MLV leukemogenesis in laboratory mouse models : (at least) two-hit mechanisms of oncogenesis

Marc Sitbon

Directeur de recherche Inserm

- Retroviral Replication and Pathogenesis -

Institut de Génétique Moléculaire de Montpellier (IGMM)

CNRS-UM1 & UM2

sitbon@igmm.cnrs.fr

Classification of vertebrate Retroviruses

(based on *gag-pol* sequence phylogeny)

Genus	Examples
Alpharetrovirus	*(Avian leukemia and sarcoma viruses, ALV, RSV…)*
Betaretrovirus	*(MMTV, MPMV, sheep Jaagsiete retrovirus...)*
Gammaretrovirus	*(Murine leukemia viruses MLV, feline FeLV, gibbon GaLV, human (?) XMRV)*
Deltaretrovirus	*(Human HTLV, simian STLV, bovine BLV)*
Epsilonretrovirus	*(Walleye dermosarcoma virus, WDSV…)*
Lentivirus	*(primate HIV, SIV; ongulate Visna, CAEV…; feline FIV…)*
Spumavirus	*(Foamy viruses, primate, feline, bovine…)*

Transposable elements in the human genome

Transposable Elements, ~45%; $3x10^6$

RNA intermediate

Transposons, 2.8%; $0.3x10^6$

Retroelements, 42.2%; $2.7x10^6$

non LTR, 33.9%; $2.4x10^6$

SINE (Alu, 10.6%; MIR, 2.5%)

LINE (L1, 16.9%; L2, 3.2%)

processed pseudogenes, <1.0%

LTR, 8.3%; $0.3x10^6$

class I ERV + MER4, 2.8%

class II ERV, 0.3%

class III ERV, 4.6%

others (MST, MLT, etc.), 0.6%

Bannert N., Kurth R. PNAS 2004;101:14572-14579

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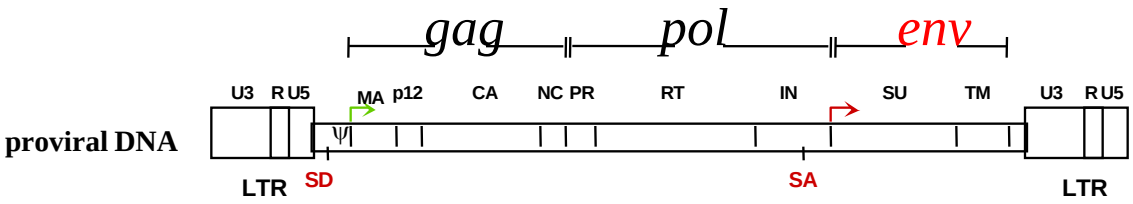
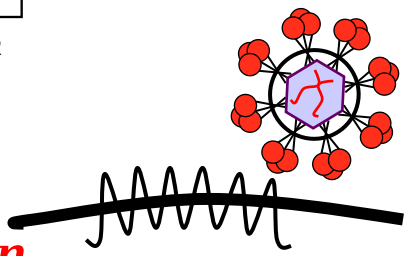
A rapid overview of the MLV group

✧ **MLV belong to the gammaretrovirus genus**

- Extracellular budding, no doubly-spliced mRNA





✧ **Several classes of MLV**

as defined by <u>specific Env/receptor recognition</u>

- Ecotropic MLV (mouse cells)
- Xenotropic/Polytropic MLV (wt mice + non mouse cells)
- Amphotropic MLV (mouse + non mouse cells)
- 10A1, MDEV, McERV (mouse + non mouse cells)
- M813 (mouse cells)

Two main types of MLV-driven oncogenesis

✧ **Rapid onset (2-3 weeks)**

- o Rapid also in adult mice
- o MLV harbors an oncogene
  - from recombination with cellular proto-oncogenes
    - . in *gag* : *abl*, *ras*, *fos*…
    - . in *env* : *mos*, *mpl*...
- o Replication-defective (helper-dependent, i.e needs other MLV to spread)
- o Leukemia/lymphoma; sarcoma

✧ **Slow (4 to > 12 months)**

- o Often faster in newborn mice (2-3 months versus > 6 mos. in adults)
- o No oncogene; insertional mutagenesis (*see presentation by Linda Wolff*)
- o Replication-competent
- o Leukemia/lymphoma

Main parameters influencing MLV-driven leukemogenesis

✧ **Host parameters**

- Age and route of inoculation
  - Newborn versus adults (increased latency broadens leukemic cell types)
  - i.v. versus intrathymic (disease acceleration)
- Genetic background
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- LTR- U3 enhancers (latency and leukemia cell types)
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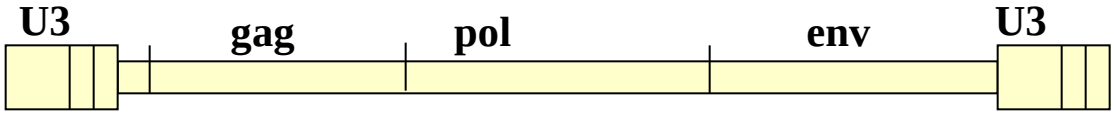
The Friend-MLV and Moloney-MLV leukemia models : Molecular bases for their distinct leukemia cell types

Age of inoculation	Adult mice (Balb/c, CBA, Swiss, etc.)		Newborn mice	
	Leukemia cell type	Latency	Leukemia cell type	Latency
	Erythroid/Lymphoid/ Myelomonocytic	4-10 mos.	Erythroid (100%)	1.5-2.5 mos.

Friend-MLV



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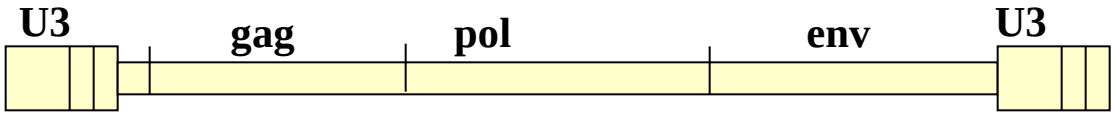
Mouse strains (susceptible)	**Age of inoculation (Balb/c, CBA, Swiss, etc.)**		
	Newborn mice		
		Leukemia cell type	**Latency**
		Erythroid (100%)	**1.5-2.5 mos.**
Mouse strains (resistance genes)	**C57Bl, DBA, etc.**	**Newborn mice**	
		Lymphoid/ Myelomonocytic	**6-15 mos.**

Friend-MLV



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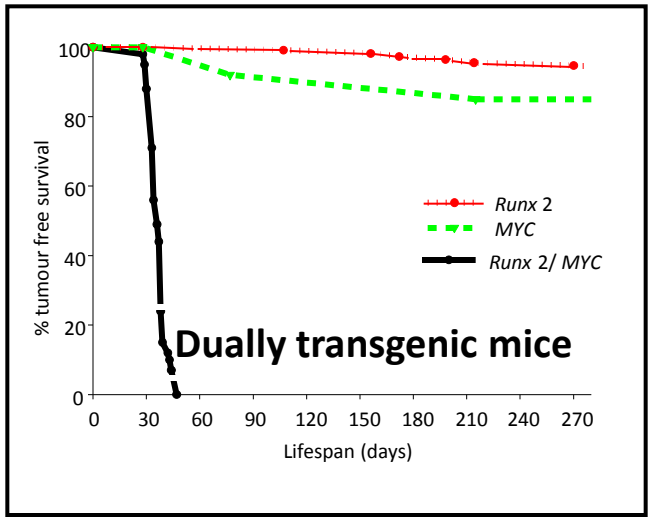
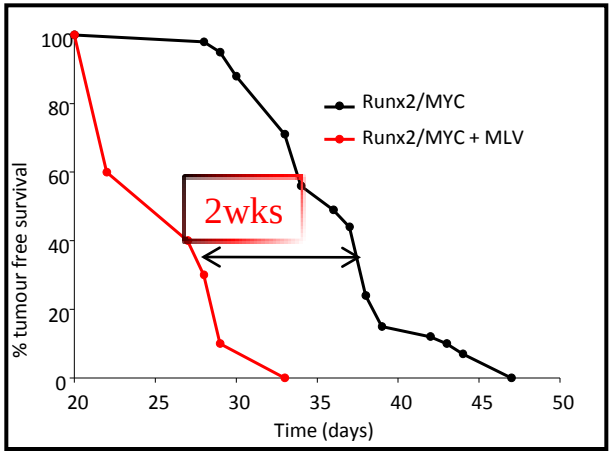
MYC/Runx collaboration : search for a third hit

RUNX and MYC synergism → A 2 week acceleration by MLV



+ Mo-MLV →

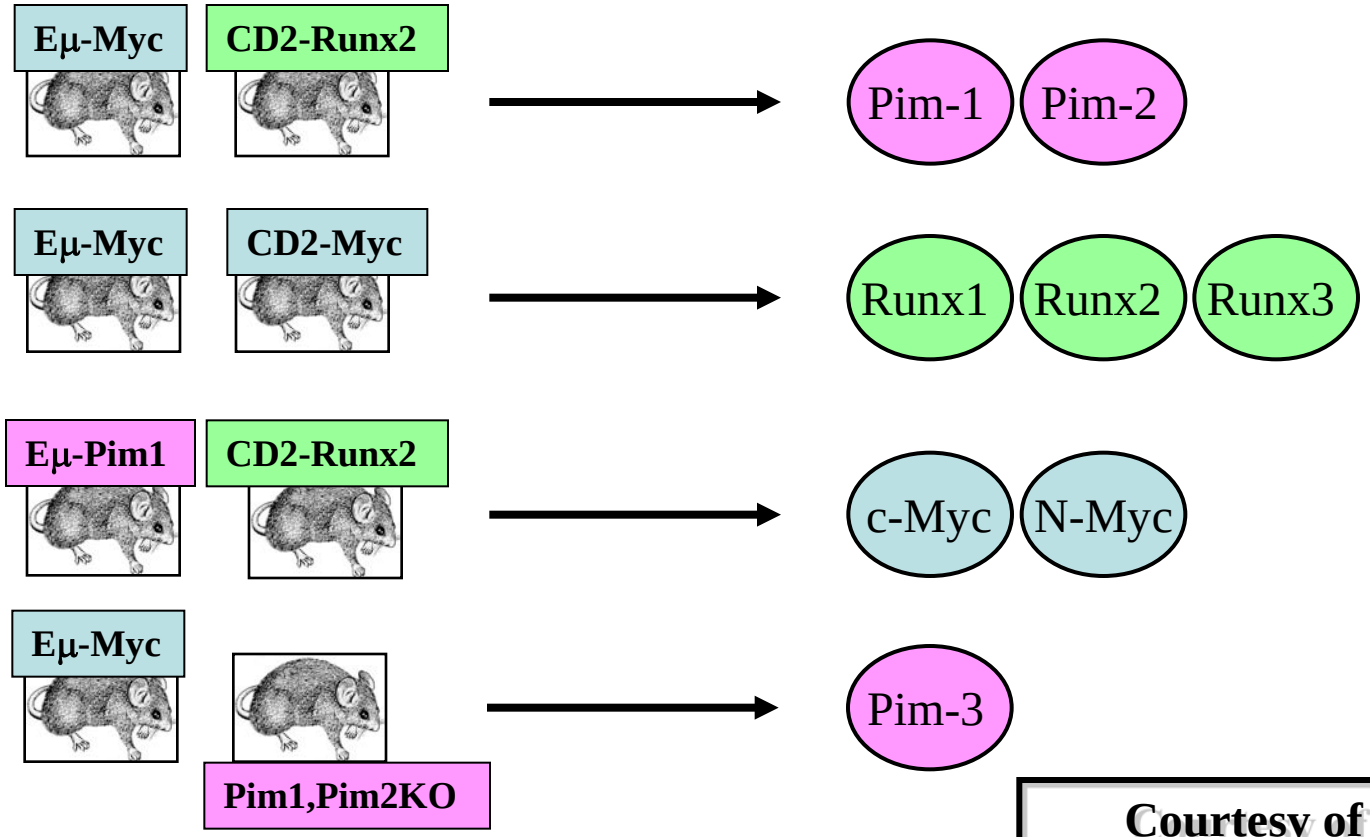


from a 2-hit/oncogene mechanism to a a 3rd hit ?

Courtesy of **James Neil**
Faculty of Veterinary Medicine
Glasgow

Collaboration tagging : 2nd hit genes show reciprocity and complementation

Strain background | Retroviral targets



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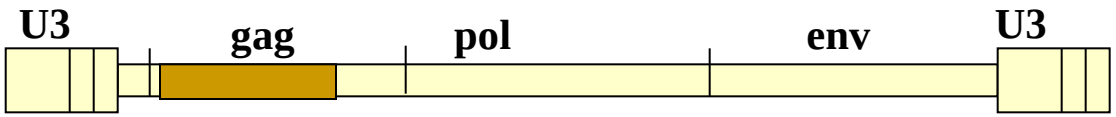
The Friend-MLV and Moloney-MLV leukemia models : Molecular bases for their distinct leukemia cell types

Conditioning	Age of inoculation (Balb/c, CBA, Swiss, etc.)			
	Pristane-pretreated adult mice		Untreated adult mice	
	Leukemia cell type	Latency	Leukemia cell type	Latency
Moloney-MLV	Promonocytic (60%)	4-6 mos.	T cell leukemia (< 50%)	> 6-10 mos.
Main sequences involved in leukemia cell types	*gag (+env)*		U3 (enhancer)	



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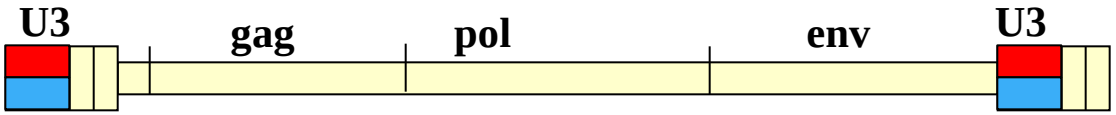
MLV strain	Age of inoculation (Balb/c, CBA, Swiss, etc.)	
	Newborn mice	
	Leukemia cell type	Latency
Friend-MLV	Erythroid (100%)	2-3 mos.
Moloney-MLV	Thymoma (100%)	3 mos.
Main sequences involved in leukemia cell types	U3 (enhancer)	



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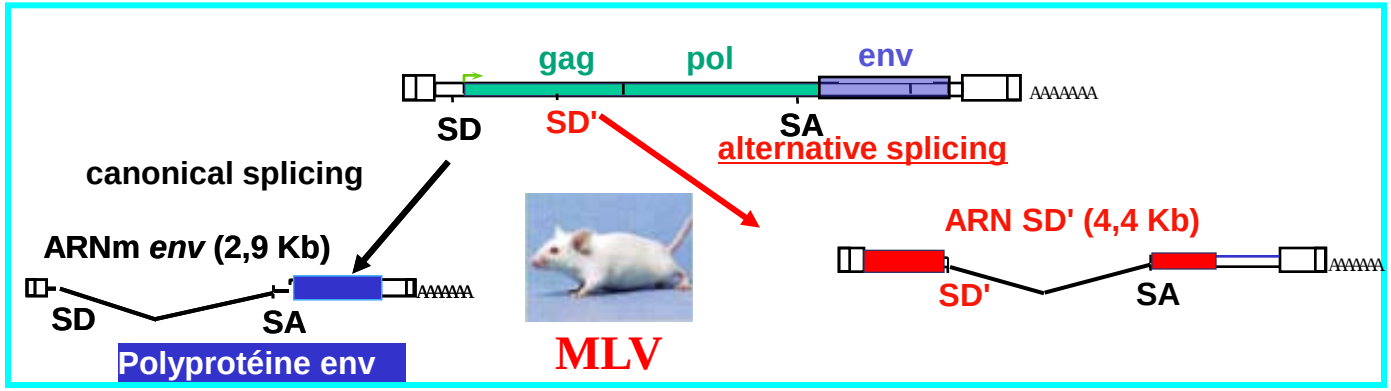
A cryptic splic site in MLV *gag* influences the leukemogenic cell type

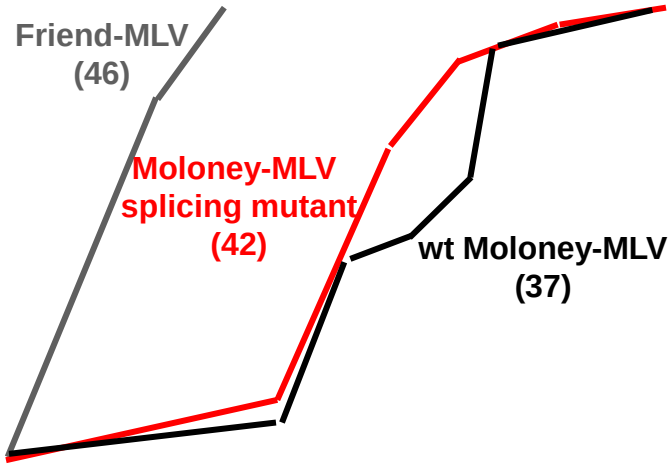
a cryptic splice site in MLV

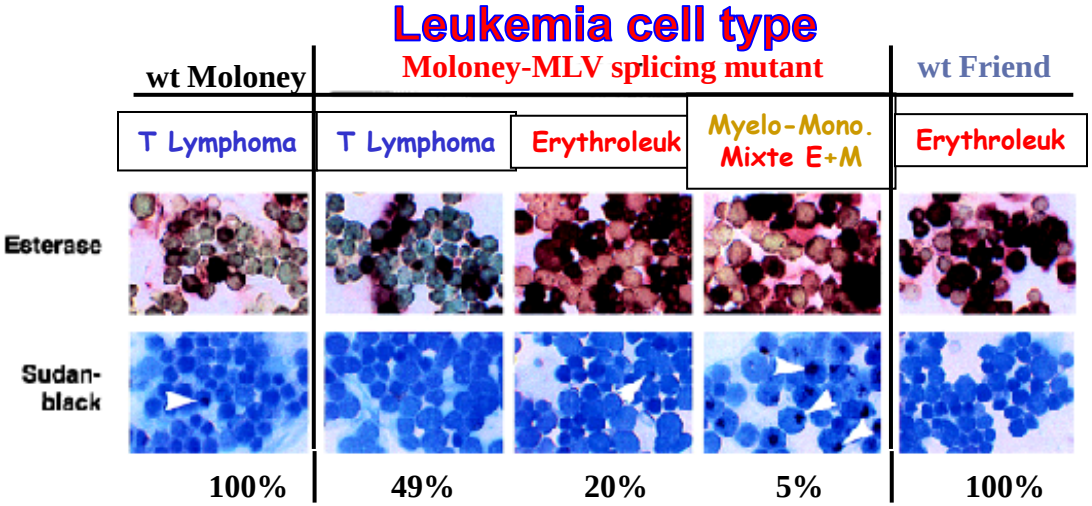
The SD' cryptic splice site is present in all mammalian oncogenic retroviruses



FeLV **GaLV**

Elimination of SD' maintains virulence but broadens the Moloney-MLV leukemia spectrum





Audit et al., J Virol. 1999

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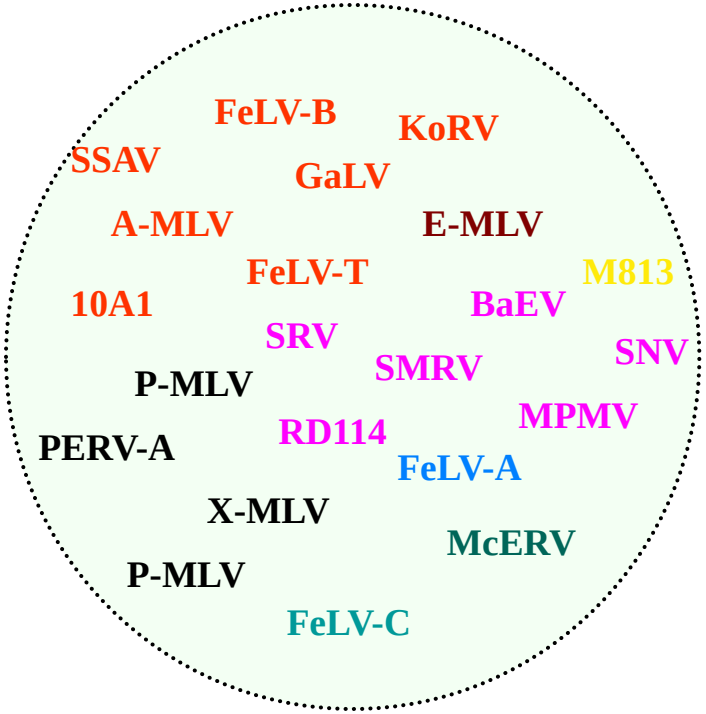
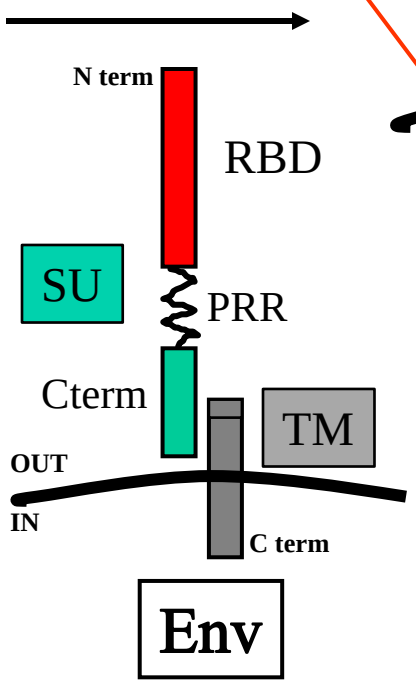
All known receptors of gammaretro Envs are nutrient and metabolite transporters

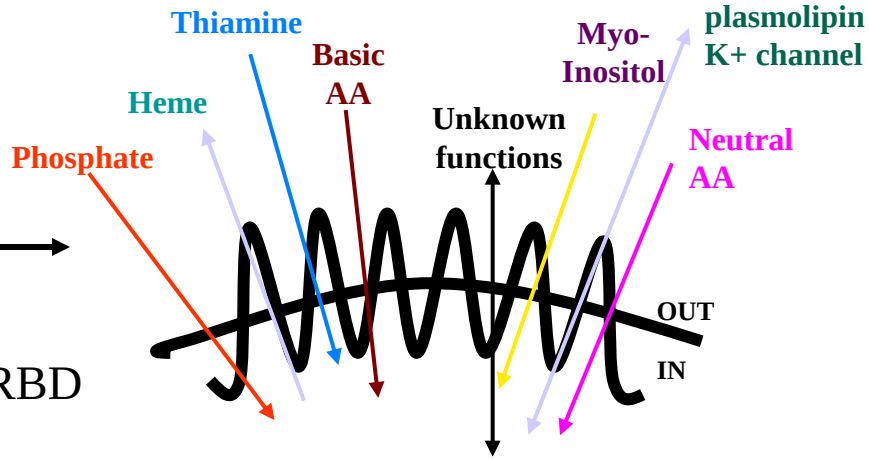
Gammaretroviruses



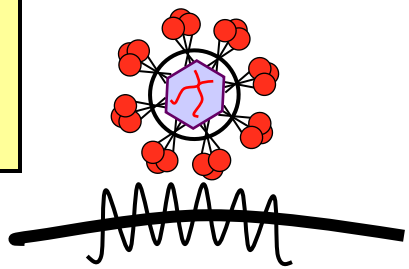
RBD = Receptor-Binding Domain



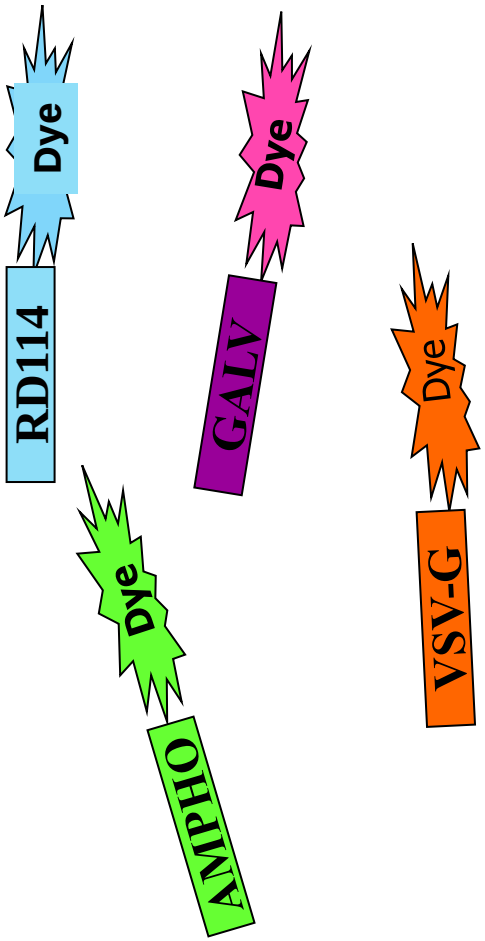
Receptors

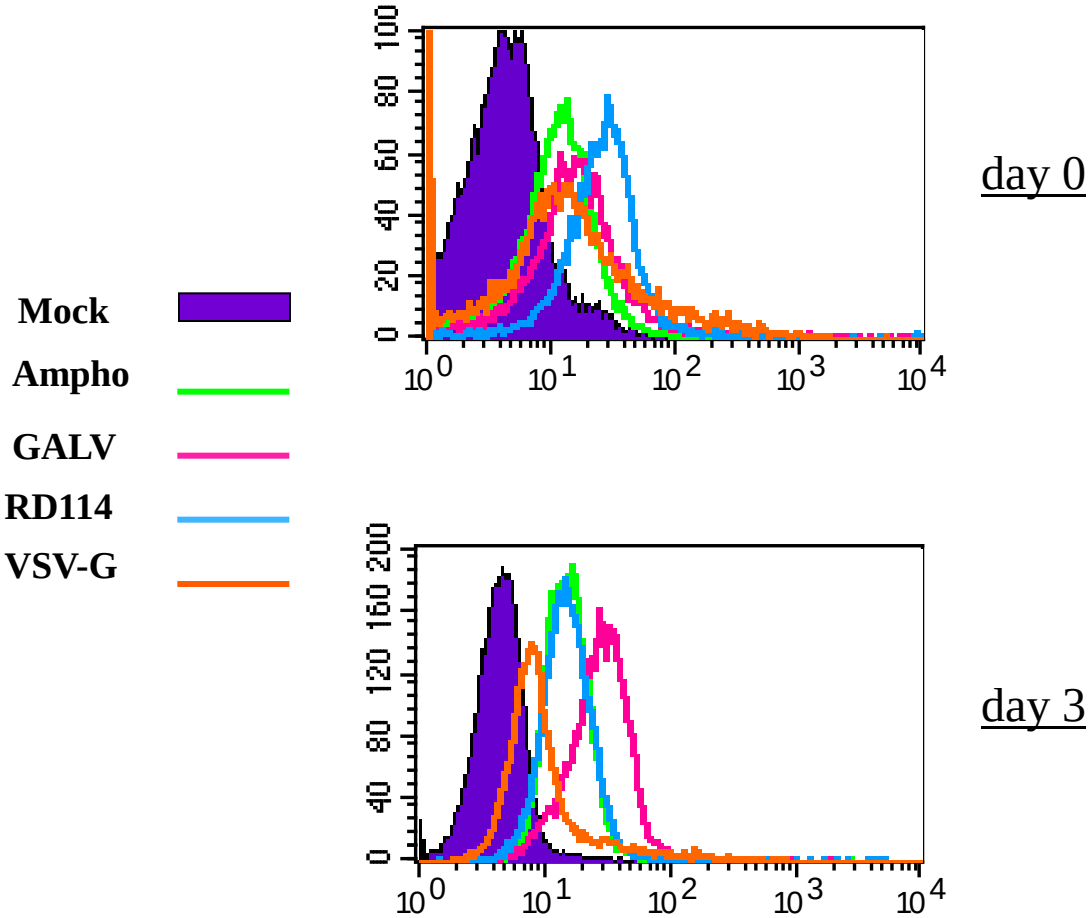


Variation of Env receptor expression during ex vivo culture of CD34+ hematopoietic cells



Env RBD to monitor receptor expression





MLV-driven leukemogenesis : many ways to (multi-) hit

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