Effective Therapeutic Targeting of Leukemia Initiating Cells in Chronic Myeloid Leukemia

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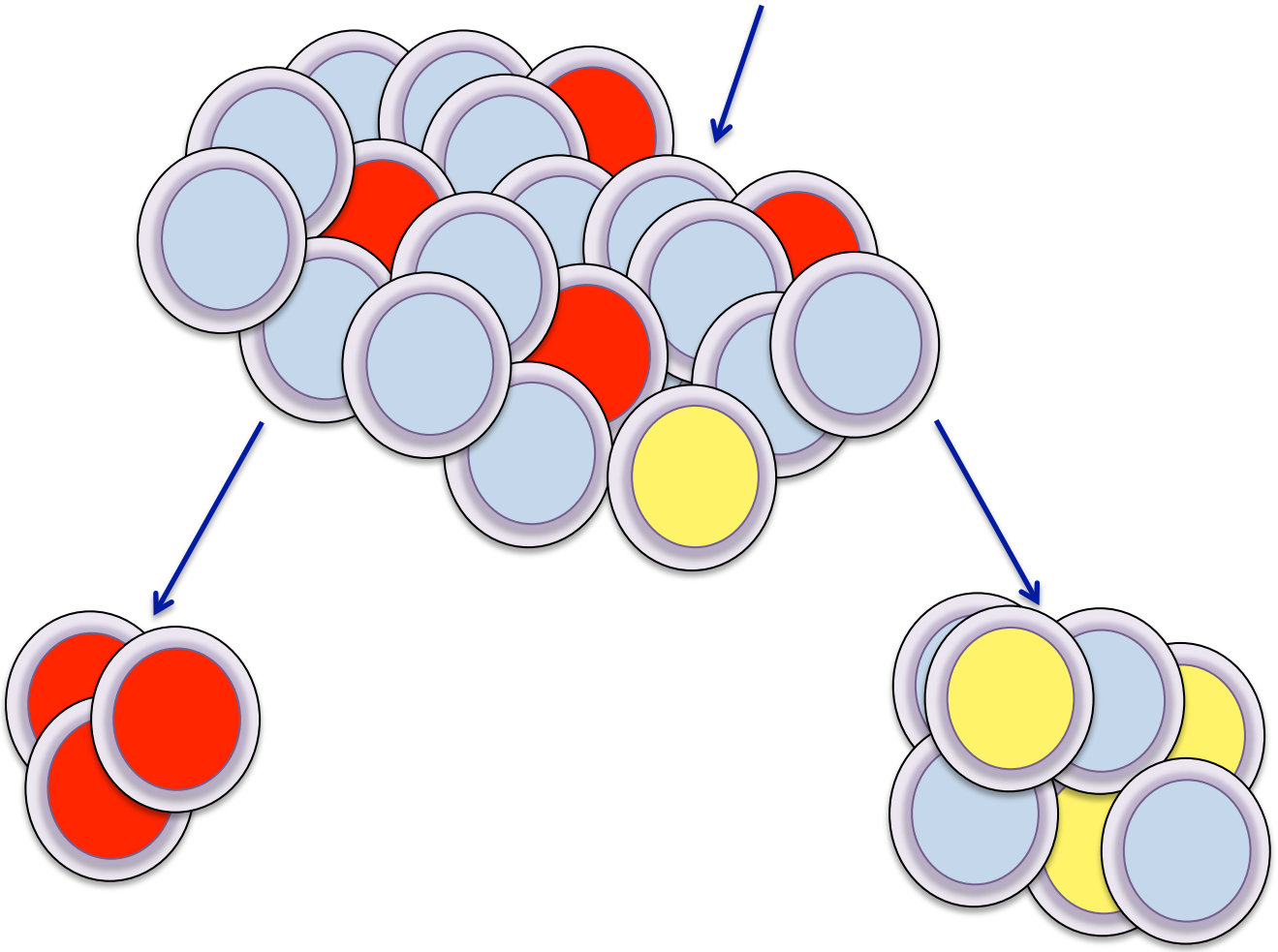
Cancer 1

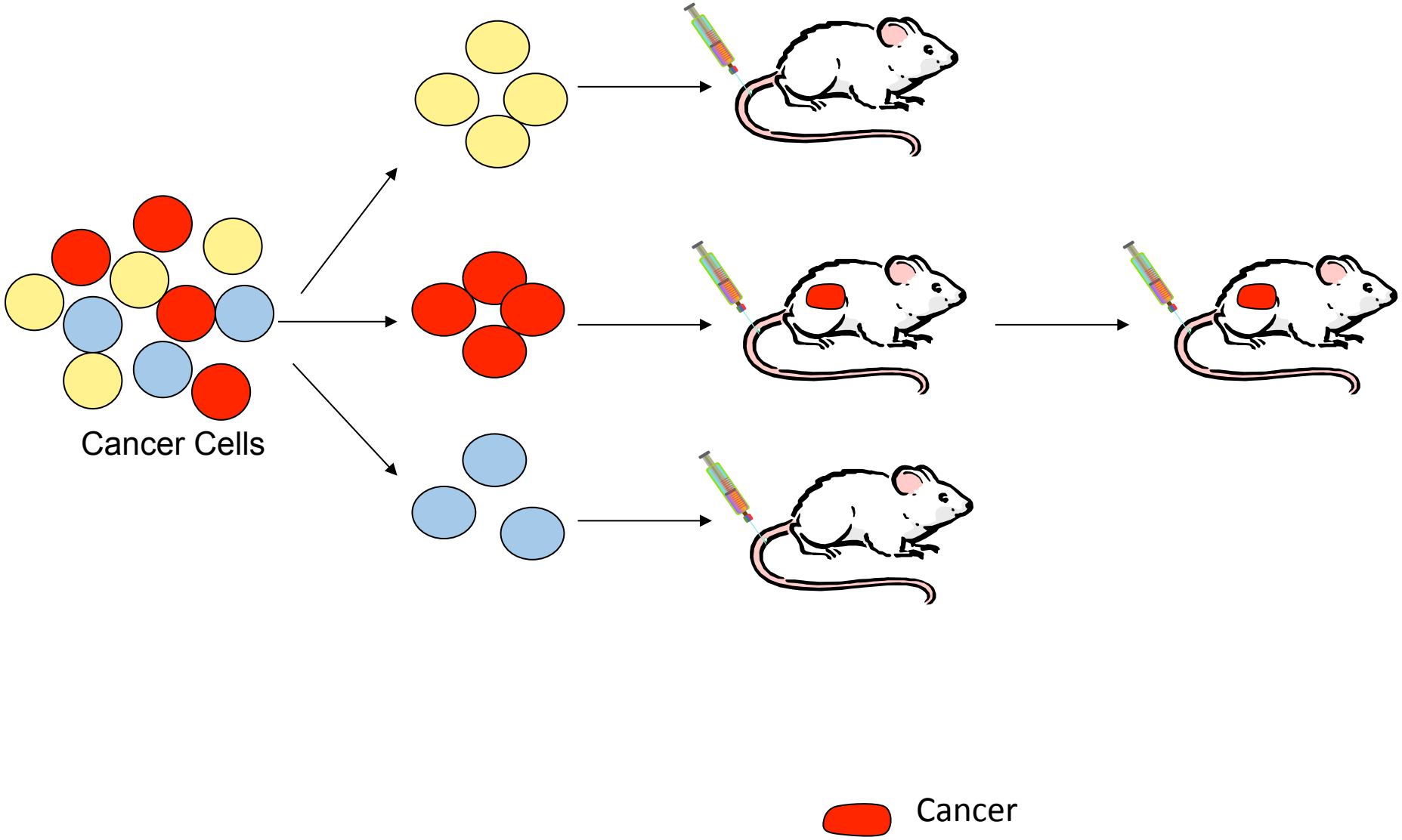
Cancer 2

Cancer 3

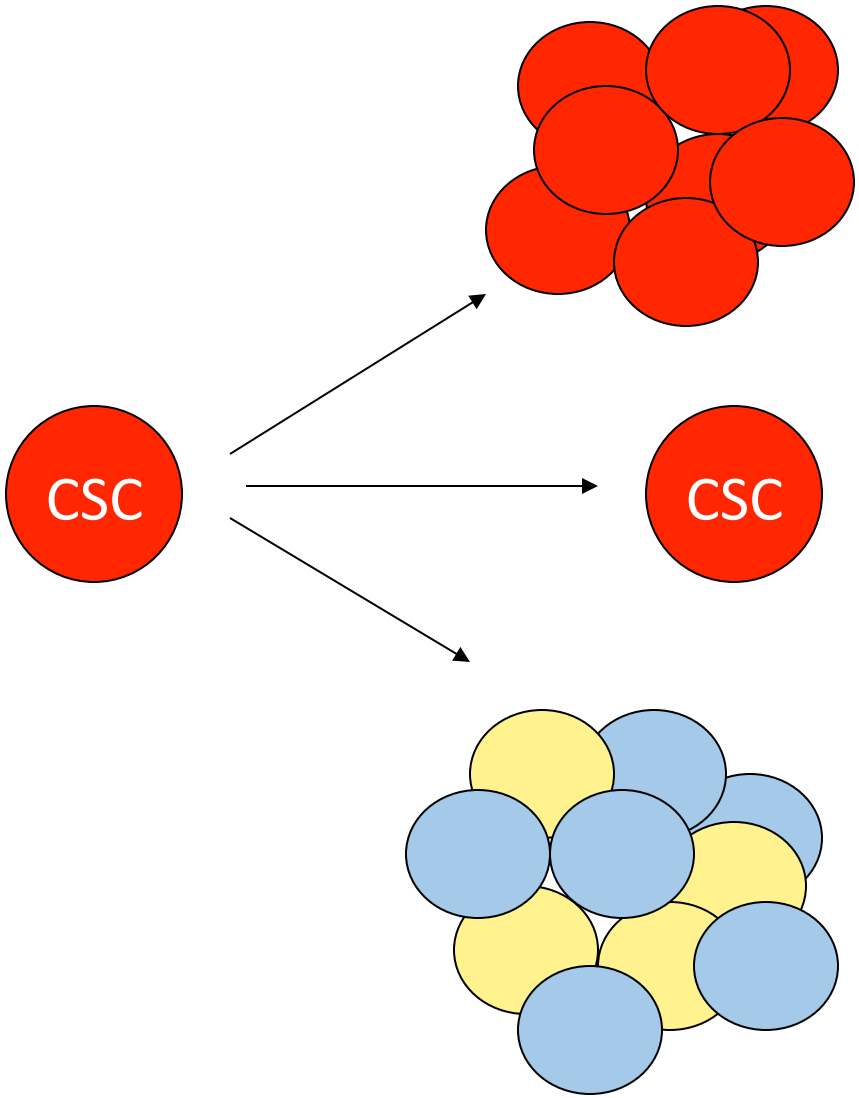
Cancer 4



Serial Transplantation Assay in mice



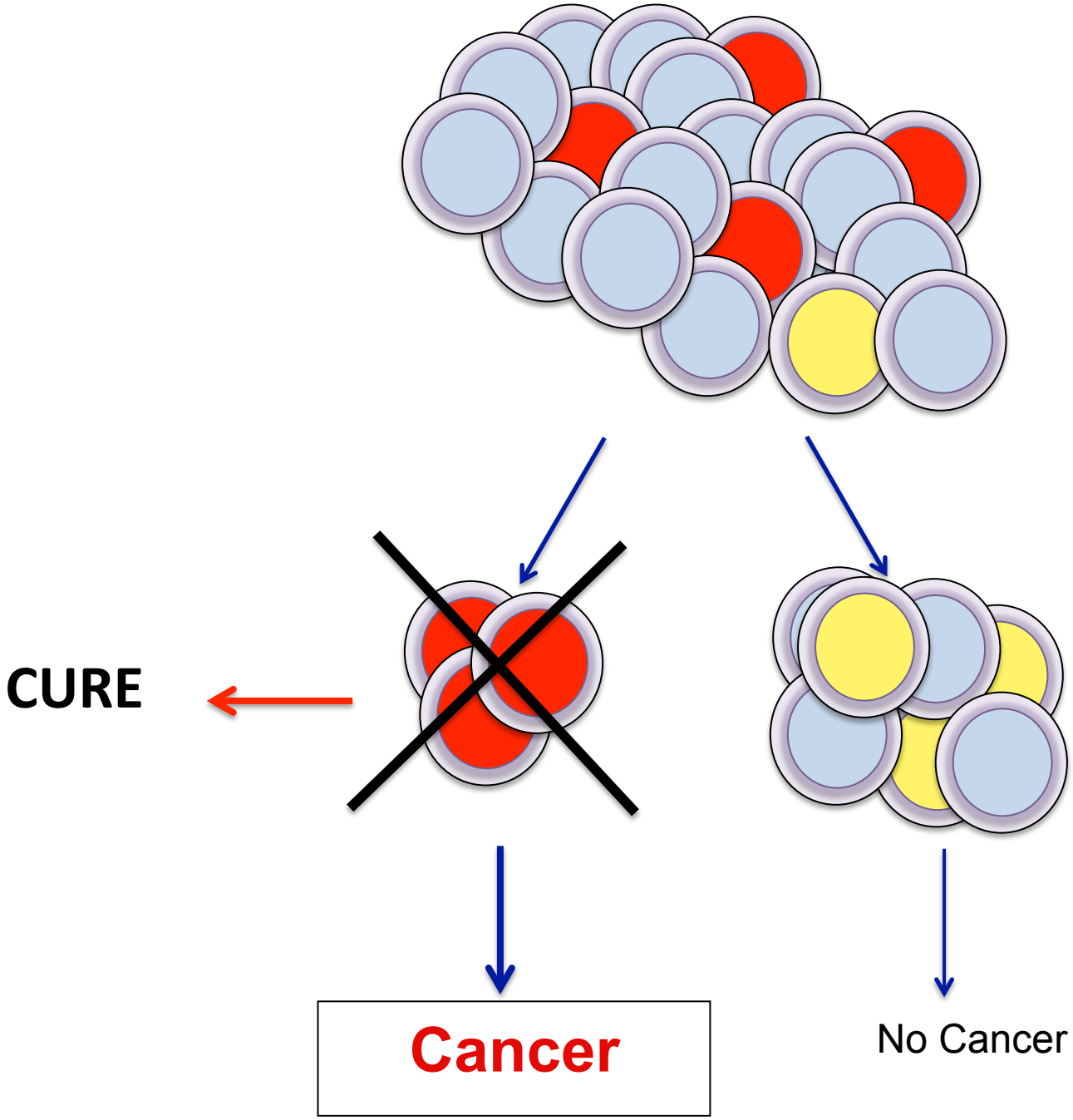
Cancer Stem Cells (CSC)

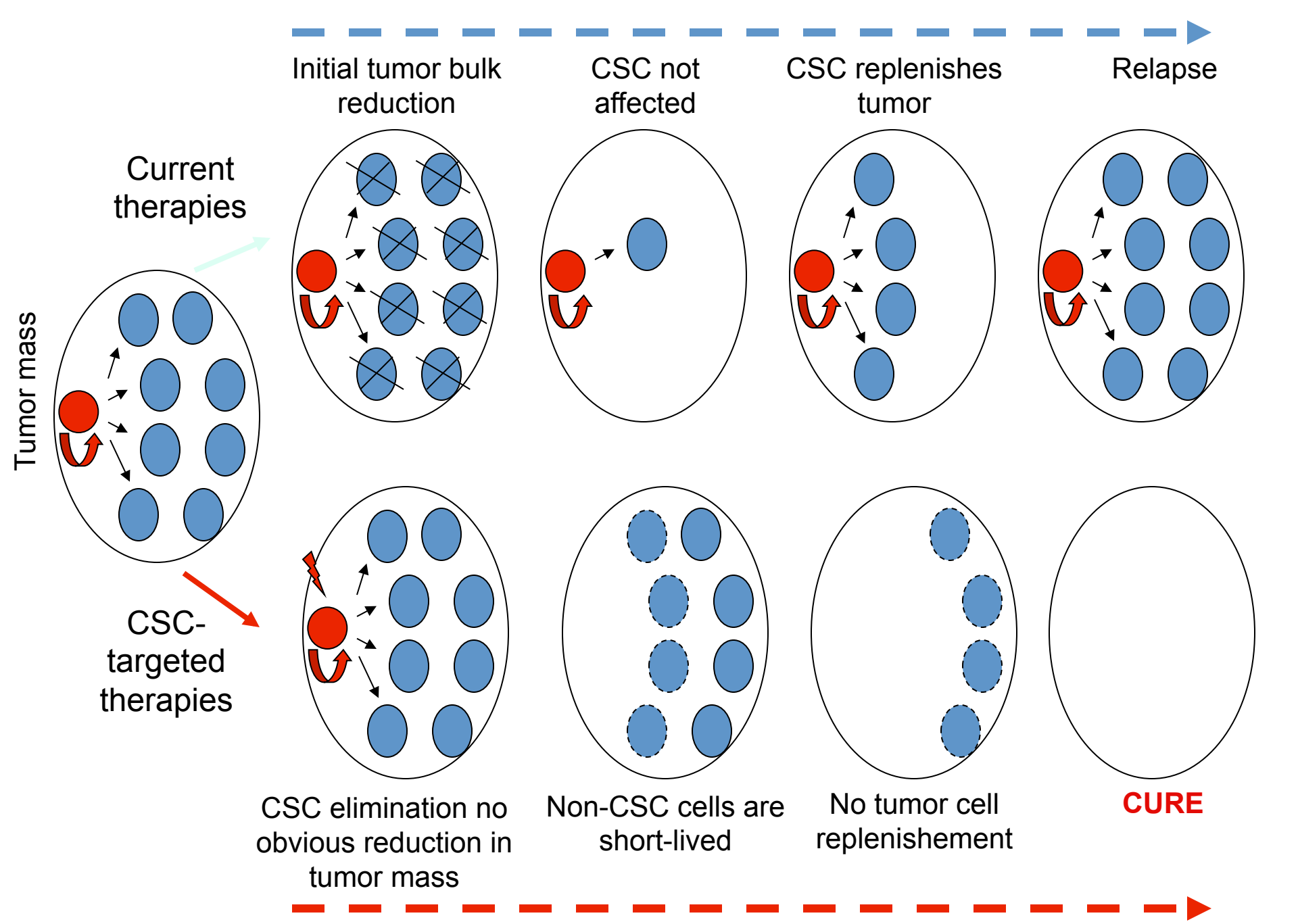


Enhanced capacity for **proliferation** and tumorigenic growth

Ability to create endless copies of CSC through **self-renewal**

Potential to give rise to more differentiated tumor bulk populations

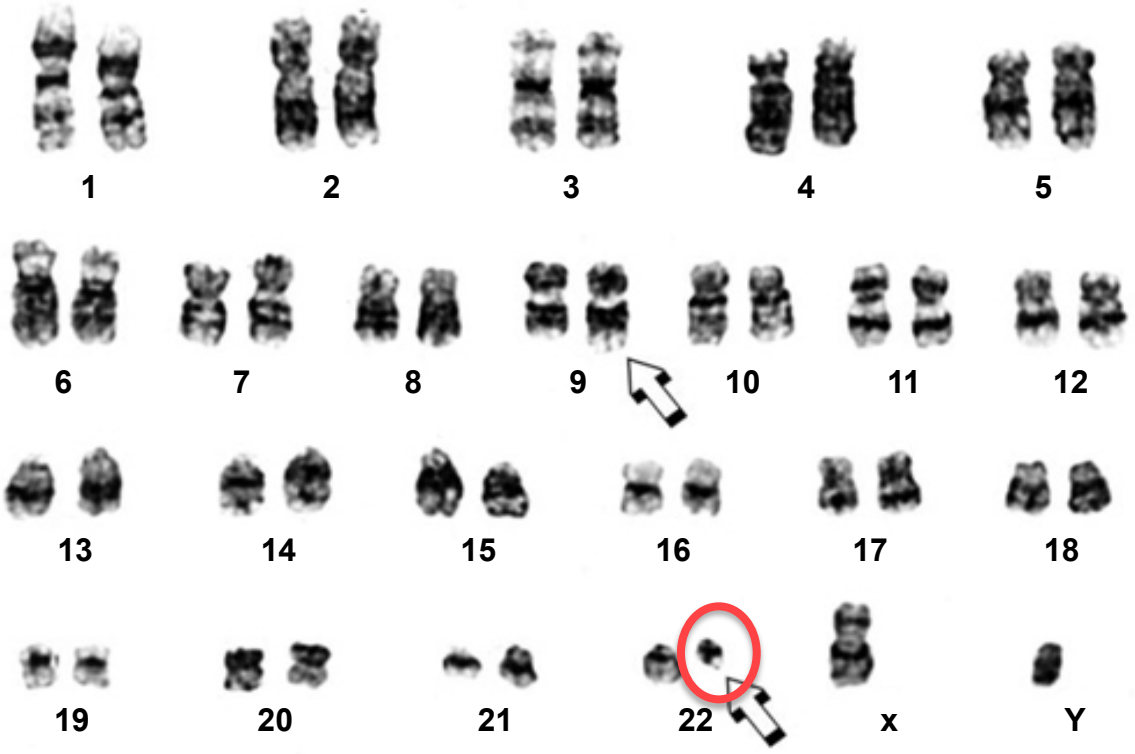

CURE
Cancer
No Cancer



Adapted from Frank et al., 2007

Chronic Myeloid Leukemia (CML)

In 1960, Nowell and Hungerford discovered an abnormal chromosome (Philadelphia chromosome)



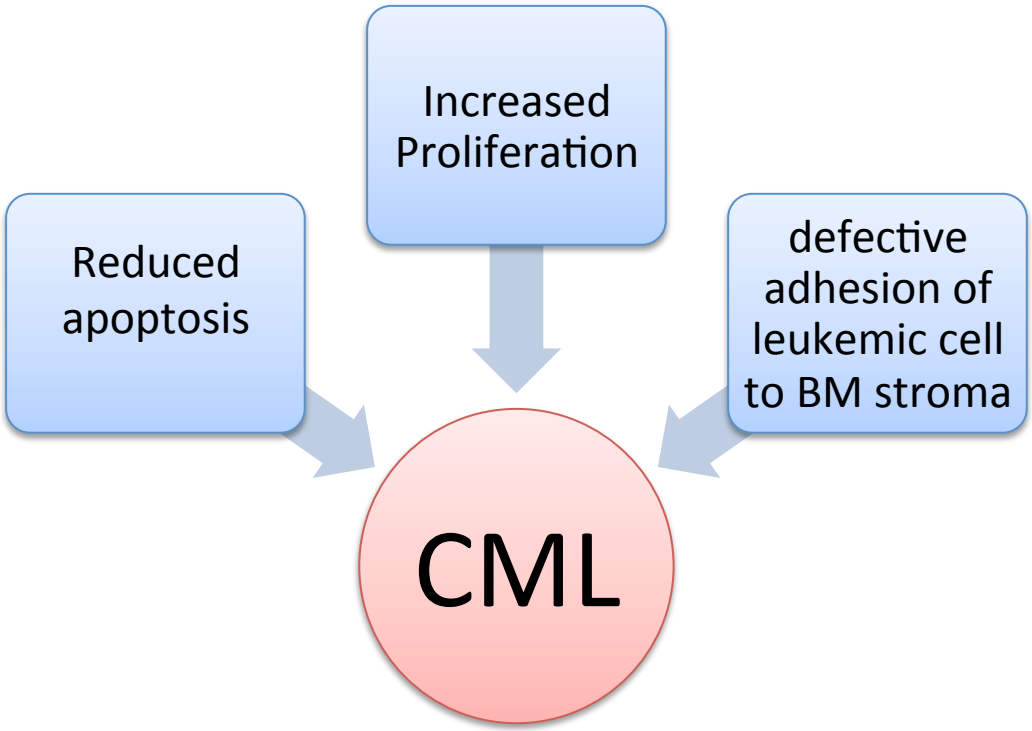
In 1973, Rowley suggested " there may be undetected translocation between the long arms of 22 and 9"

***bcr-abl* fusion gene**

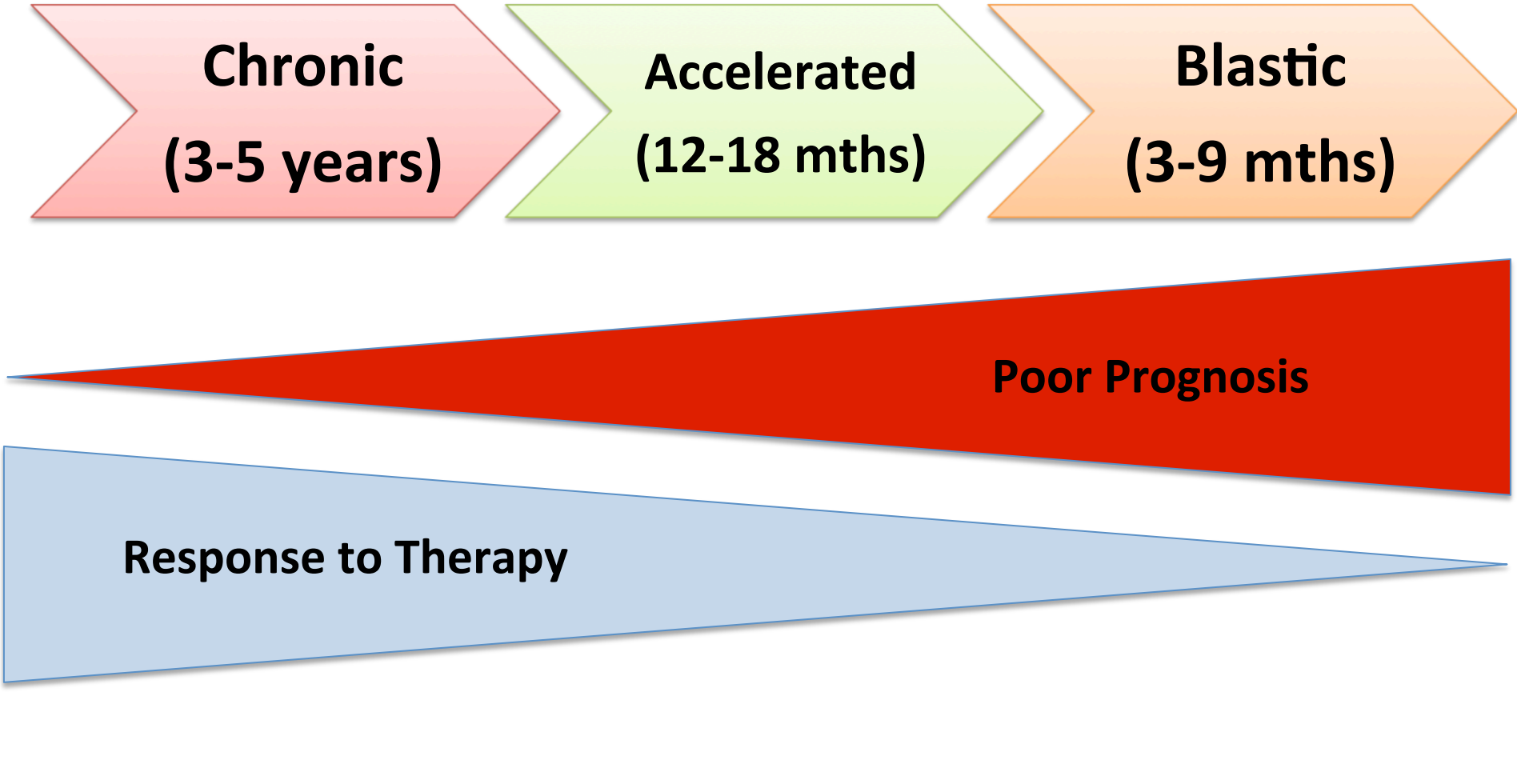
↓

BCR-ABL fusion protein
(constitutively active tyrosine kinase)

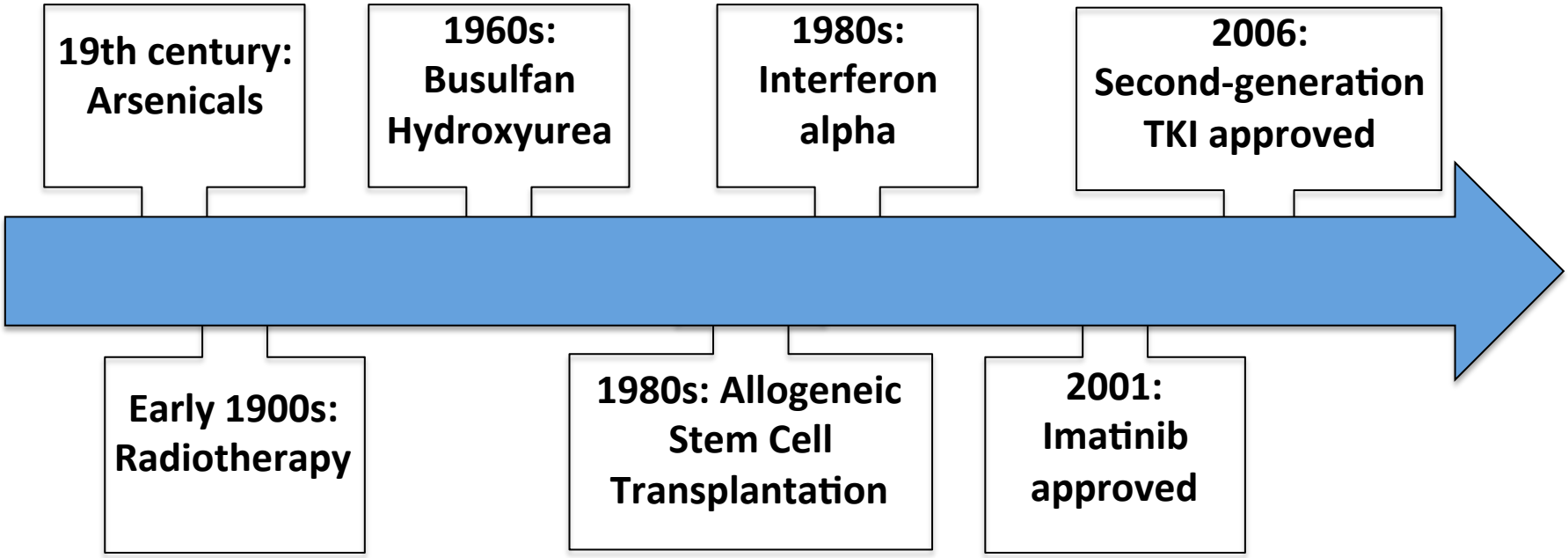
↓



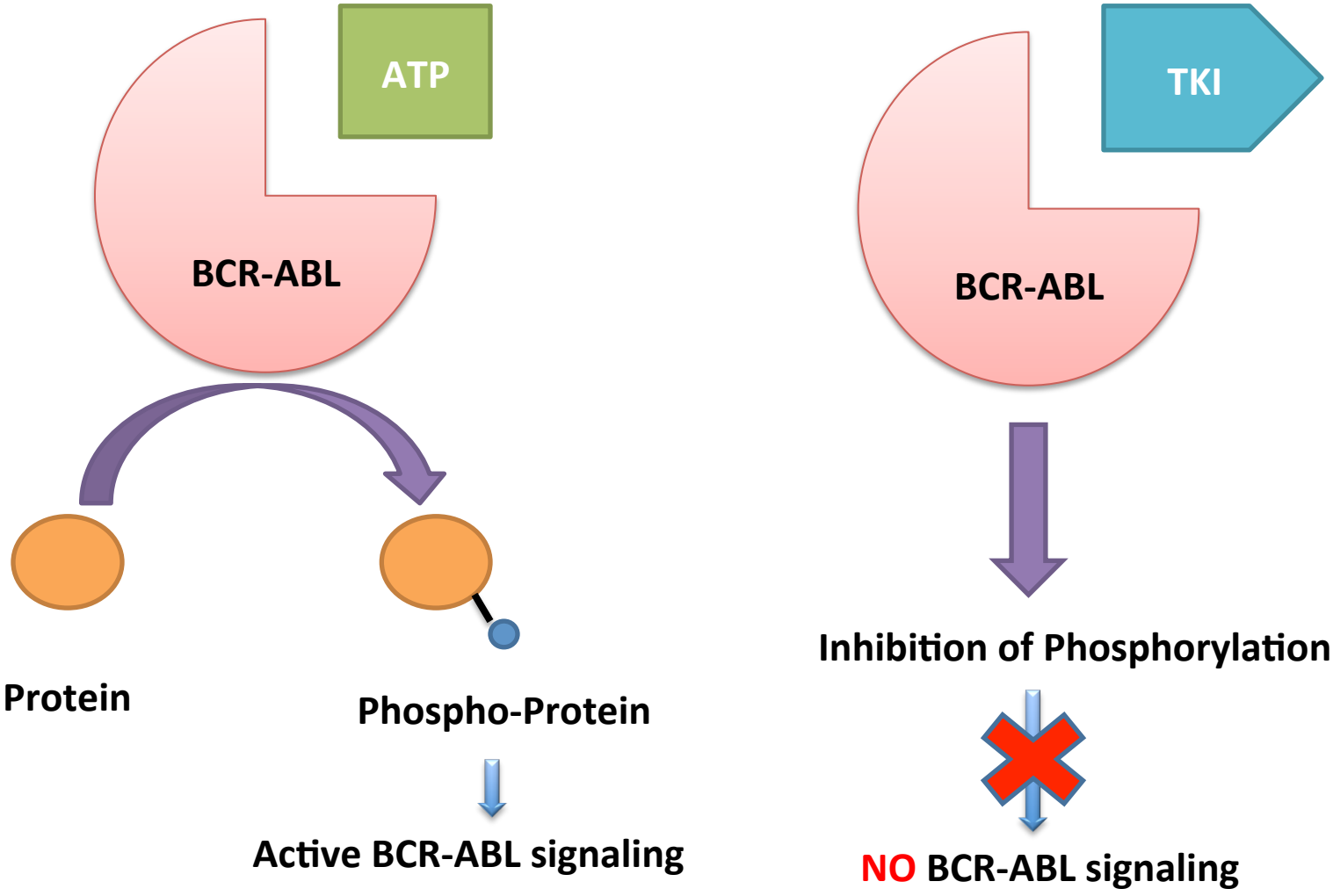
CML Phases


Chronic
(3-5 years)
Accelerated
(12-18 mths)
Blastic
(3-9 mths)
Poor Prognosis
Response to Therapy

Evolution of CML Treatment



Tyrosine Kinase Inhibitors (TKI): Standard of care in CML treatment



Can TKI cure CML?

- **Are patients alive and well?**

TKIs prolong survival of CML patients **BUT**

- Less effective in advanced stages
- Resistance
- Intolerance
- Economic burden

Can TKI cure CML?

- **Off all treatment?**

Patients relapse occurs shortly after therapy discontinuation

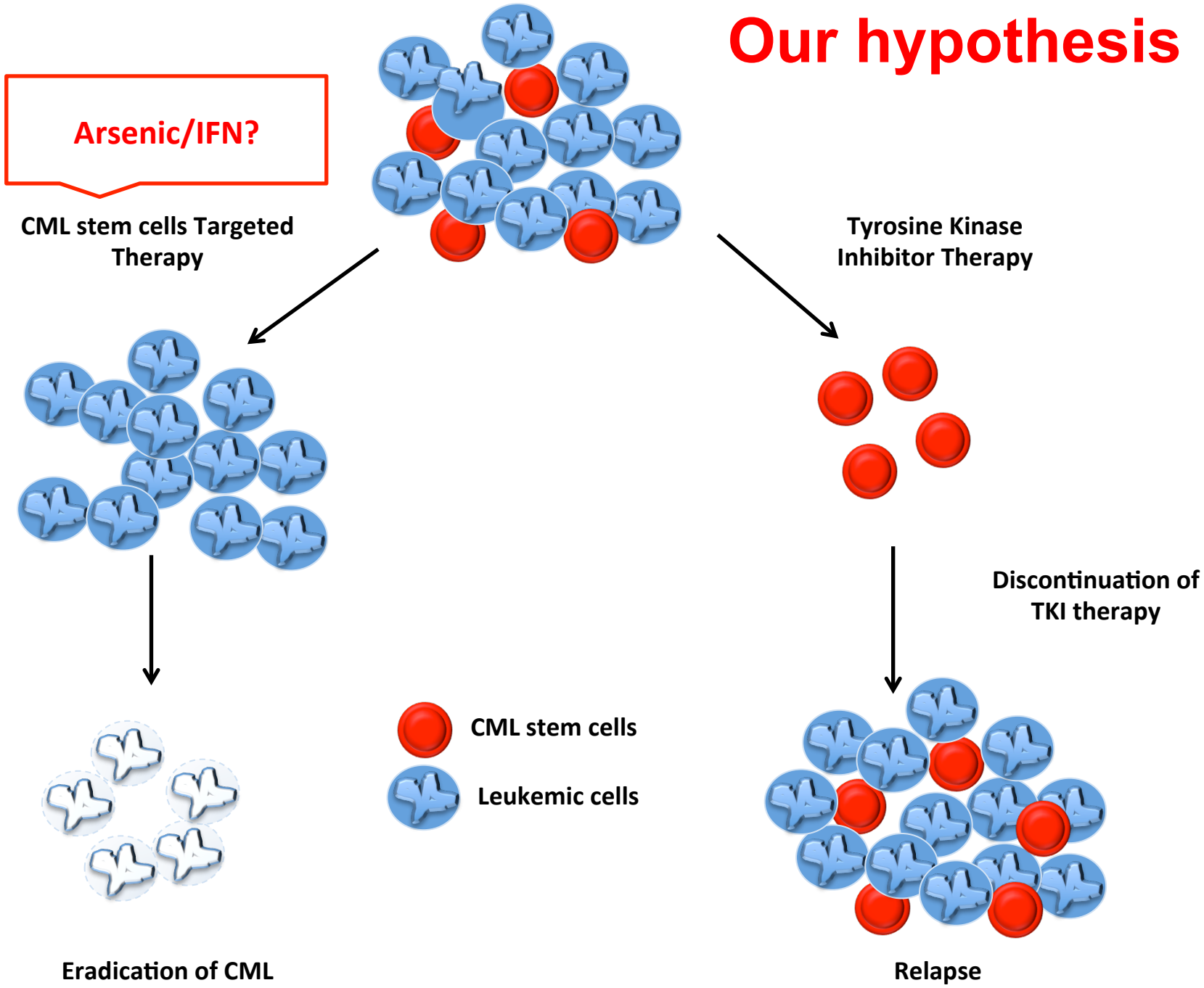
- **No detectable CML CSC?**

TKIs target only leukemic cells, while quiescent CML CSC remain intact: Resistant to all TKIs

For Cure of CML,

new therapeutic options are needed

Our hypothesis

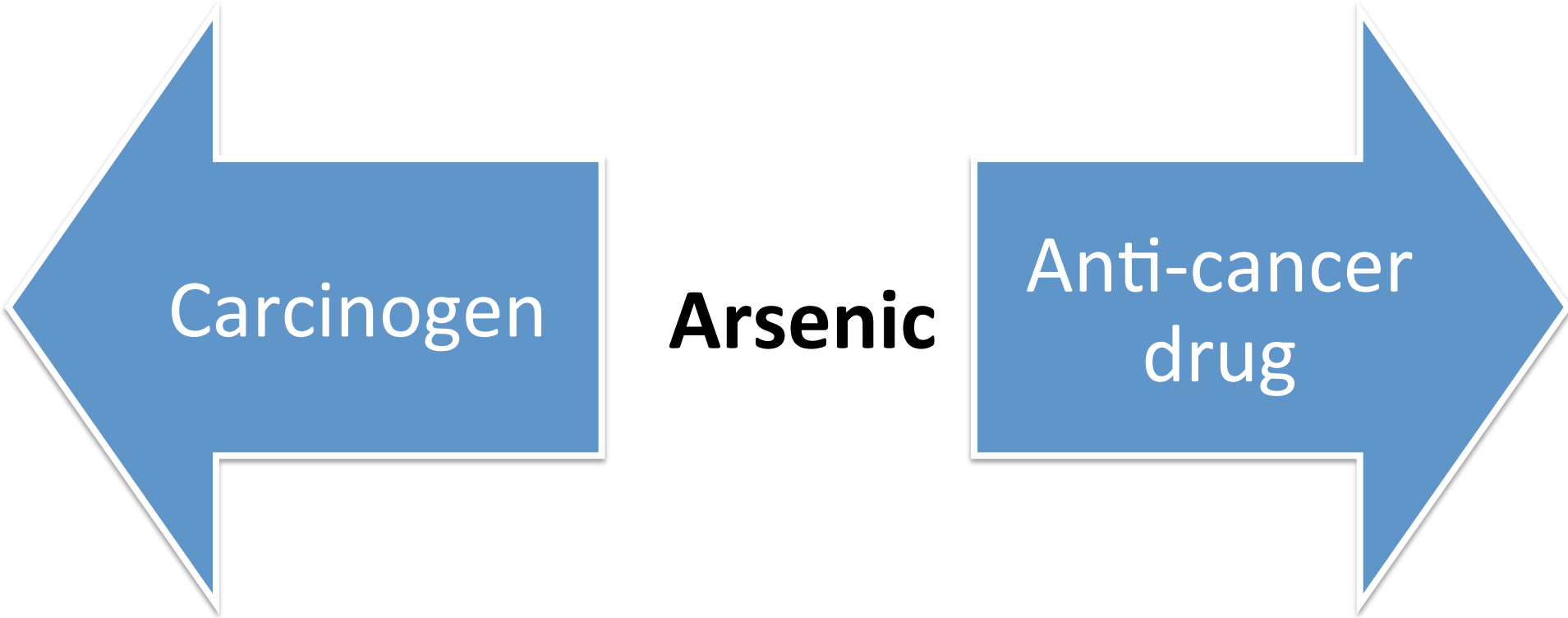


Why Interferon?

- Some CML patients pretreated with IFNa and then switched to imatinib experienced persistent remission (Predhomme et al., 2010)

- Combining IFN with TKI minimize failure and increase the number of CML patients with deep MR (Nicollini et al., 2010)

Why arsenic?



Why arsenic?

Arsenic → oncoprotein degradation

Acute Promyelocytic Leukemia

eradicates APL LIC through PML-RAR degradation

(*Nasr et al.,* Nature Medicine, 2008)

Adult T-Cell Leukemia

eradicates ATL LIC through Tax degradation

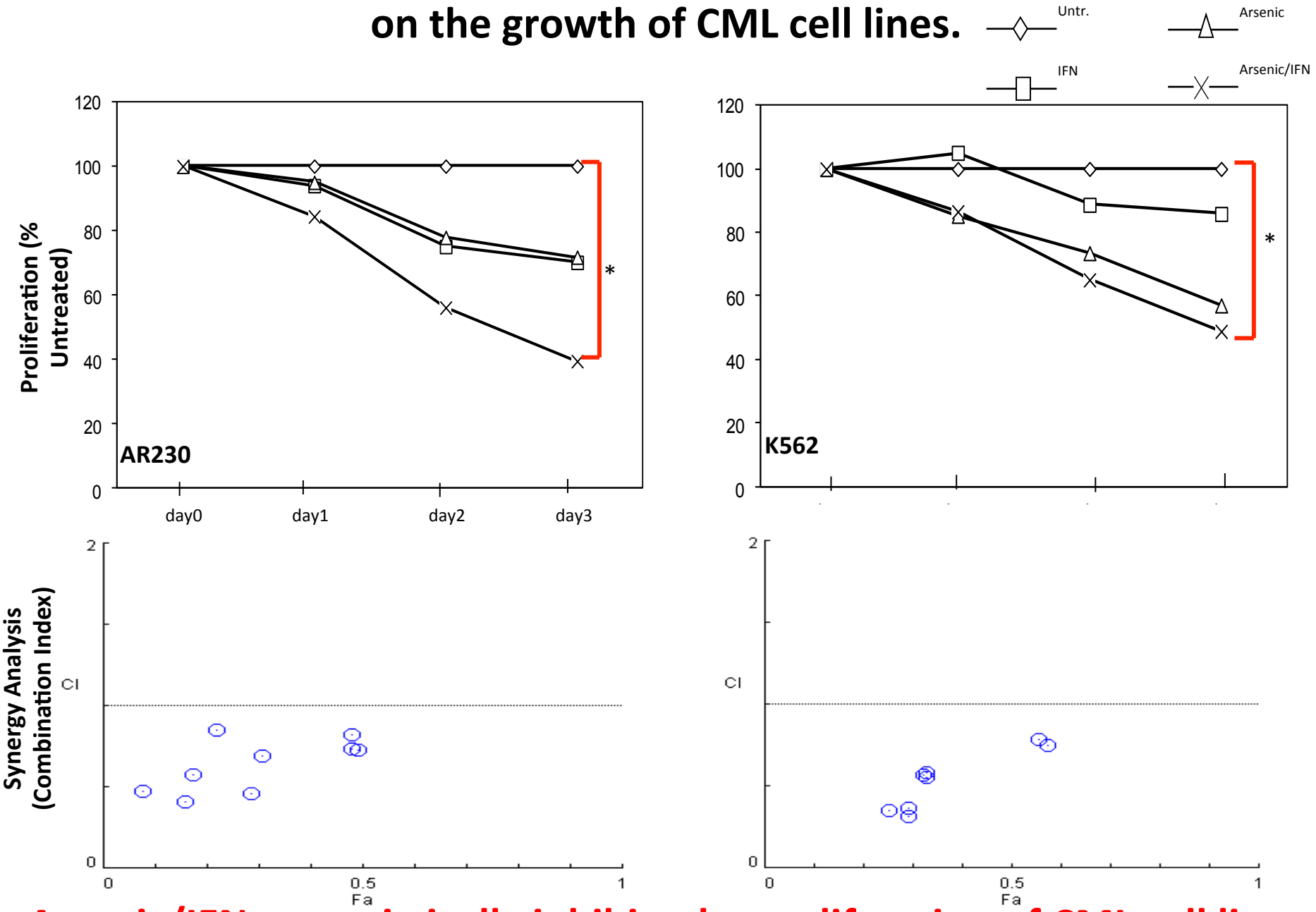
(*Nasr et al.,* Blood, 2003; El hajj et al., JEM 2010)

Arsenic was FDA approved and is already used in the therapy of leukemia with considerable efficacy and acceptable toxicity

What type of models?

- CML cells lines sensitive and resistant to imatinib
- Bone marrow cells from CML pts **(IRB approval)**
- CML mouse model sensitive and resistant to TKI **(IACUC approval)**

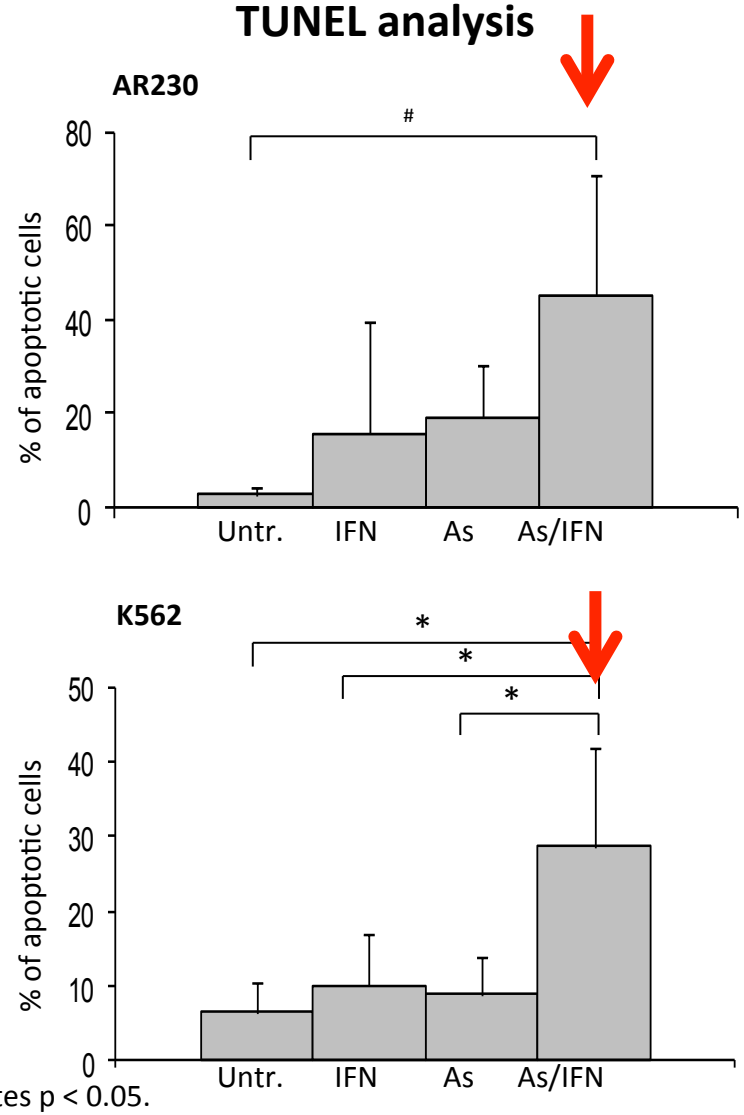
Effects of arsenic trioxide and interferon alpha on the growth of CML cell lines.



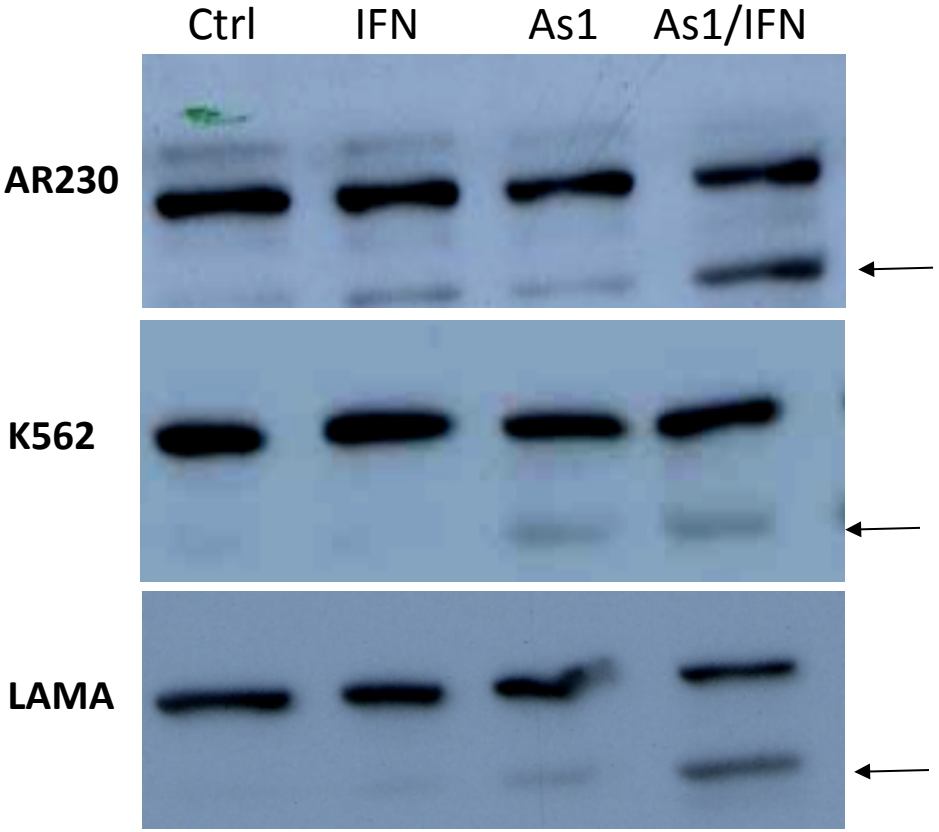
Arsenic/IFN synergistically inhibits the proliferation of CML cell lines

Effects of arsenic and interferon on the induction of apoptosis in CML cell lines

TUNEL analysis



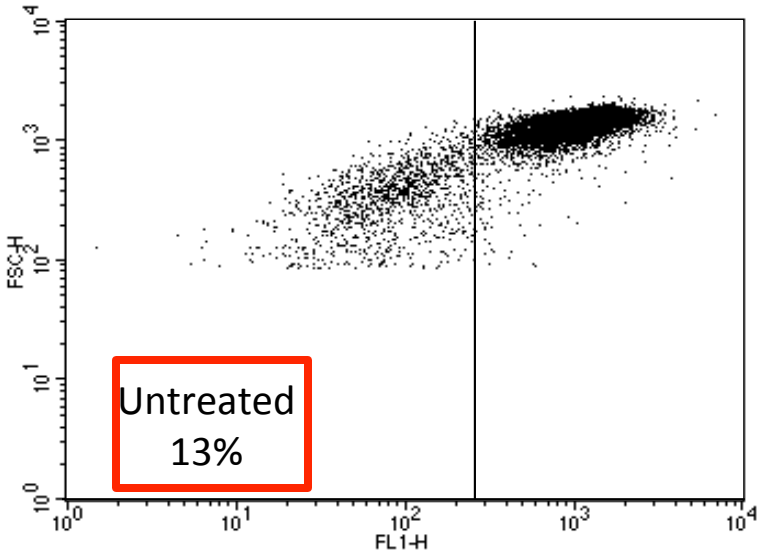
PARP cleavage

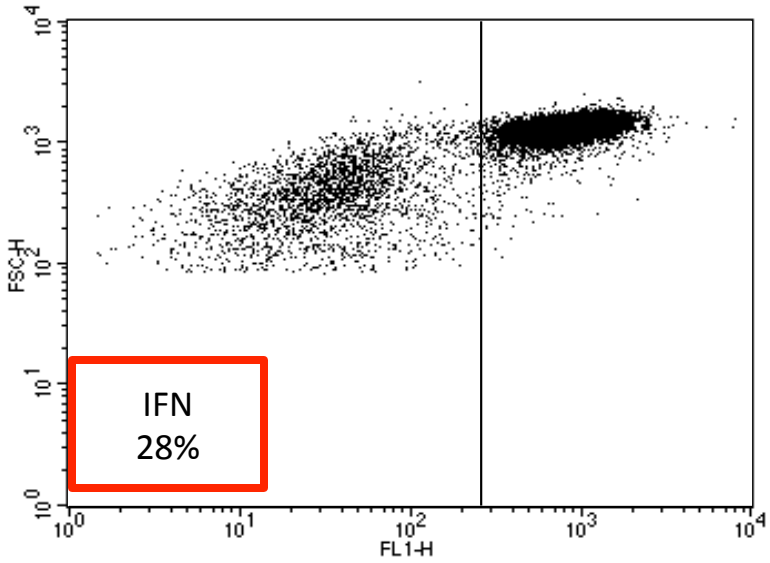


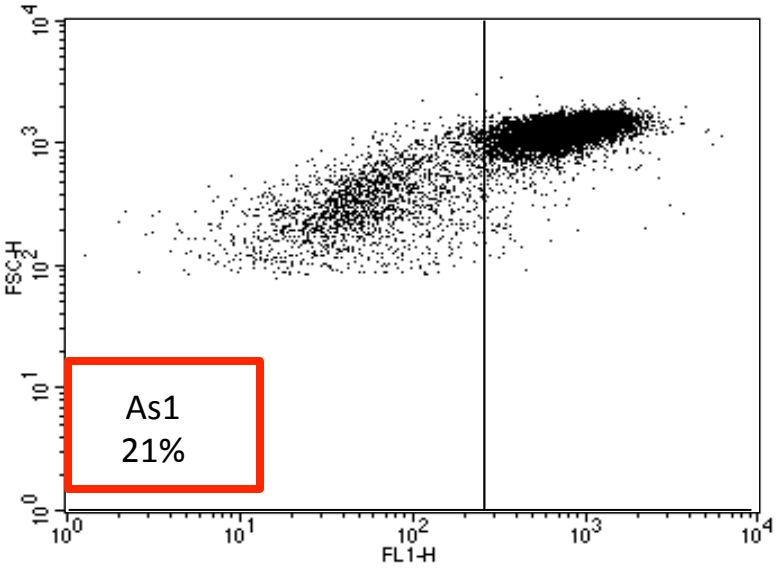
# denotes $p < 0.05$.
* denotes $p < 0.01$

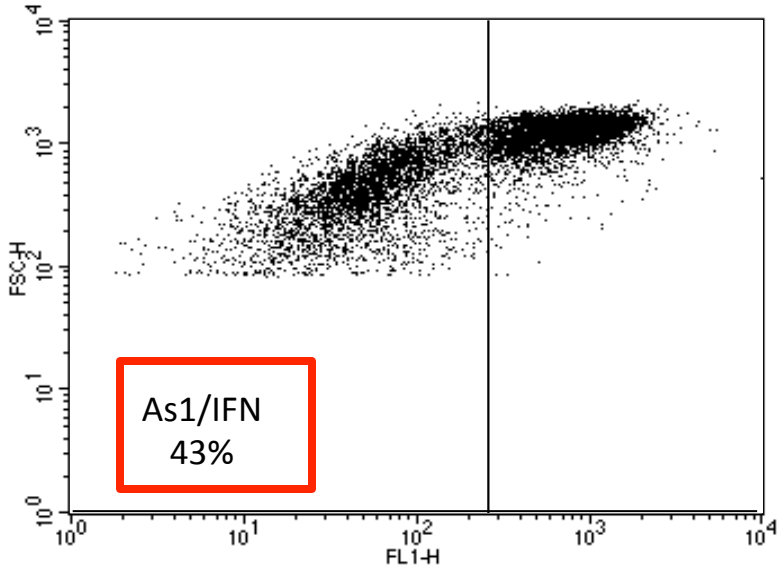
Arsenic/IFN induces caspase-dependent apoptosis in CML cells

Rhodamine-123 staining of AR230



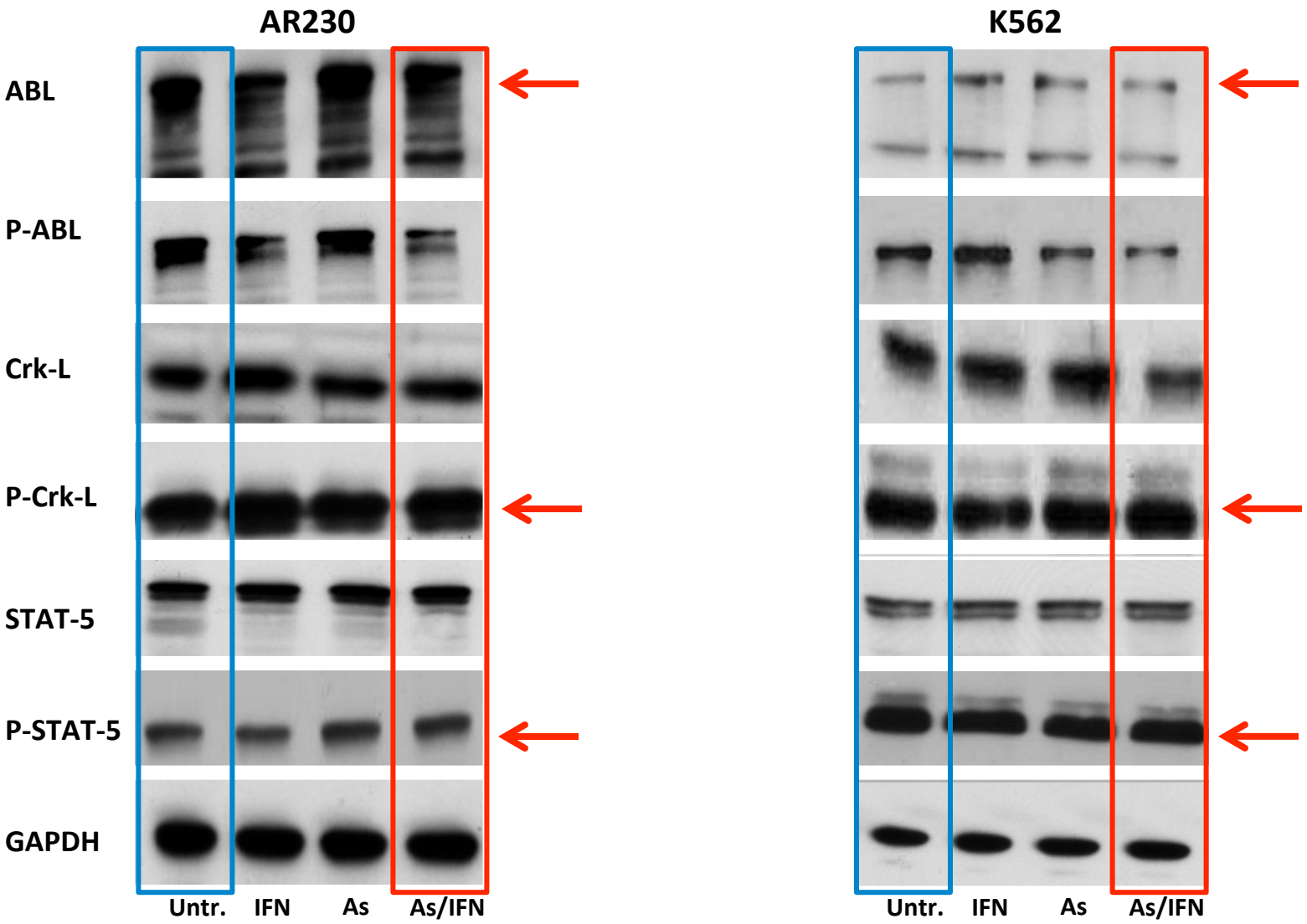






Arsenic/IFN-induced apoptosis was accompanied by collapse of mitochondrial membrane potential in CML cells

Effect of Interferon alpha and arsenic trioxide on BCR-ABL level and activity



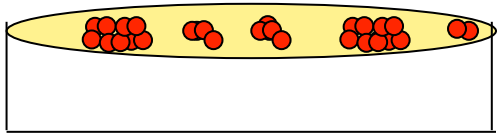
Arsenic/IFN did not affect BCR-ABL levels and activity in CML cells

Effects of arsenic/IFN on primary CML cells

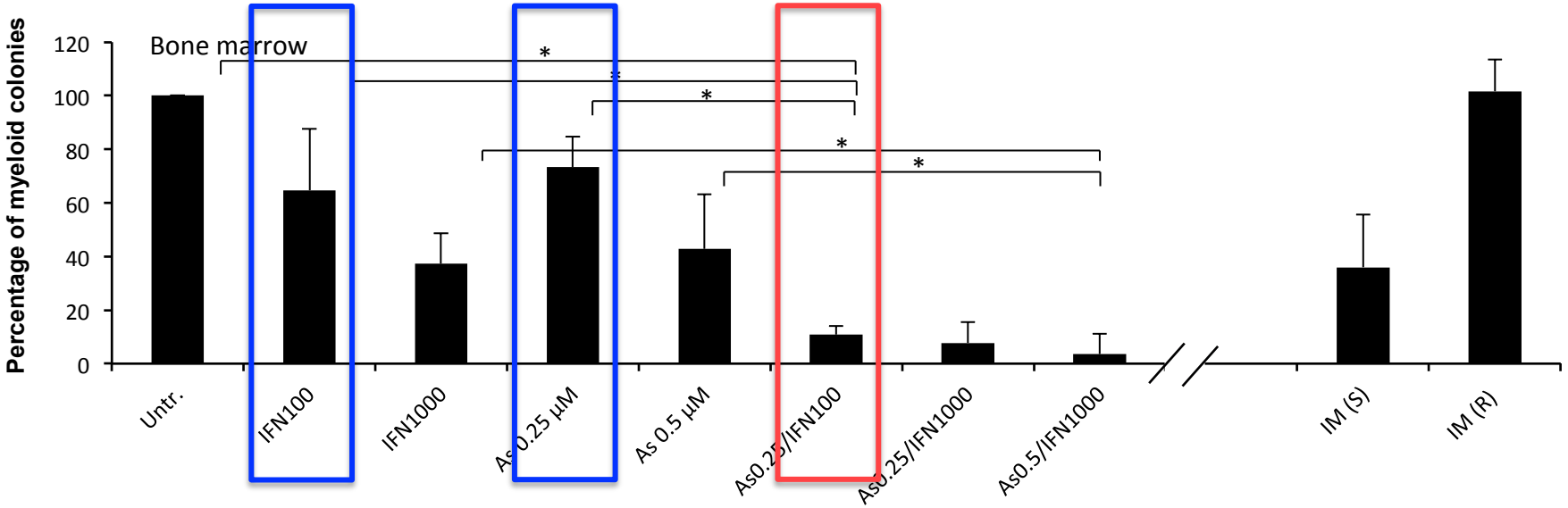
BM or CD34+ cells

Colony Forming Cell Assay

2 weeks in MC +
IL-3, SCF, G-CSF, EPO

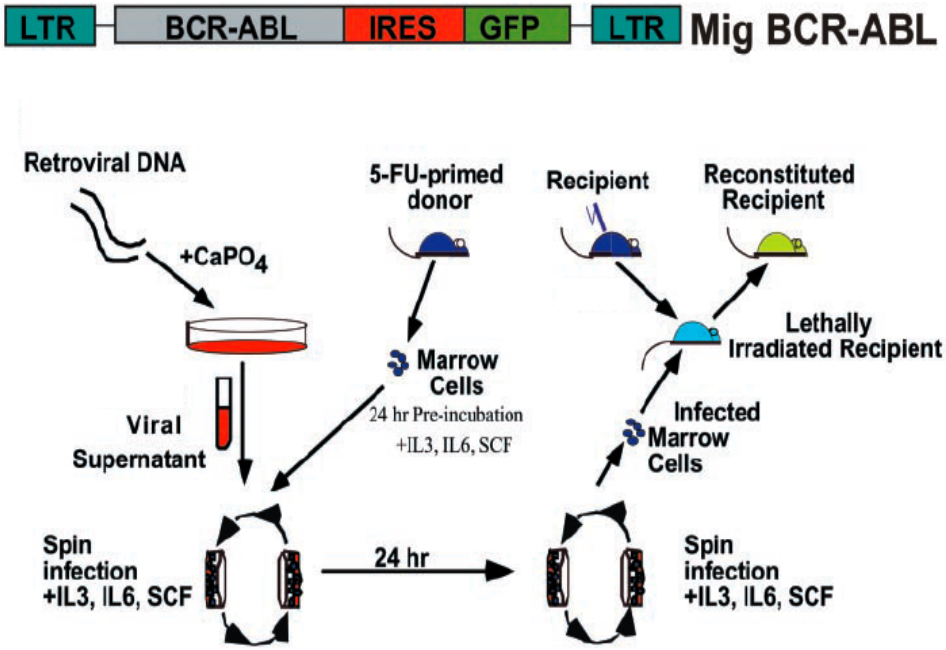


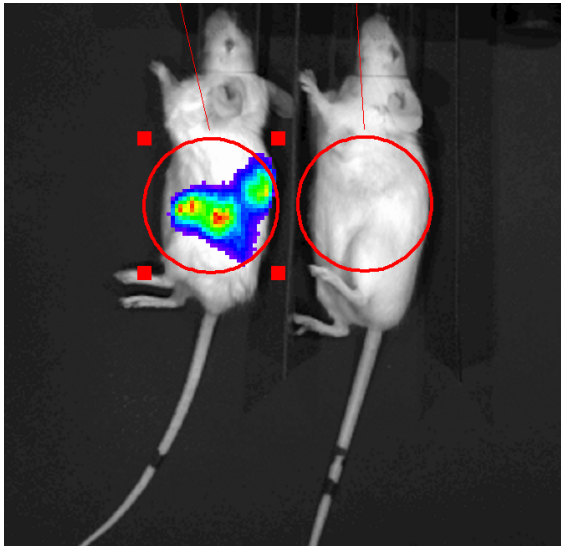
CFC (G, GM, E, GEMM)



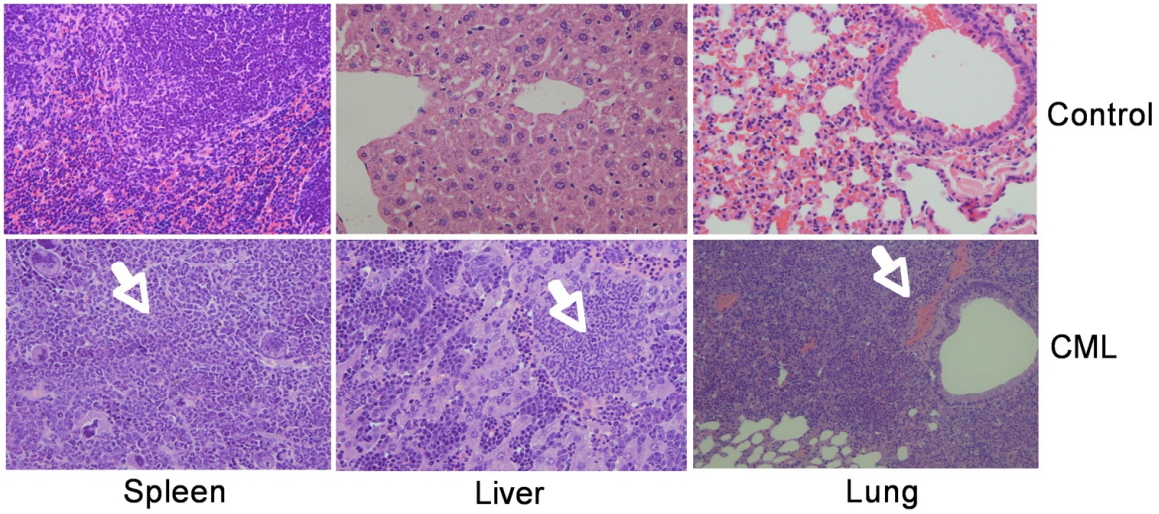
Arsenic and IFN inhibit the clonogenicity of primary CML progenitors

CML mouse model

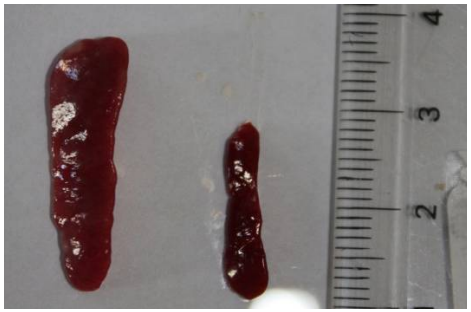




Luciferase imaging of a normal and leukemic Balb/c mice.

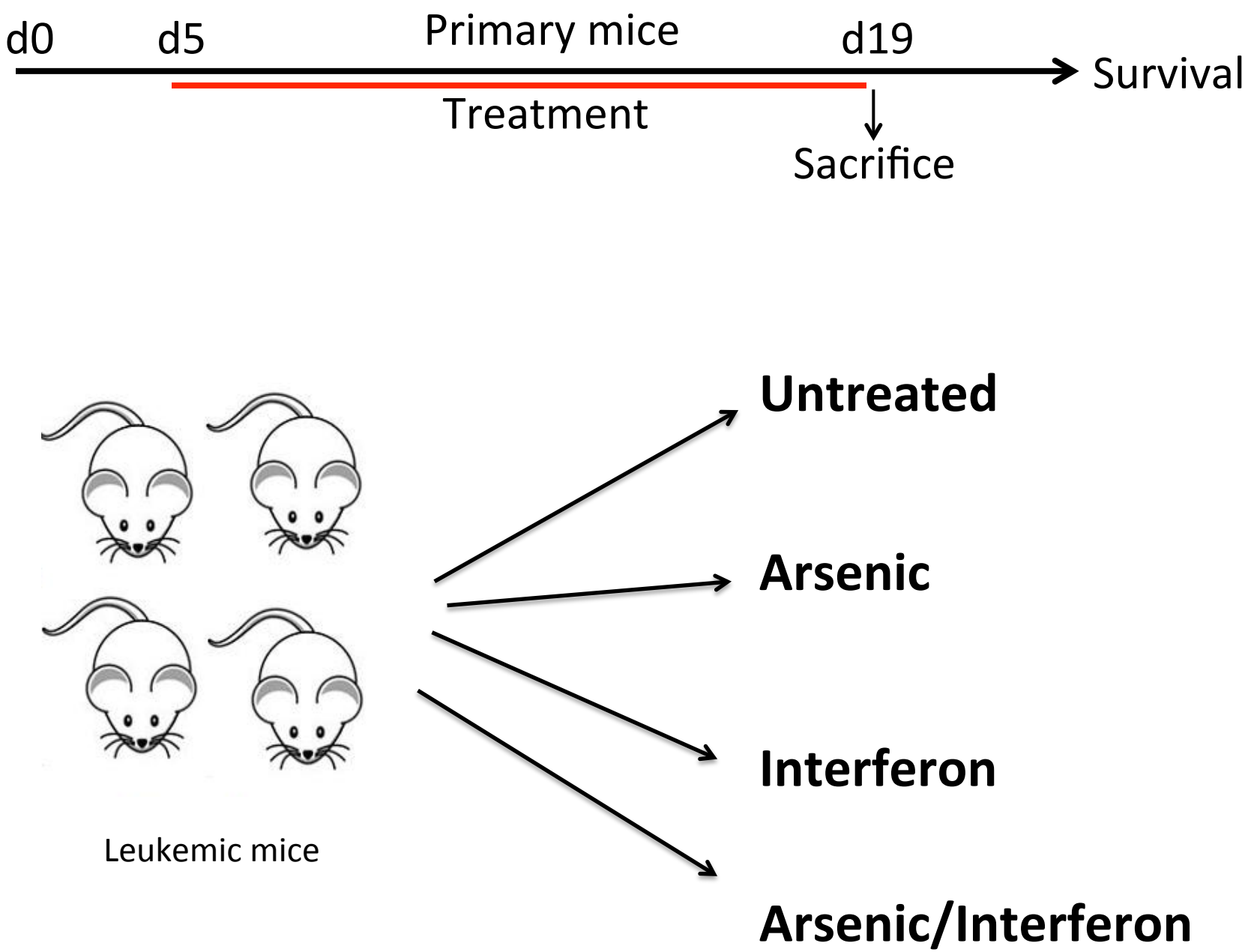


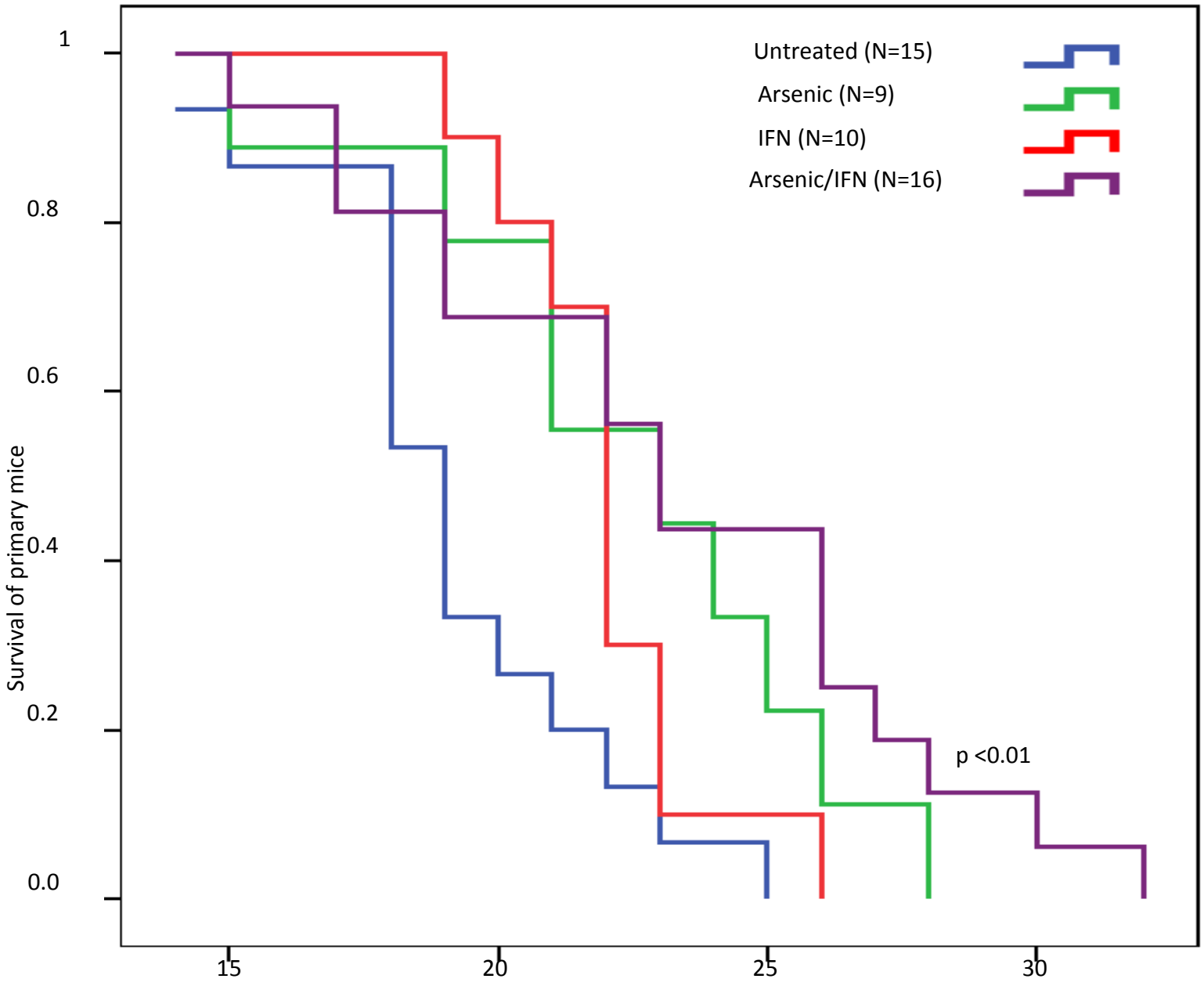
Tissue infiltration with leukemic cells



Comparison of spleen size between a normal (0.1 g) and leukemic Balb/c mice (0.6 g).

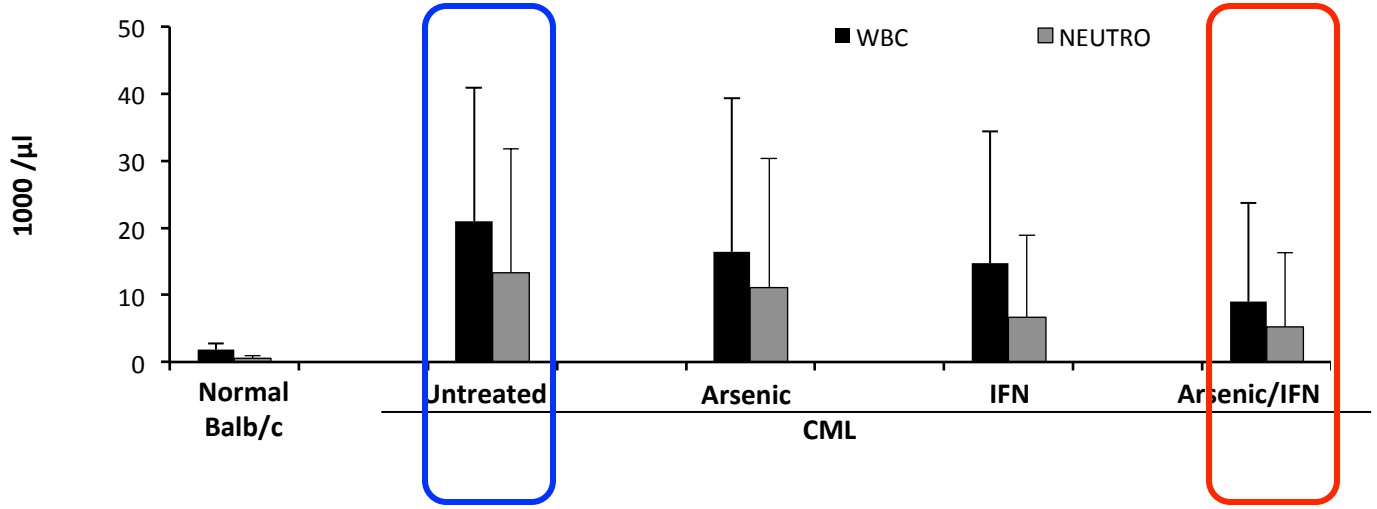
Pear et al., 1998

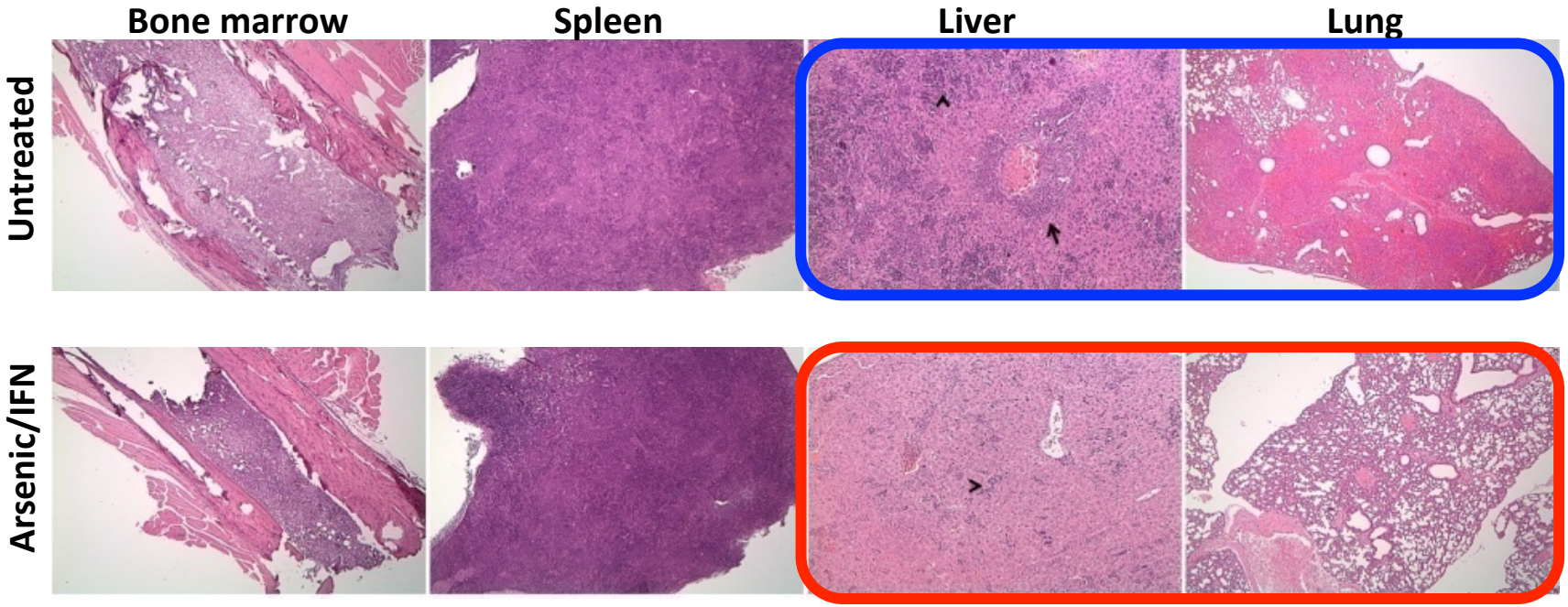

d0
d5
Primary mice
d19
Survival
Treatment
Sacrifice
Leukemic mice
Untreated
Arsenic
Interferon
Arsenic/Interferon



Arsenic/IFN prolongs the survival of primary CML mice

Effect of As/IFN on leukemic infiltration





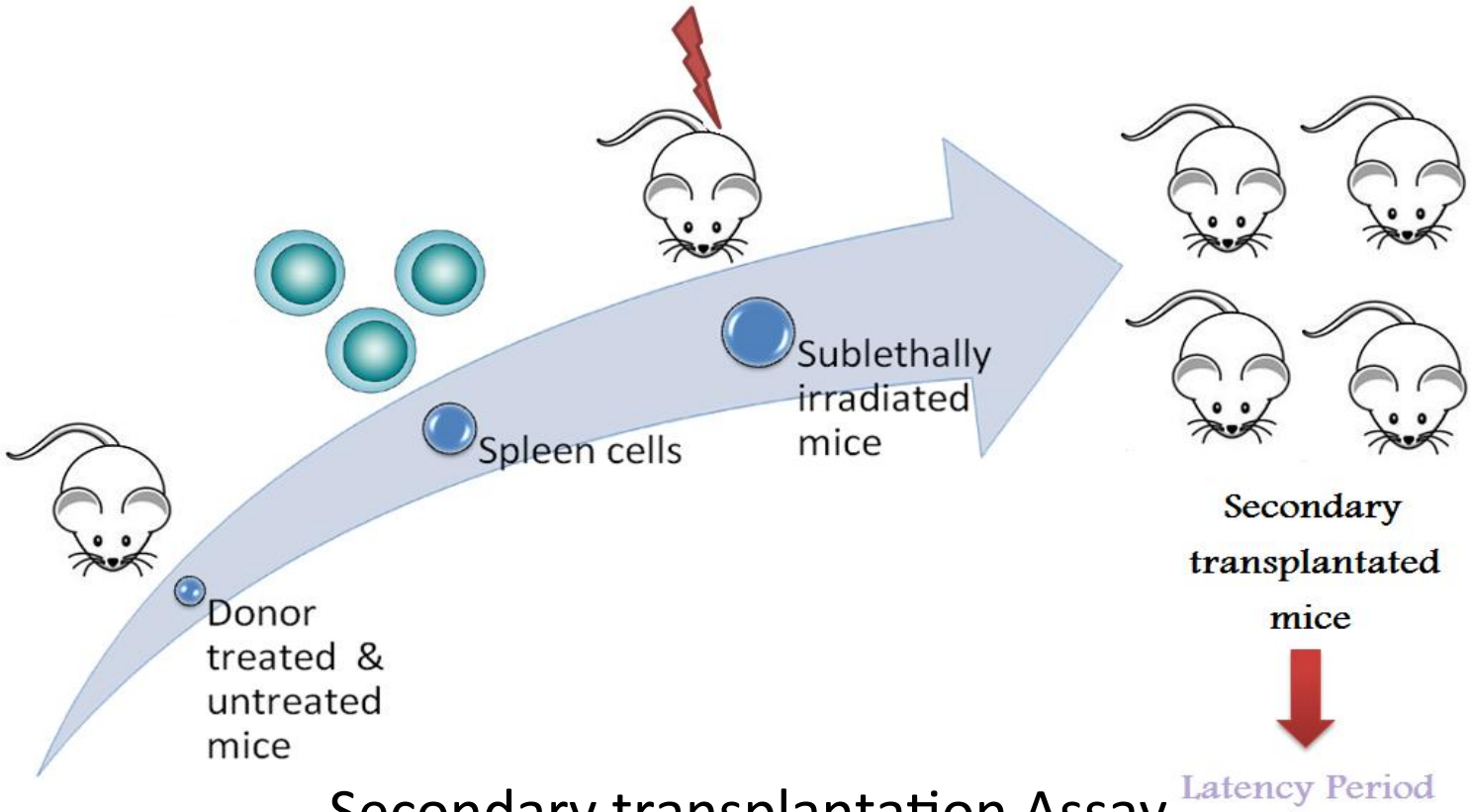
Arsenic/IFN decreases leukemic infiltrate mainly in liver and lung but did not significantly decrease liver and spleen weights

d0 d5 Primary mice d18

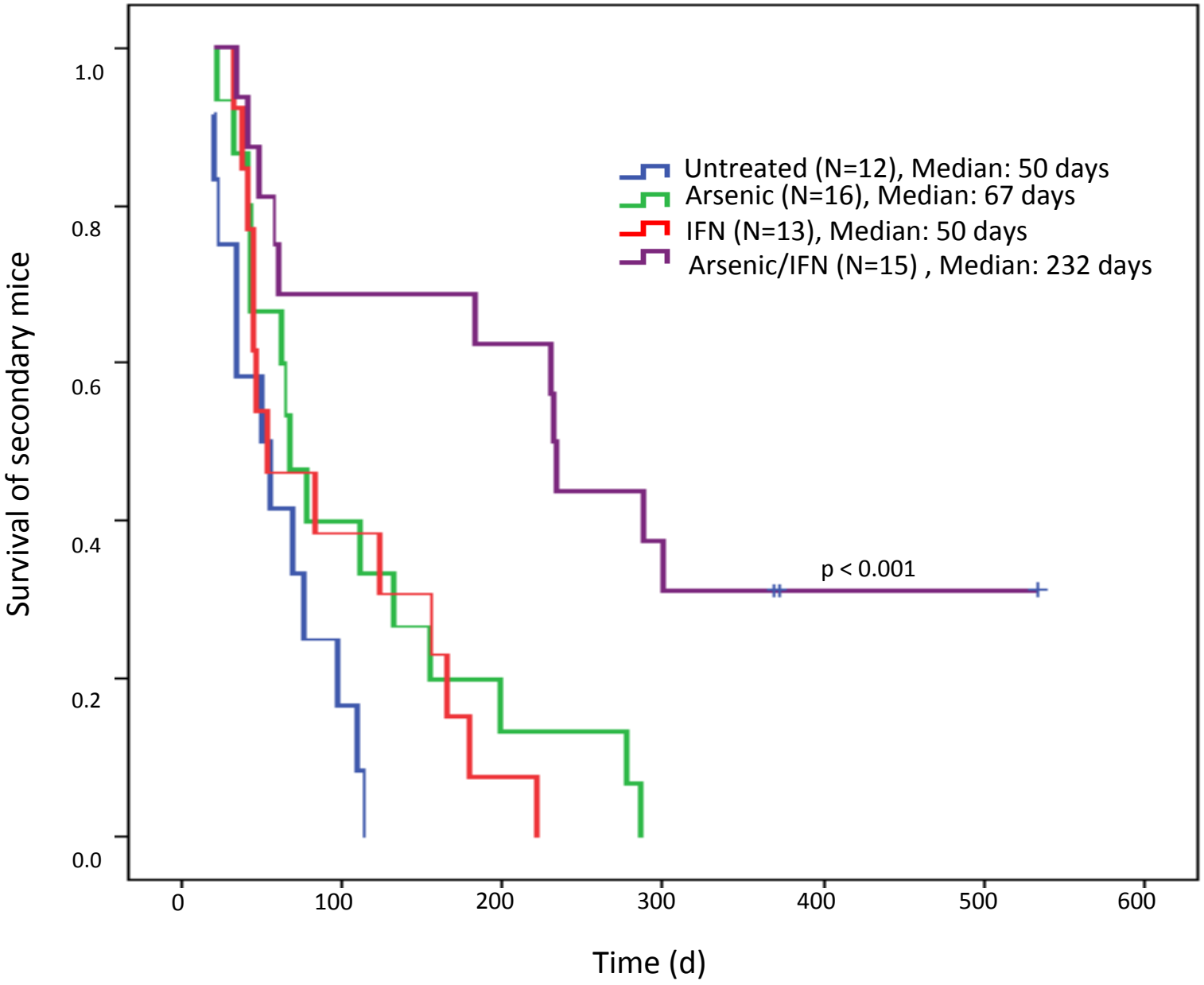
Treatment

Sacrifice

Transplantation into untreated secondary mice → Survival



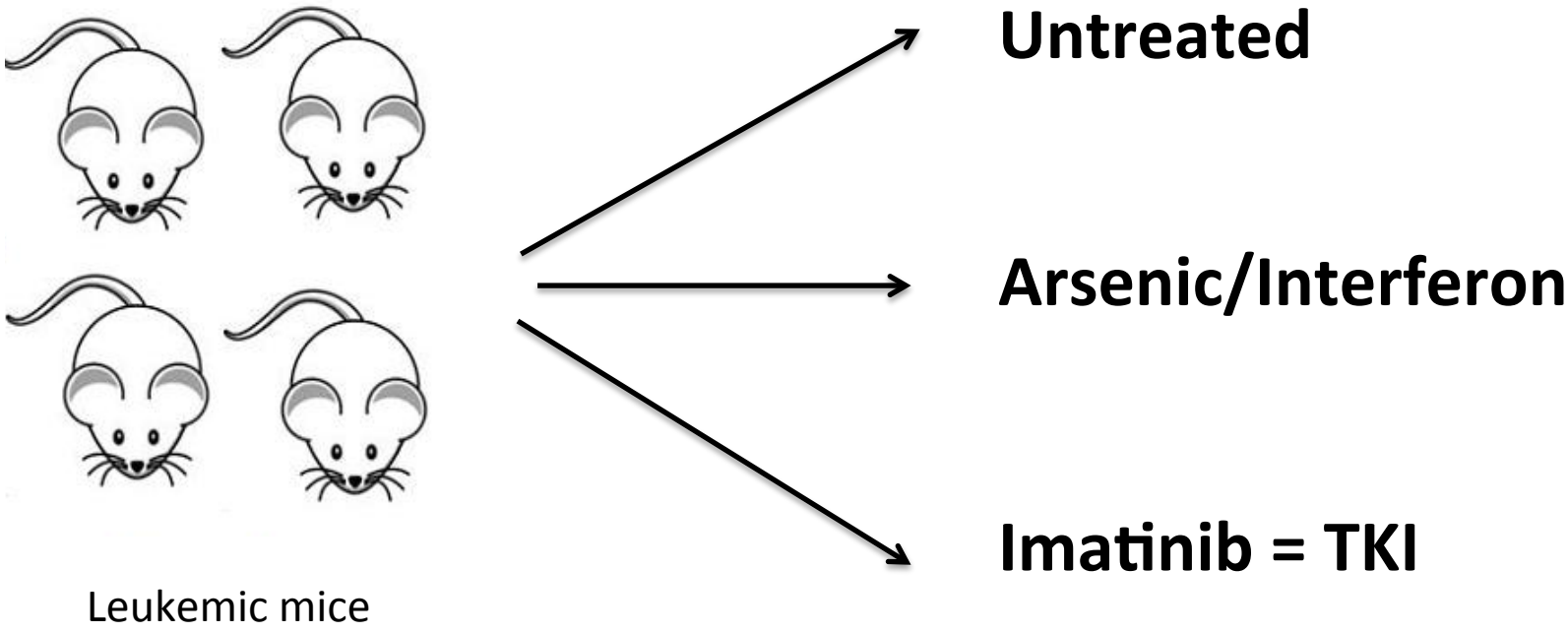
Secondary transplantation Assay

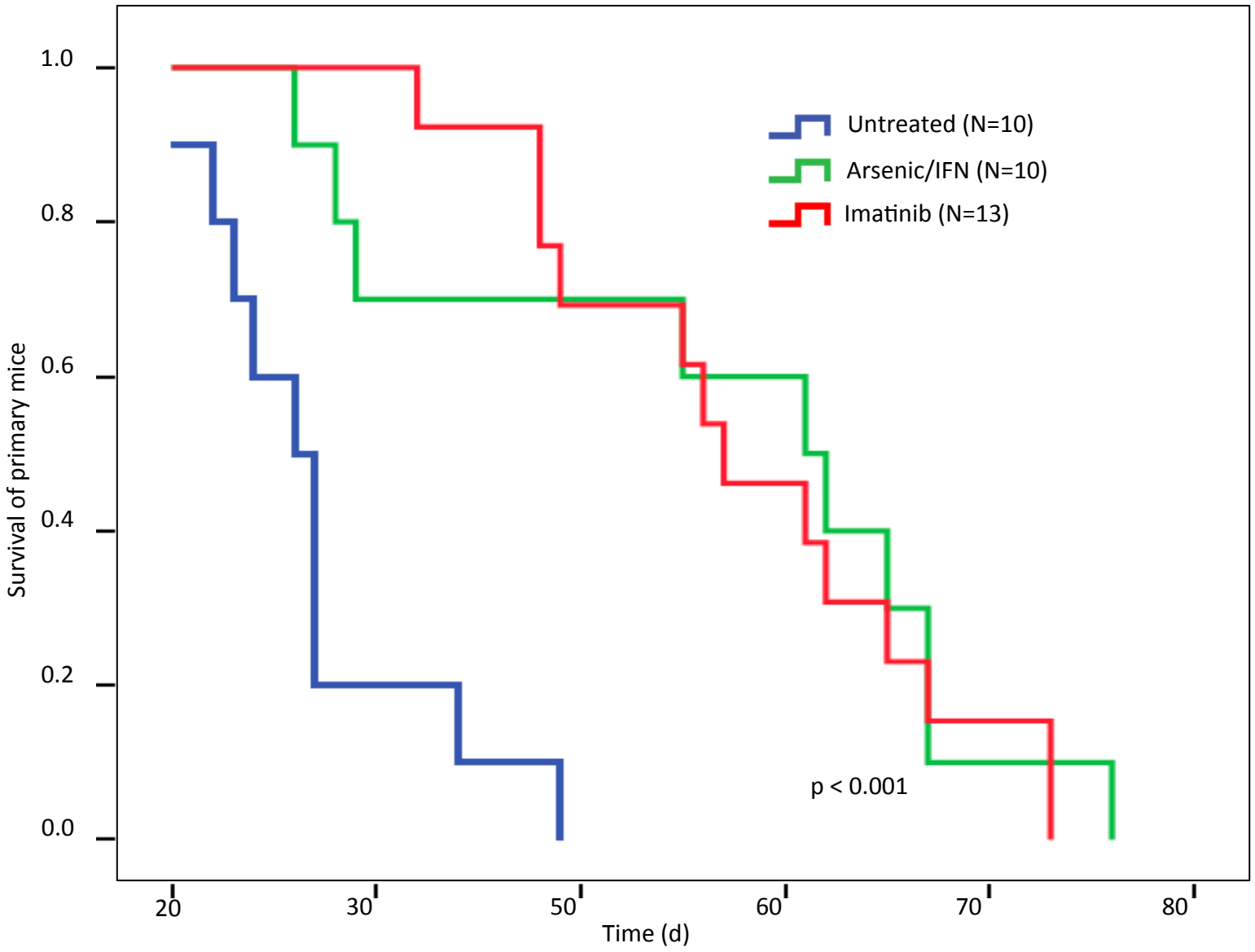


Arsenic/IFN treatment targets CML LIC and cures CML mice

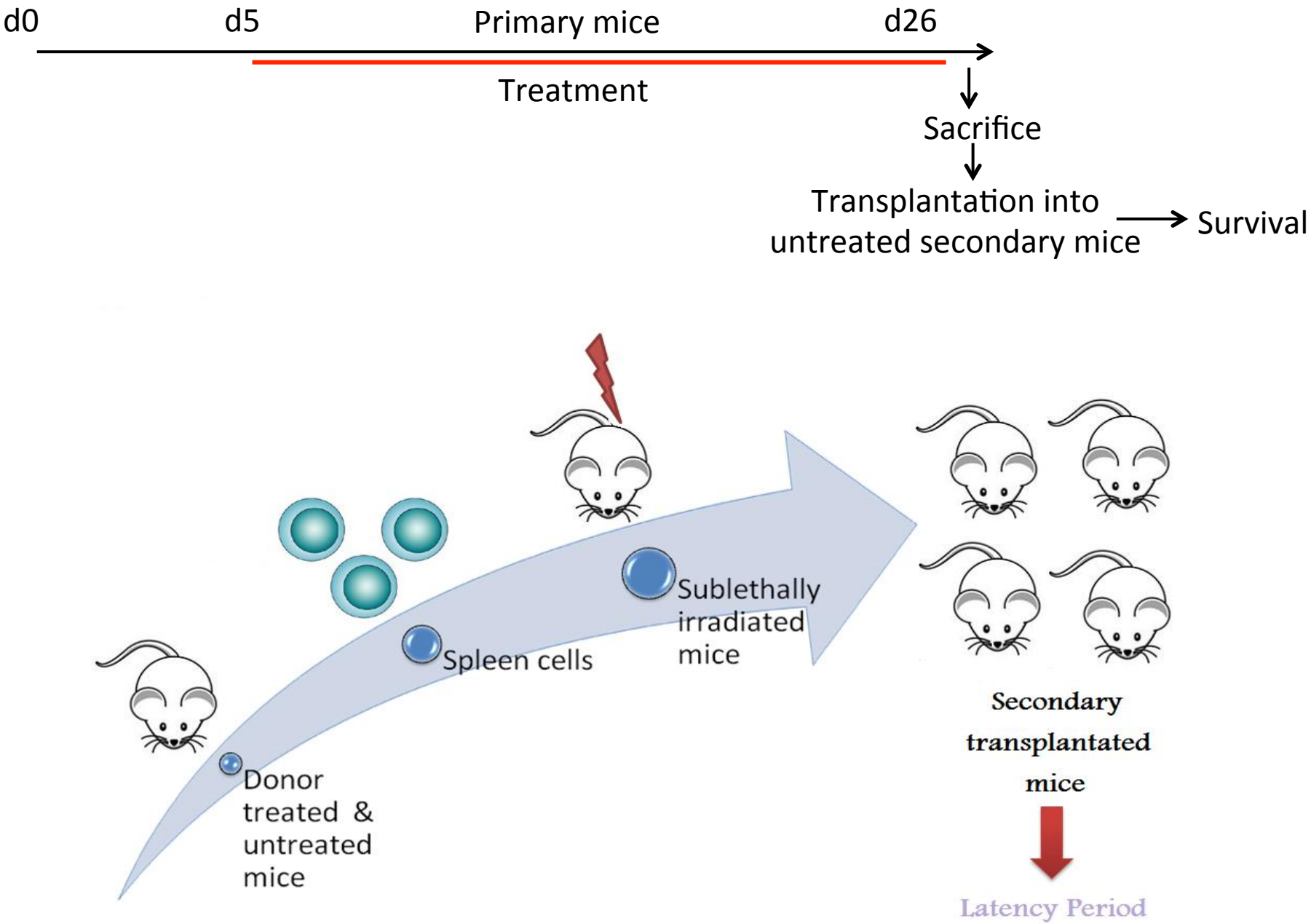
Is Arsenic/IFN better than Imatinib?

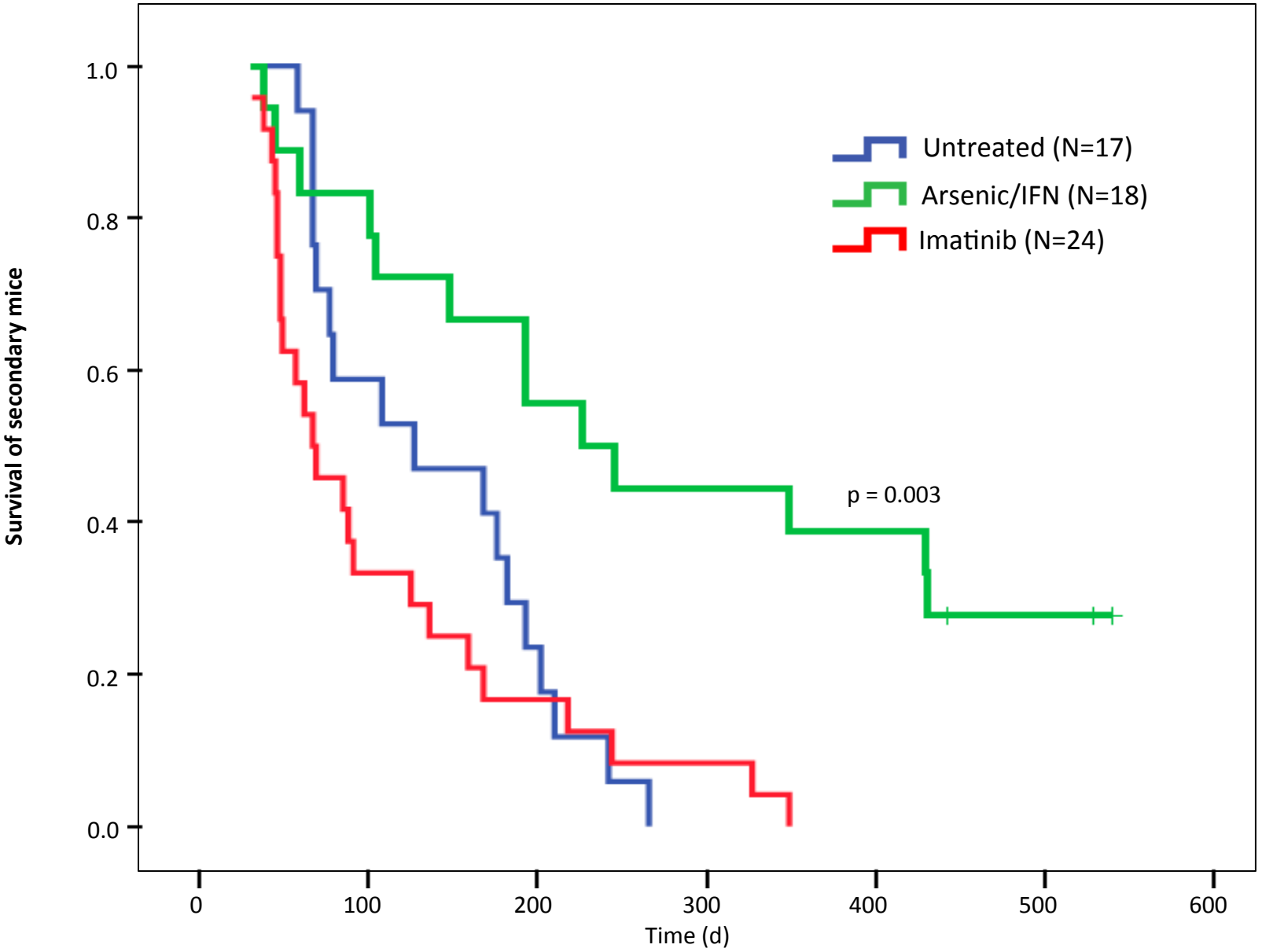

d0
d5
Primary mice
d26
Survival
Treatment
Sacrifice


Untreated
Arsenic/Interferon
Imatinib = TKI
Leukemic mice

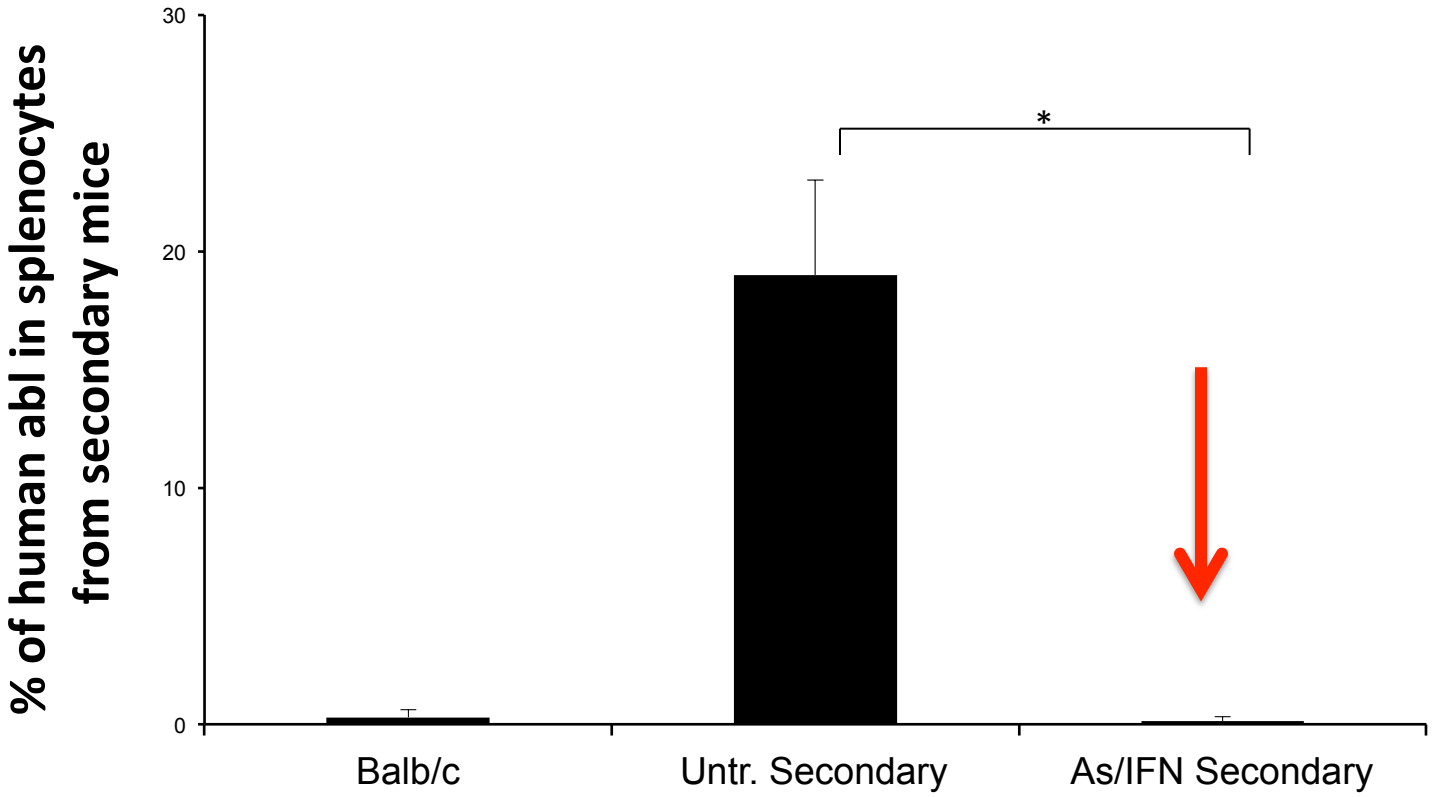


Arsenic/IFN is clinically comparable to IM in primary CML mice.


d0
d5
Primary mice
d26
Treatment
Sacrifice
Transplantation into
untreated secondary mice
Survival
Sublethally
irradiated
mice
Spleen cells
Donor
treated &
untreated
mice
Secondary
transplantated
mice
Latency Period



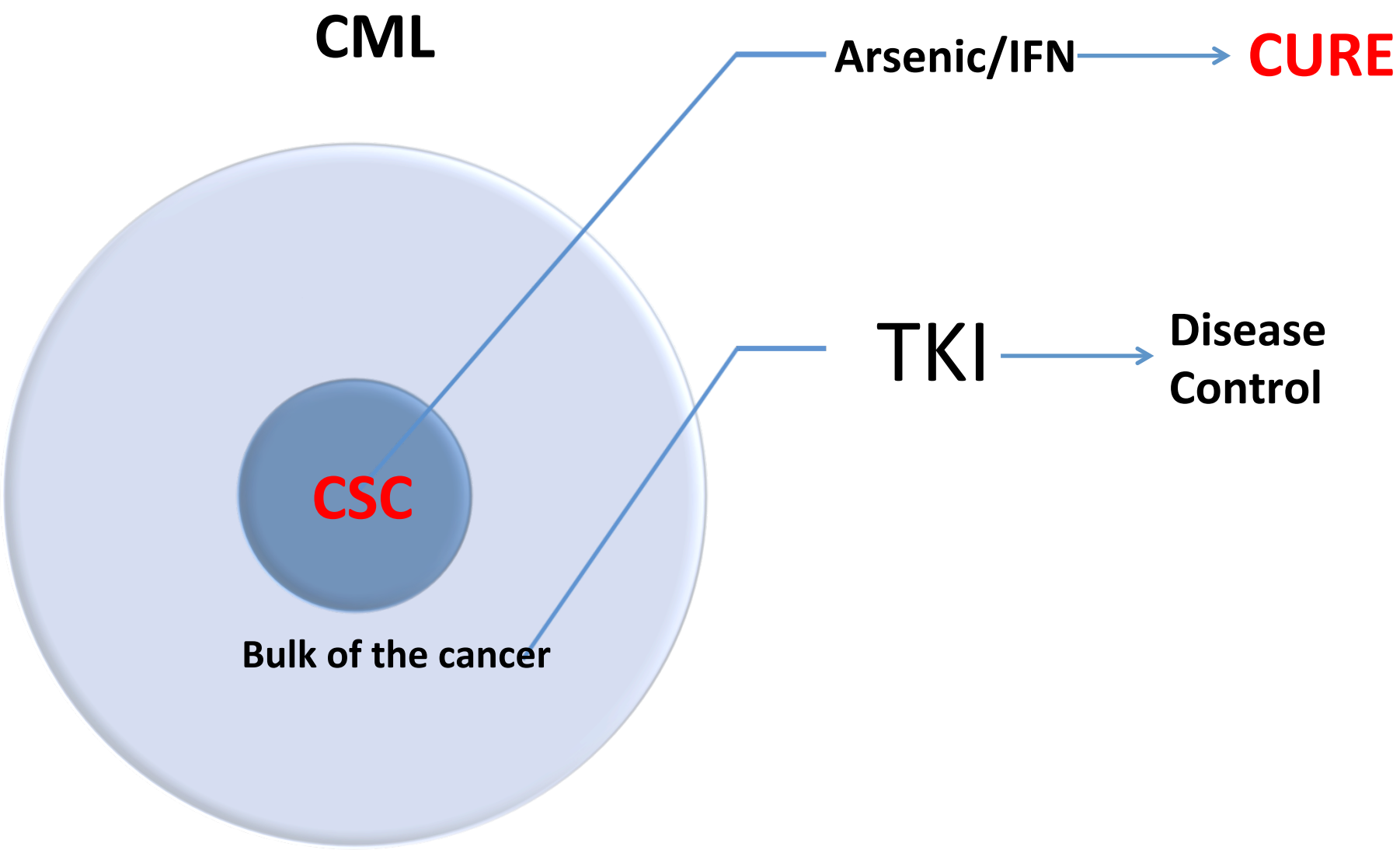
A significant prolongation in survival was observed in secondary recipients of spleen cells from primary mice treated with arsenic/IFN



Arsenic/IFN treatment cures CML mice

Conclusions

- Arsenic/IFN inhibits proliferation and induces apoptosis of CML cell lines *in vitro*
- Arsenic/IFN inhibits clonogenic capacity of CML cells *ex vivo*
- Arsenic/IFN prolongs survival of CML mice *in vivo*
- Arsenic/IFN cures some CML mice *in vivo*



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Effective targeting of chronic myeloid leukemia initiating activity with the combination of arsenic trioxide and interferon alpha.

El Eit RM, Iskandarani AN, Saliba JL, Jabbour MN, Mahfouz RA, Bitar NM, Ayoubi HR, Zaatari GS, Mahon FX, De Thé HB, Bazarbachi AA, Nasr RR.

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To raise awareness about the importance of cancer research


AMALOUNA
Cancer is limited.
Research isn't
the smallville hotel
Four Seasons Hotel
Beirut
the postoffice

THANK YOU