The Influence of Host HLA on Antiretroviral Drug Resistance Mutation in HIV-1

M John, C Moore, I James and S Mallal
Centre for Clinical Immunology and Biomedical Statistics (CCIBS), Royal Perth Hospital and Murdoch University, Perth, Australia

Western Australian HIV Cohort Study

BACKGROUND

Antiretroviral (ARV) drugs select characteristic 'drug escape' mutations in HIV sequence. Host HLA-restricted anti-HIV CTL responses also select escape mutations in HIV-1.

We hypothesised that;
-Selection effects (current and primordial) of drugs *and* CTL responses are evident at a population level.

-Drug resistance mutations should be drug-specific and CTL escape mutations should be specific for HLA class I alleles.

-Drug pressure and CTL pressure may compete, leading to synergistic or antagonistic interactions.

-Such interactions may help explain variable rates of emergence of drug resistance between individuals, discordance of drug resistance patterns *in-vivo* and *in-vitro* and genotype/phenotype discordance.

METHODS

HIV-1 RT and protease (proviral DNA) sequencing in 492 subjects of the WA HIV Cohort, performed over 2303 person-years of observation, were analysed.

Polymorphism(s) (versus consensus) of each single AA residue across the pooled viral sequences of the cohort were determined.

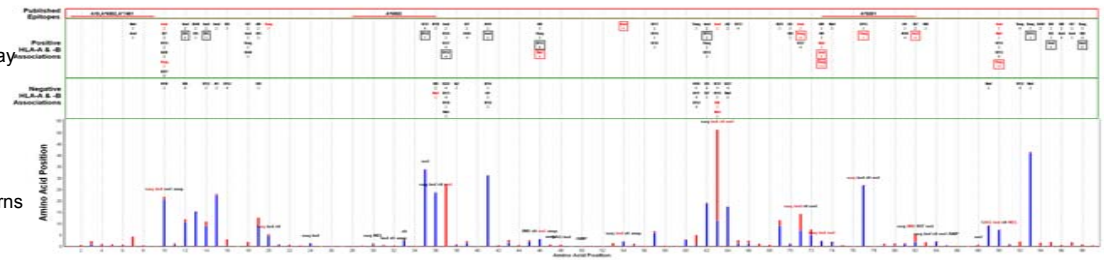
We then carried out a multivariate analyses (logistic regression models using Epipop™ method) for each residue (eg 184 of RT) where;
outcome= specified polymorphism (eg M184V, M184x)
-covariates=ARV drugs and HLA-A &-B alleles present in the population.

This process was repeated for all residues making up the full RT/protease proteins, giving a residue-specific view of the independent & interactive selection effects *in-vivo* of host CTL **and** ARV drugs on HIV-1, at a population level.

RESULTS

Map of HIV-1 RT; HLA & drug associated polymorphisms

Map of HIV-1 protease; HLA & drug associated polymorphisms



HLA allele-specific polymorphisms have characteristic AA substitution(s)

HLA allele	Site(s) of allele associated polymorphism in HIV-1 RT	CTL epitope (if known) containing/flanking polymorphism	Predominant amino acid substitution(s) (%)
A11	166	166-166 LAI	K166R (92%)
A28	32		K32R (63%)
B5	135	128-135 IIIB	I135T/V (83%) reduced HLA binding in-vitro shown
B7	158	156-165	A158S (70%)
	165		T165I (57%)
	169		E169D (63%)
B12	211	203-212 (HLA-B44)	R211K (74%)
B15	207		Q207E (58%)
B17	214		F214L (65%)
B18	66		S68G (88%)
	135		I135I (67%)
	142		I142T (63%)
B35	121	118-127	D121Y (58%)
	177	175-185	D177E (80%)
B37	200		T200I (63%)
B40	197	192-201 (HLA-B60)	Q197K (58%)
	207	207-216 (HLA-B60)	Q207E (54%)

HLA allele	Site(s) of allele-associated polymorphism in HIV-1 protease	Predominant amino acid substitution(s) (%)
B7	10	L10I (73%)
B12	35	E12D (94%)
B13	62	I62V (82%)
	65	E65D (60%)
	90	L90M (67%)
	93	I93L (93%)
B37	35	E35D (100%)
	37	N37D (57%)
B40	13	I13V (83%)

Selection of drug resistance mutations is evident at a population level

Amino acid substitutions examined in HIV-1 RT	Published primary drug association(s)	Drug association(s) detected at a population level in study cohort	OR	P-value
M41L	thymidine NRTI	ZDV	3	<0.001
D67N	ZDV	ZDV	10	<0.001
K70R	thymidine NRTI	ZDV	2	<0.001
L74V	ddI ABC	ddI	8	<0.001
K103N	NNRTI	nevirapine	6	<0.001
		efavirenz	6	<0.001
Y181C/I	nevirapine delavirdine	nevirapine	9	<0.001
M184V	3TC ddC ABC	3TC ddC	19 3	<0.001 0.004
G190A/S	nevirapine	nevirapine	11	<0.001
L210W	ZDV	ZDV	2	0.016
T215 Y	thymidine NRTI	ZDV	4	<0.001
K219Q/E	ZDV	ZDV	4	<0.001

Amino acid substitutions examined in HIV-1 protease	Published primary drug association(s)	Drug association(s) detected at a population level in study cohort	OR	P-value
L10I/R	secondary broad PI	indinavir	2	0.005
		saquinavir	3	<0.001
D30N	nelfinavir	ND		
M46I/L	primary indinavir	indinavir	3	0.008
G48V	primary saquinavir	ND		
I54V/L	indinavir	indinavir	5	<0.001
A71V/T	secondary broad PI	indinavir	2	0.017
		saquinavir	3	<0.001
73	secondary broad PI	indinavir	4	0.002
		saquinavir	10	<0.001
V77I	secondary broad PI	indinavir	2	0.026
V82A/T/F	indinavir	indinavir	3	0.01
	ritonavir	ritonavir	2	0.03
I84V	indinavir	indinavir	6	<0.001
N88S	nelfinavir	nelfinavir	11	<0.001
L90M	saquinavir nelfinavir	saquinavir	2	0.012
		nelfinavir	9	<0.001

Interactions between HLA alleles and ARV drugs

Particular HLA alleles independently increased the odds of;
(RT) M41L, K70R, Y181C/I,G190A/S, T210W, T215Y/F
(protease) L10I/R, M46I/L, A71V/T, 73,V77I,V82A/T/F,I84V, N88S, L90M

e.g. Of all ZDV-treated individuals (n=225),
Prevalence of HLA-A28 in-total cohort M41L + 8% M41- 16% 5% RR (of M41L on ZDV if HLA-28) 3.9, p=0.005 (Fisher's exact test).

Other +ve interactions; RT

	total cohort	K70R+	K70R-
A10	2%	17%	8%
		Y181C/I+	Y181C/I-
B8	23%	47%	20%
		G190A/S+	G190A/S-
B8	23%	64%	20%

protease

	total cohort	I84V+	I84V-
A9	22%	50%	15%
		N88S+	N88S-
B15	18%	64%	17%

HLA alleles reduced the odds of;RT- K103N, M184V and in protease- L10I/R/VI

CONCLUSIONS

These findings support a highly dynamic, host-specific model of HIV-1 adaptation *in-vivo*, in which host CTL responses & ARV therapy act as concurrent, competing or parallel interacting evolutionary forces at the level of single viral residues.

The selection of characteristic drug resistance mutations (& expected associations with their causative drugs) are evident at a population level.

The selection of characteristic CTL escape mutations is also evident at a population level, as HLA allele-specific polymorphisms. These are present in drug treated individuals.

For 15 known drug resistance mutations in RT & protease, HLA alleles influence risk of having the mutation following drug therapy.

Better understanding of CTL-driven effects on HIV evolution prove useful for individualisation of ARV therapy.

ABSTRACT

Background:Antiretroviral treatment (ART) induced mutations in HIV-1 reverse transcriptase (RT) and protease that allow viral escape from drug suppression are well characterised. Similarly, host CTL responses select viral variants that can escape CTL responses. We have shown in a large population study, that CTL escape mutations are evident as HLA allele-specific mutations. We examined the hypothesis that selection pressures of both host HLA-restricted CTL responses and ART interact at specific sites, causing variable susceptibility to drug resistance mutation between HLA diverse individuals. **Methods**:492 subjects with antiretroviral drug exposure, HLA-A and -B typing, pre- and post-ART RT(20-228) and protease(1-99) sequences over 2303 patient-years were studied. Multivariate logistic regression models were used to analyse associations between the development of ART resistance mutations (as the outcomes) and ART (as covariates). We then analysed the effects of HLA alleles in these models. **Results**:Associations between ZDV, 3TC, ddI, ddC, ABC, NVP, EFV, IDV, RTV, SQV, NFV and one or more of their corresponding, known primary and secondary drug resistance mutations were demonstrable using this population-based approach [eg.M184V in RT, was associated with use of 3TC(OR=69,p<0.001) and L90M in protease was associated with use of NFV (OR=3.1,p=0.003)]. At other sites (46 RT and 12 protease residues) drug exposures decreased risk of mutation. In models incorporating HLA alleles, there were 57 residues in RT and 33 in protease at which mutation was associated with specific HLA-A and –B alleles (p<0.05). Like drug resistance mutations, HLA-specific mutations (putative CTL escape mutations) were site-specific, associated with stereotypic amino acid substitutions and secondary mutations. At 8 RT and 7 protease drug mutation sites, we detected HLA associations independently increasing or decreasing the risk of mutation [eg,T215Y in RT was associated with ZDV(OR=3.7,p<0.001) and HLA-B7(OR=2.3,p=0.007), but was less likely in those with HLA-B13(OR=0.2,p=0.03)]. In protease, V82A/T/F was associated with IDV(OR=4.3, p<0.001) and HLA-A2 (OR=5.4, p=0.002). **Conclusions**:Knowledge of characteristic HLA-specific effects on viral sequence, including on drug resistance mutations may help explain variable susceptibility to drug resistance and be useful for individualisation of HIV therapy.

CONTACT-Dr Simon Mallal, Director
Centre for Clinical Immunology and Biomedical Statistics (CCIBS)
Level 2 North Block, Royal Perth Hospital
Wellington Street, WA, 6000, AUSTRALIA
mallal@prodigal.murdoch.edu.au
Ph:(61) 8 9224 2899, Fax:(61) 8 9224 2920
Website-http://www.maths.murdoch.edu.au/ccibs/

Western Australian HIV Cohort Study