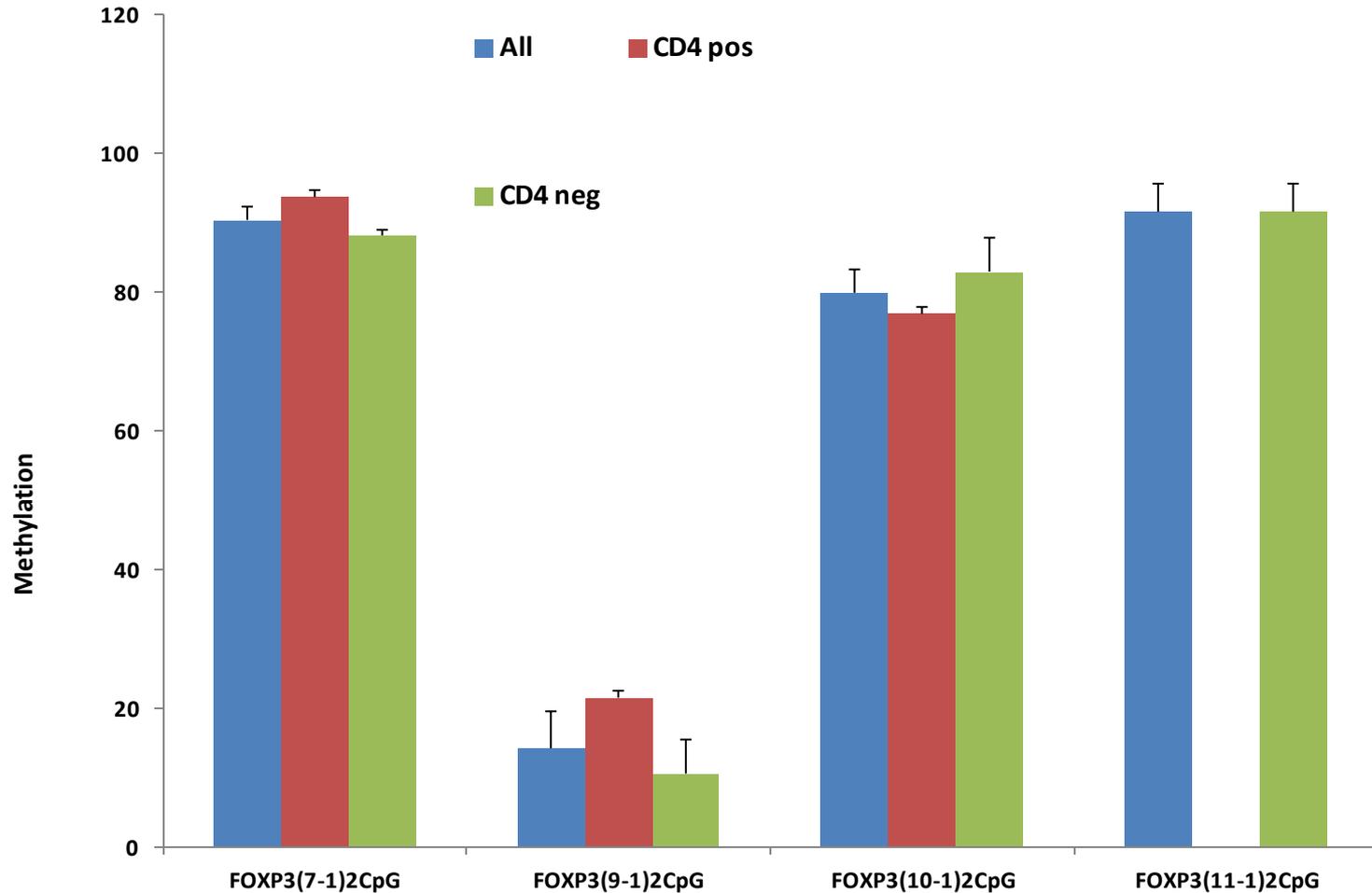
# Azacitidine affects expression of the LAAs NE and P3 (PR1 precursors) in leukemia cell lines



Methylation levels at the FOXP3 gene are decreased during treatment with low dose AZA

**Simrit Parmar - MDACC**

# FOXP3 Demethylation in amplicon 9 in CD4 + and negative cells (2 subjects)



7-color flow for: CD4, CD8, CD14, CD16, CD19, CD25, CD127

# Goodyear et al Blood 2012

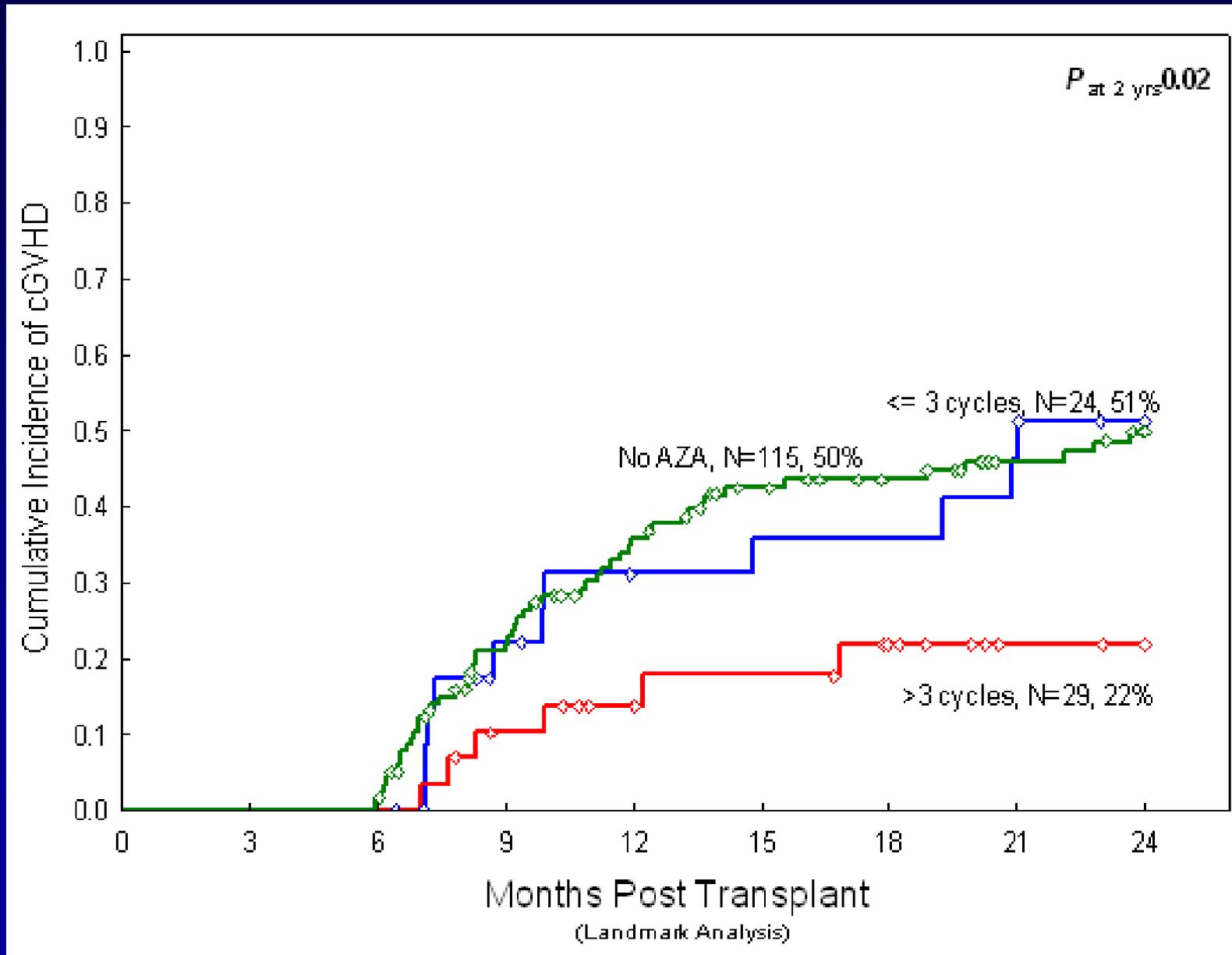
- 27 patients (median age 59)
- Median follow up 7 months (3-21)
- 11 sib, 16 VUD
- CR1=18, CR2=7, relapse 2
  - Control cohort: FMC 50 no AZA
- Induction of an increase in circulating T-regs in the early post transplant period
- Induction of a memory T cell-mediated response in the bone marrow to putative tumour antigens
- 15/18 pts (80%) who had 6 cycles of treatment had detectable CD8+ T cell responses to tumour specific peptides

# **Low dose AZA and cGVHD**

# Patient and disease characteristics

|  | Controls (n=230) | AZA≤3 cycles(n=48) | AZA>3 cycles (n=37) | P(controls X AZA>3 cycles) |
|--|------------------|--------------------|---------------------|----------------------------|
| Median age   | 52               | 60                 | 52                  | 0.9                        |
| Ablative preparative regimen                         | 63%              | 25%                | 30%                 | <0.001                     |
| Disease status (remission (CR1/CR2)/ active disease) | 64%/36%          | 31%/69%            | 41%/59%             | 0.01                       |
| AML/MDS  | 95%              | 96%                | 86%                 | 0.07                       |
| Second allo HSCT                                     | 7%               | 15%                | 22%                 | 0.01                       |
| Peripheral blood graft                               | 86%              | 75%                | 70%                 | 0.02                       |
| <10/10 HLA match                                     | 4%               | 11%                | 13%                 | P=NS                       |
| Tacrolimus-based GVHD prophylaxis                    | 98%              | 92%                | 89%                 | 0.01                       |
| aGVHD incidence (grade II-IV/III-IV)                 | 10%/2%           | 17%/11%            | 25%/3%              | 0.01 (gd II-IV)            |

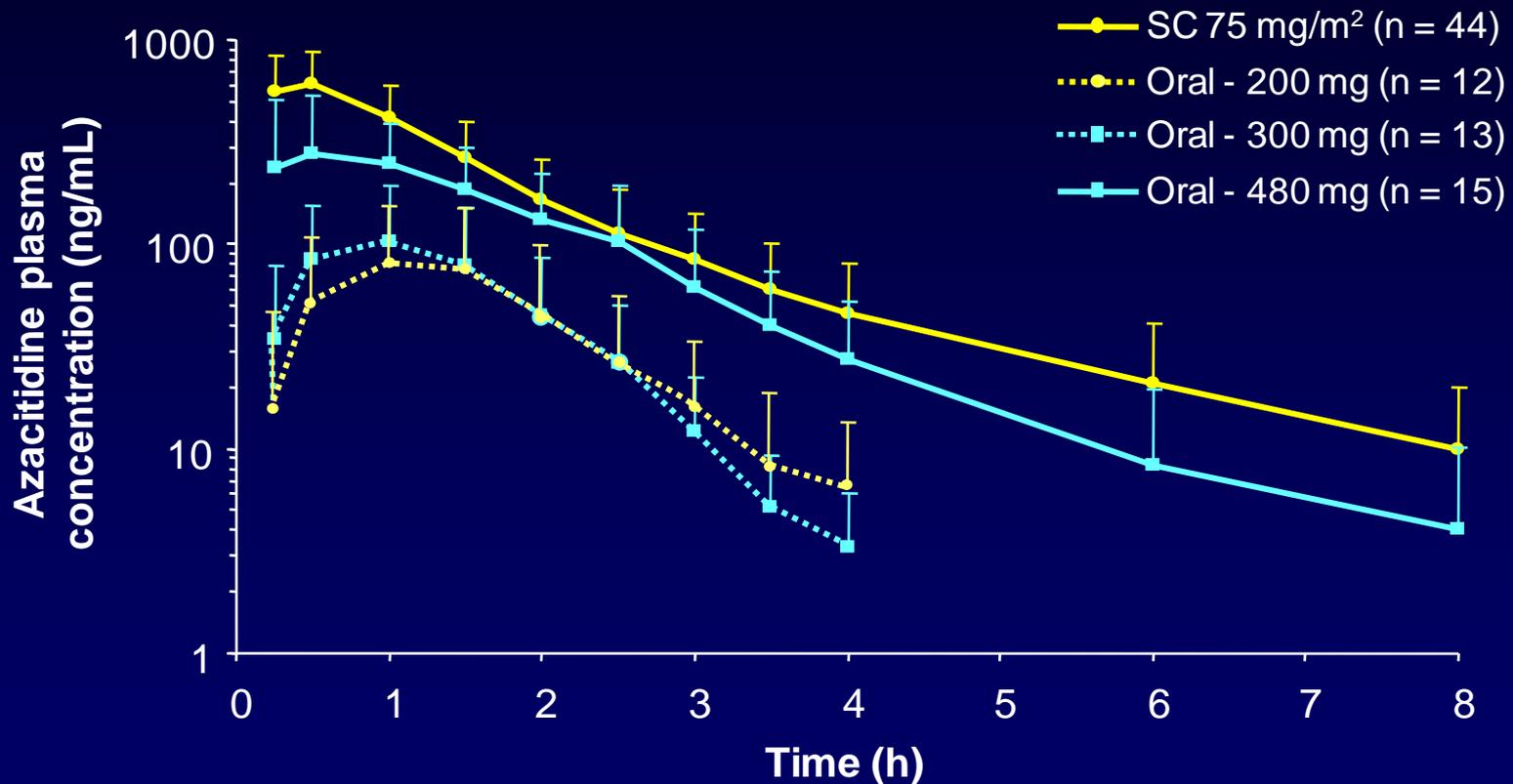
# Cumulative incidence of cGVHD. 6-month landmark analysis.



**Oral AZA**



# AZA Plasma Concentration Profiles Following SC or Oral Administration\*



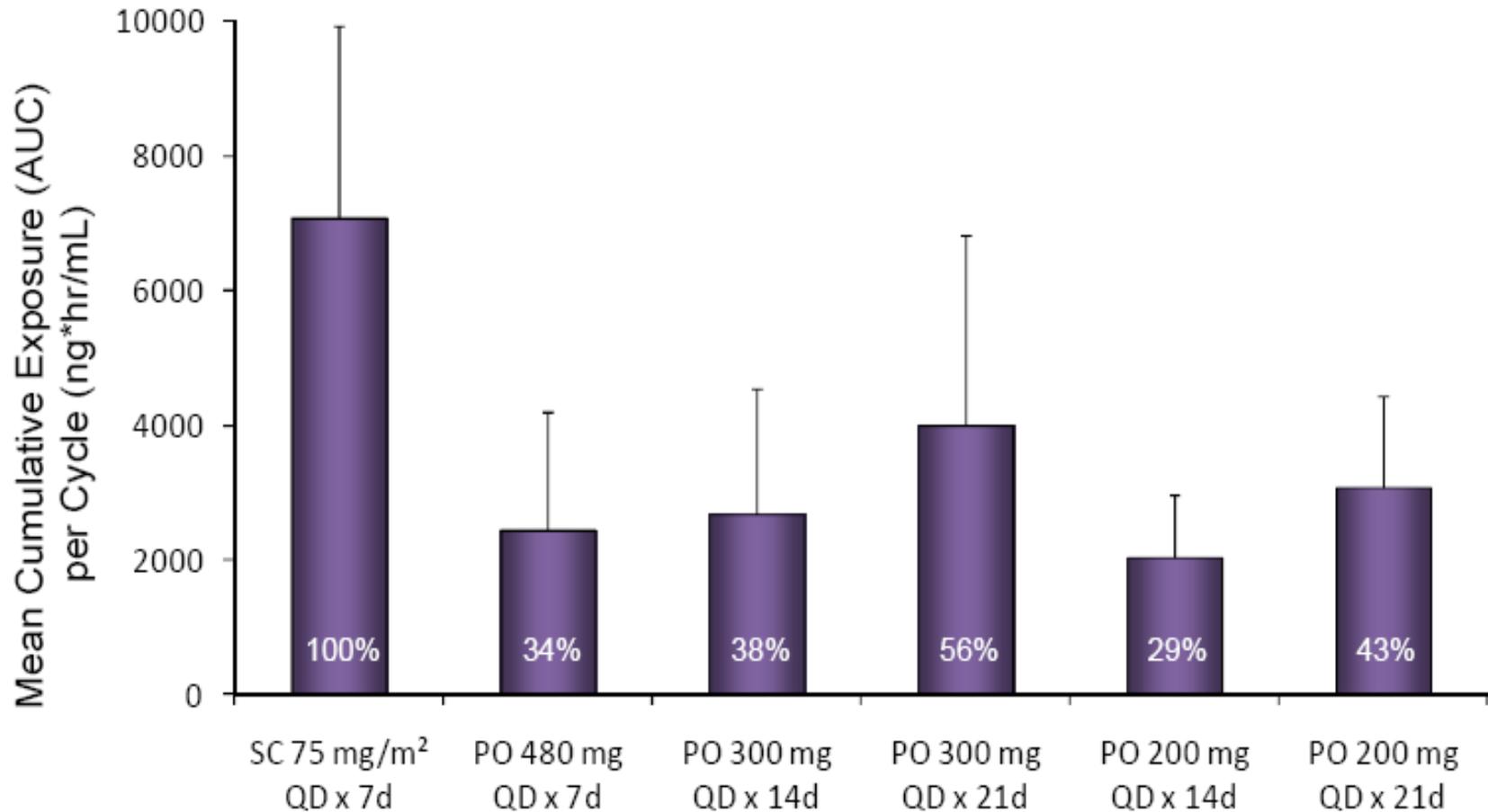
# Oral Azacitidine Absorption

Absorb approximately 11% of given oral dose

Cleared from plasma by 8 hrs (parent drug)

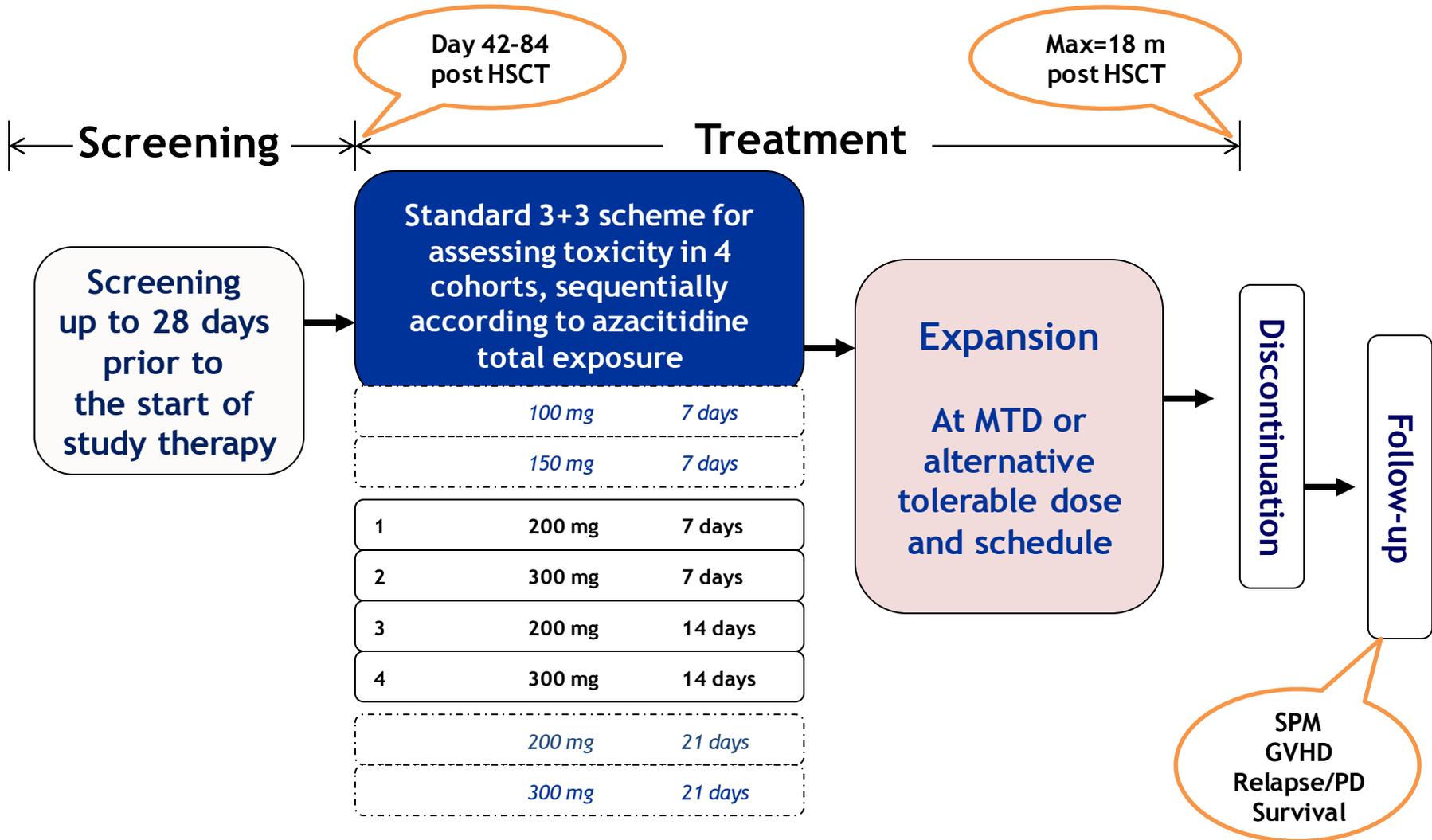
Approximately 2% of dose cleared through urinary route

# Extrapolated Cumulative Azacitidine Exposure per Cycle



AUC = area under the concentration-time curve; QD = once daily; SC = subcutaneous.  
Note: the percentage of exposure is compared to SC (shown in bars).

# Study Design Flow



# Conclusions

- Maintenance therapy may contribute to the treatment of patients with AML/MDS.
- Hypomethylating agents may modulate GVL and GVHD after allogeneic transplantation.
- The post transplant scenario, once the realm of GVHD trials, may provide an ideal arena to improve disease control now that new therapies (cellular and otherwise) are available.

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