

Resistance Mutations in the Viral Protease Alter the In Vitro Resistance Profiles of Bevirimat

Axel Fun, Noortje van Maarseveen, Renée Maas, Pauline Schipper and Monique Nijhuis

Department of Virology, University Medical Center, Utrecht, The Netherlands



University Medical Center
Utrecht

EHDRW

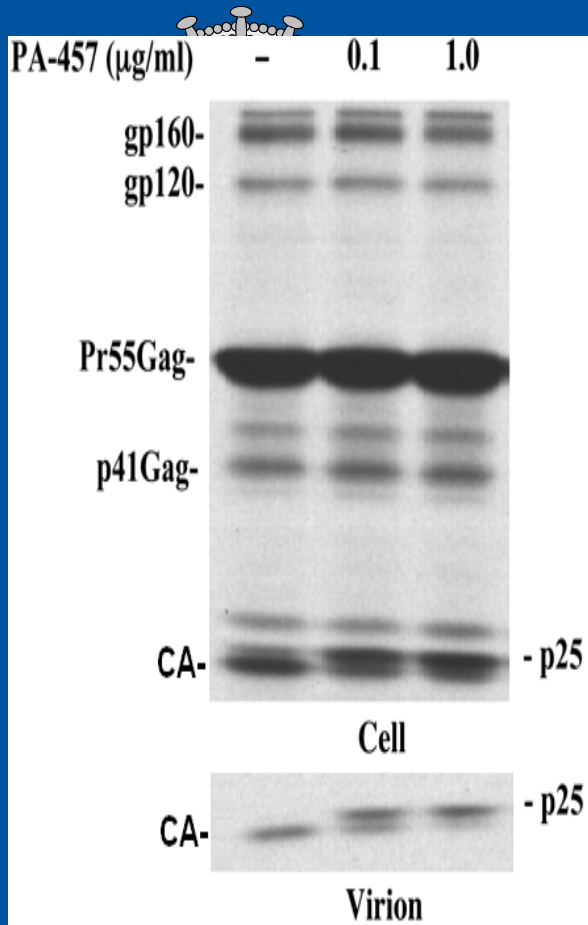
March 17, 2010

Sorrento, Italy

Particle maturation: an ordered stepwise process



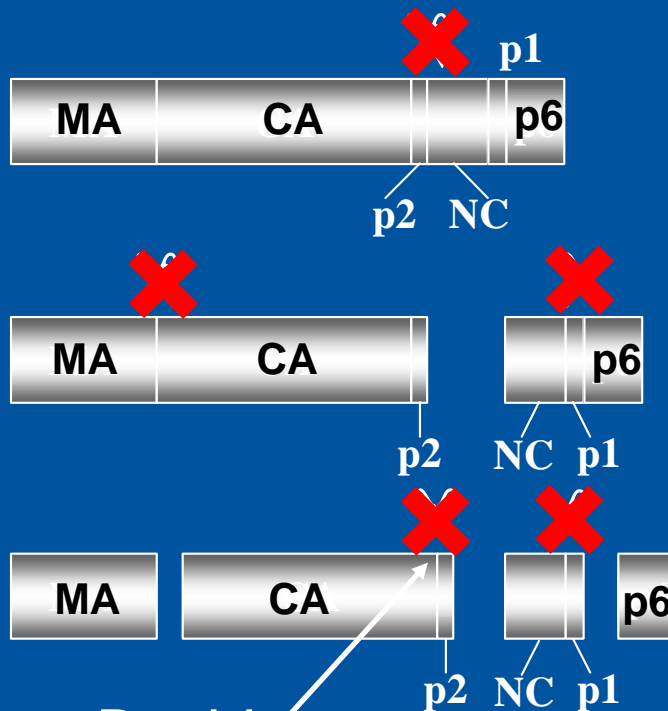
IMMATURE



Non-infectious

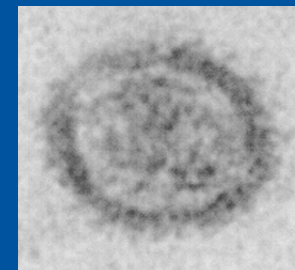
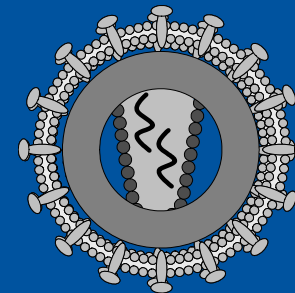
PI \longrightarrow ~~X~~ = viral protease

protease mediated cleavage of Gag precursor protein



Bevirimat

IMMATURE

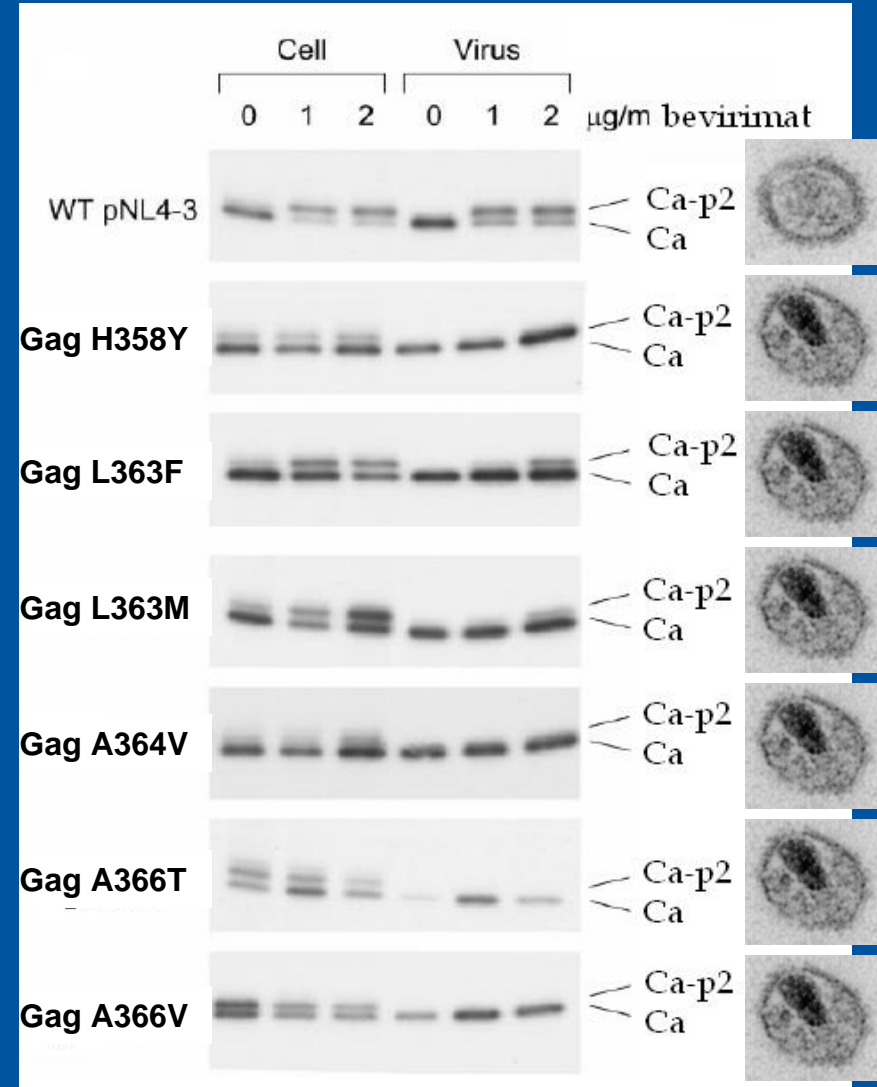
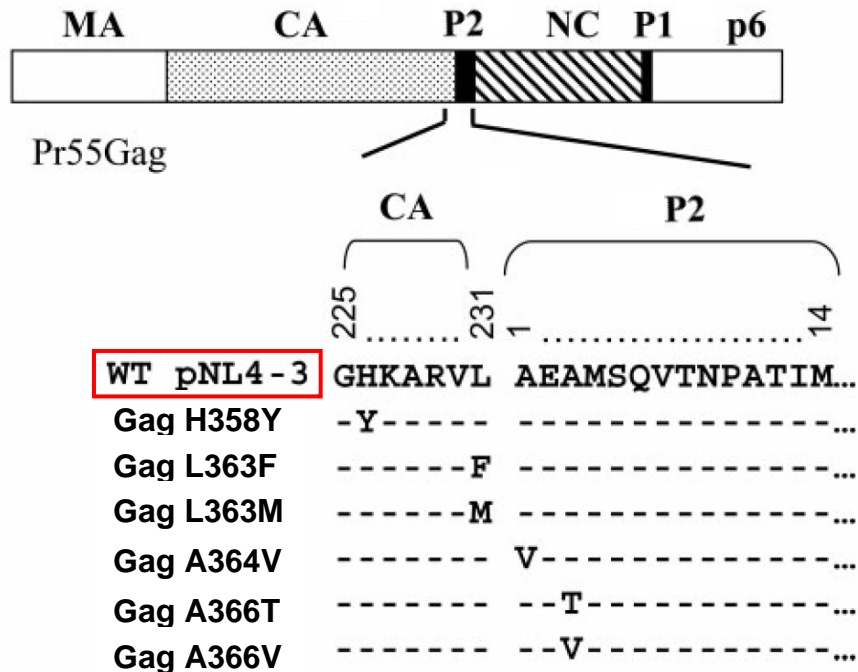


Non-infectious

In Vitro resistance against bevirimat



In Vitro selections with wild-type virus



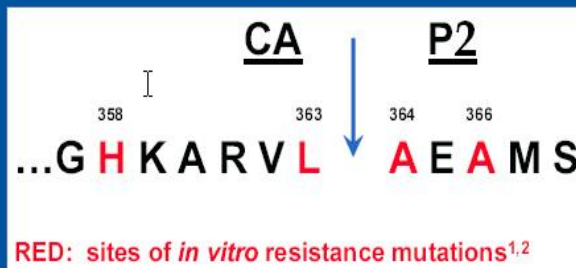
Adamson, et al., (2006) J. of Virol., 10957-10971

Results of phase IIb clinical trial:



Determinants of bevirimat response

- Approximately half of the patient population showed adequate response



		InPheno ¹	Virco ²
	CA-p2 Sequence	Fold Change in IC ₅₀	Fold Change in IC ₅₀
	...GHKARVL-AEAMSQVTNSATIM	1	1
129	...GHKARVL-AEAMSQVTS SATMM	0.57	4.8
180	...GHKARVL-AEAMSQVTNPPTIM	0.84	2.2
6	...GHKARVL-AEAMSQVTNSATIM	0.87	3.1
126	...GHKARVL-AEAMSQVTGSAAVM	1.02	1.3
1	...GHKARVL-AEAMSQVTNSATVM	1.14	2.4
182	...GHKARVL-AEAMSQVTNPATIM	1.16	1.7
14	...GHKARVL-AEAMSQVTPSATVM	1.51	58
16	...GHKARVL-AEAMSQVTNPSNIM	1.69	31.8
3	...SHKARIL-AEAMSQVTGPANIM	1.79	47.6
127	...GHKARVL-AEAMSQMTNSATAM		
127 chimera	...GHKARVL-AEAMSQMTNSATAM		
4	...GHKARVL-AEAMSQMTNPATIM		
24	...SHKARVL-AEAMSQV-NPTNIM		
10	...NHKARIL-AEAMCHVTNSATVM		
12	...GHKARVL-AEAMSQMTNSATTM		
8	...SHKARVL-AEAMCQA-NSTTVM		
125	...GHKARVL-AEAMSQATASNVIM		
15	...GHKARVL-AEAMSQA-NSSSIM		
125 chimera	...GHKARVL-AEAMSQATASNVIM		
30	...GHKARVL-AEAMSQATNSAAIM		

prevalence of BVM polymorphisms in clinical isolates



	Subtype B isolates	non-B isolates
	Therapy naïve	Therapy naïve
	n = 270	n = 167
Any mutation 369-371	112 (41.5%)	
Any bevirimat resistance associated mutation	85 (31.5%)	104 (62.3%)

(Verheyen, *et al.*, AIDS, 13 March 2010, V24, I5, p 669–673)

Interaction between PI resistance mutations and bevirimat



University Medical Center
Utrecht

Is resistance development to bevirimat affected by the presence of PI resistance mutations?

- Rate of resistance development

Affected by replication?

- Mutational pattern

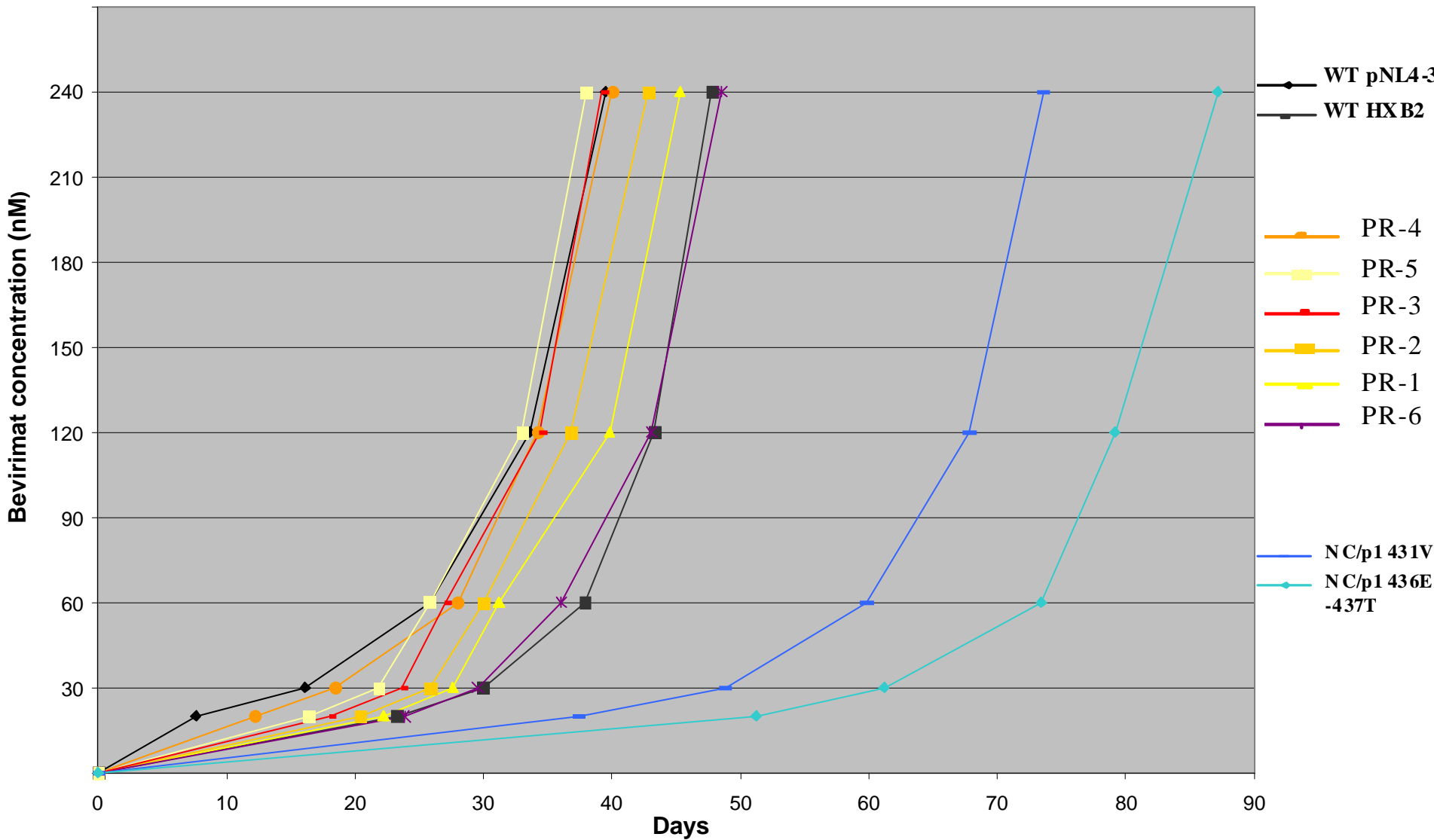
Interaction between PI resistance mutations and bevirimat



In Vitro selections

HIV-1 Variant	PI resistance mutation			RC (relative to WT virus)
	Gag		Protease	
	CA/p2	NC/p1		
Wild-type	Hxb2			+++
Wild-type	NL4-3			+++
PR1*	-	-	36I - 54V - 71V - 82T	+++
PR2*	-	431V	46I - 54V - 82A - 90M	++
PR3 [#]	-	431V	- 54V - 82A - 90M	+/-
PR4 [#]	-	431V	46I - 82A - 90M	+
PR5 [#]	-	431V	46I - 54V - 90M	+
PR6 [#]	-	431V	46I - 54V - 82A -	++
NC/p1	-	431V	-	+++
NC/p1	-	436E - 437T	-	+/-

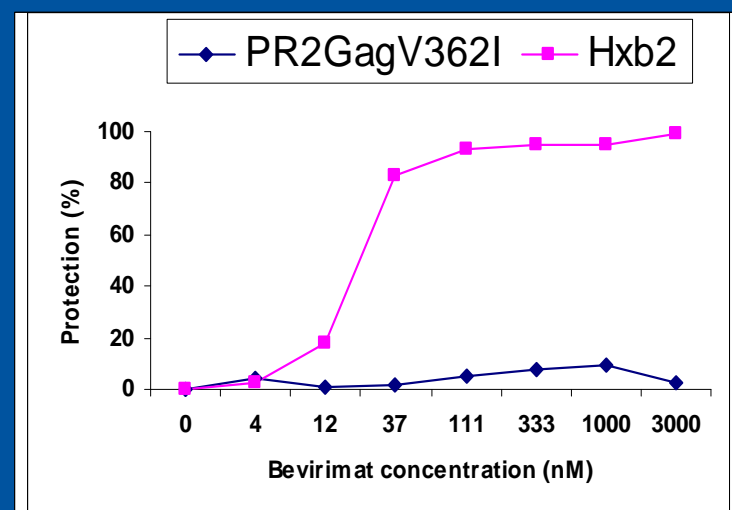
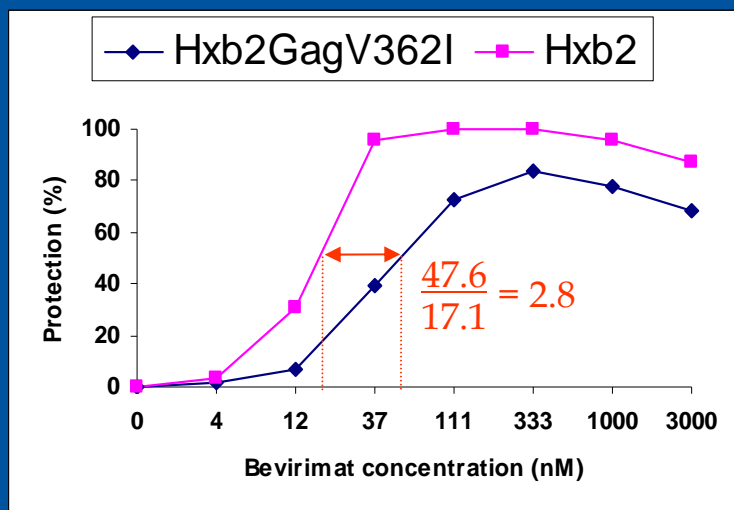
Development of bevirimat resistance



Impact of individual mutations on bevirimat resistance and viral replication



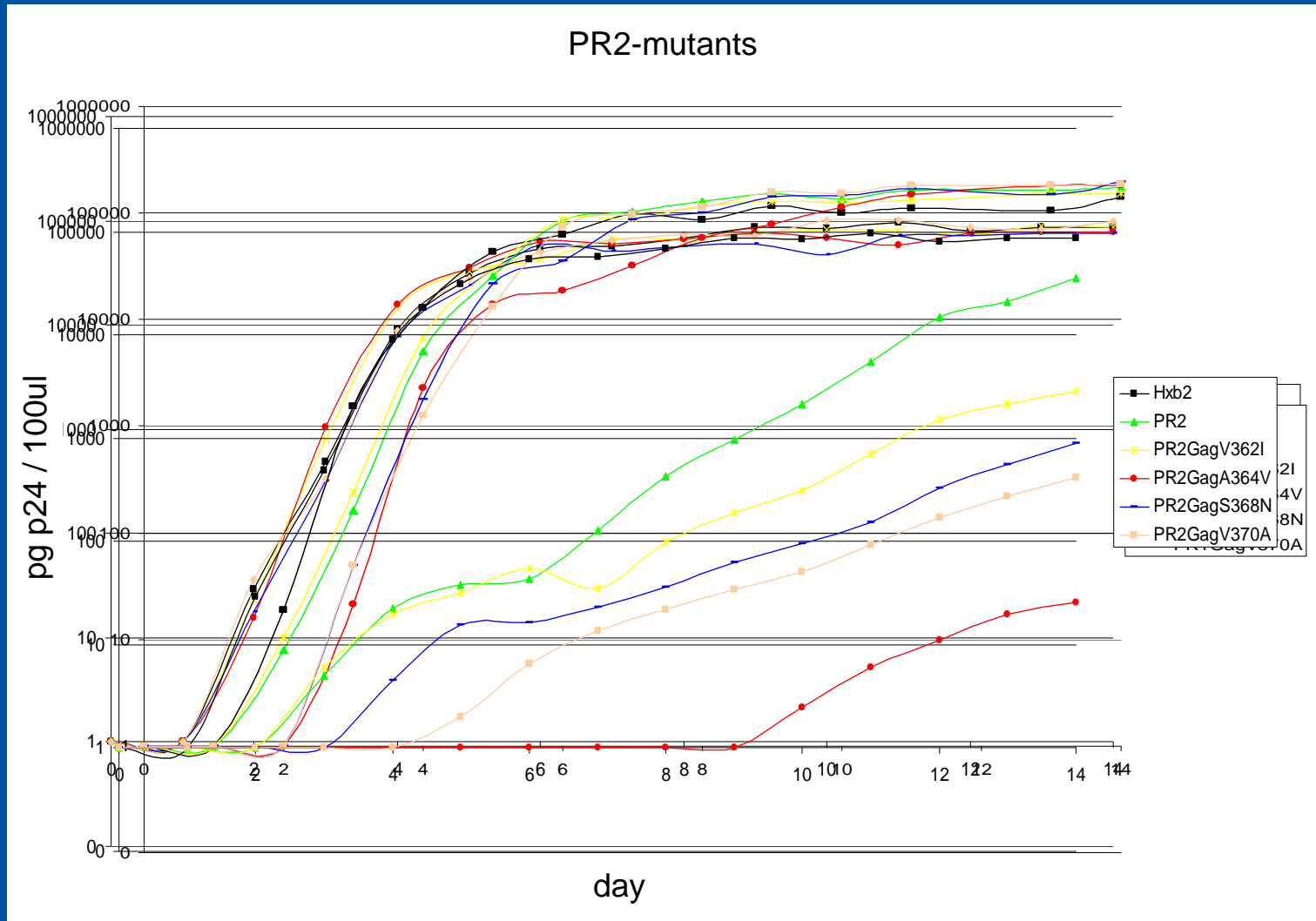
Virus	Fold resistance induced by mutation			
	V362I	A364V	S368N	V370A
Hxb2	2.8	>>150	6.6	>150



Bevirimat resistance mutations



Impact on viral replication



Conclusions



University Medical Center
Utrecht

In vivo data:

- PI resistant isolates (bevrimat naïve): significant accumulation of bevrimat resistance mutation (QVT-motif)

In vitro data

- Identified 2 new BVM resistance mutations (Gag V362I and S368N)
- Resistance mutations in the viral protease shift the bevrimat resistance pathway towards mutations in the QVT-motif
- Individual BVM resistance mutations in Gag have a different impact on replication and resistance depending on protease background

Acknowledgments



University Medical Center
Utrecht

Dept. of Medical Microbiology
UMC, Utrecht

Renée Maas
Noortje van Maarseveen
Diede Brunen
Annemarie Wensing
Monique Nijhuis

University of Cologne, Germany

Jens Verheyen
Elena Knops
Herbert Pfister
Rolf Kaiser

Aids Reference Laboratory, Ghent
University Hospital, Ghent, Belgium

Chris Verhofstede
Linus Vandekerckhove
Kenny Dauwe

