A uniquely high level of recombination at the HLA-B locus

(chimpanzee/bonobo/major histocompatibility complex class I/evolution)

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ABSTRACT Major histocompatibility complex (MHC) loci are some of the most polymorphic genes in the animal kingdom. Recently, it has been suggested that although most of the human MHC loci are relatively stable, the HLA-B locus can undergo rapid changes, especially in isolated populations. To investigate the mechanisms of HLA-B evolution we have compared the sequences of 19 HLA-B homologues from chimpanzees and bonobos to 65 HLA-B sequences. Analysis of the chimpanzee and bonobo HLA-B homologues revealed that despite obvious similarities between chimpanzee and human alleles in exon 2, there was little conservation of exon 3 between humans and the two chimpanzee species. This finding suggests that, unlike all other HLA loci, recombination has characterized the HLA-B locus and its homologues for over 5 million years.

The products of the polymorphic (1) classical human major histocompatibility complex (MHC) class I loci (HLA-A, -B, -C) are molecules that bind peptides (2-4) and present them to CD8⁺ T cells. The T-cell receptor of the CD8⁺ T cell binds to the peptide/MHC complex, triggering destruction of the target cell. β_2 -Microglobulin-deficient mice that do not express MHC class I molecules show delayed clearance of viruses and are susceptible to a variety of other pathogens (5-10). Selection pressure appears to have been responsible for the maintenance of diversity at the amino acid residues that line the Ag-recognition site of MHC class I molecules (11). This diversity is thought to increase the number of possible different viral or tumor antigens that can be bound and presented to cytotoxic T lymphocytes. Thus, the products of these polymorphic classical MHC class I loci are critical to the surveillance function of an intact immune system.

Despite the enormous polymorphism of the MHC loci, allelic lineages of some of these MHC loci have been maintained over millions of years (12, 13). Comparison of HLA-A alleles and alleles of the orthologous locus of chimpanzees have provided evidence that the origin of such lineages may predate speciation events (14-16). All known chimpanzee A locus alleles belong to one of the six families of alleles found at the HLA-A locus, indicating that this allelic lineage originated prior to the divergence of humans and chimpanzees (5-7 million years ago). By contrast, the relationship between alleles at the highly polymorphic HLA-B locus and its chimpanzee homologue have not been easy to determine on the basis of the limited data available to date (17). To understand how evolution of this locus has occurred, we cloned and sequenced 18 HLA-B homologues from nine chimpanzees

and two bonobos and compared these alleles to their human counterparts (Table 1).^{††}

MATERIALS AND METHODS

PCR and Cloning of Chimpanzee and Bonobo HLA-B Homologues. PCR from cDNA or DNA was carried out as described (18, 19). Briefly, RNA was extracted from $2-5 \times$ 10⁶ lymphocytes using oligo(dT) bound to magnetic beads and DNA was extracted using standard techniques (20). For full-length amplification from cDNA, the LPHIII and MM2 primers were used (21). For partial-length amplification from RNA and DNA, GCBH3 [5'-GCAAGCTTGACGACAC(G) C)C(A/T)GTTCGTGA-3'] and NuA2ERI [5'-GCGAATTC-CAGC(G/T)T(G/C)TCCTTCCCGTTCTC-3'] were used. PCR products amplified from cDNA or genomic DNA were ligated into pSP65 and at least three full-length copies of each insert were sequenced to avoid PCR errors (22). At least four MHC class I-specific sequencing primers were used in each direction to generate double-stranded sequence and compressions were resolved using dITP. Sequences were analyzed using software from IBI.

Tree Construction. The trees were constructed by the neighbor-joining method (23) based on number of nucleotide substitutions per site (d) (24). Standard errors of branch lengths were estimated by Rzhetsky and Nei's method (25).

RESULTS

Similarity Between Chimpanzee, Bonobo, and Human $\alpha 1$ **Domains.** Comparison of 19 chimpanzee (*Pan troglodytes*) Patr-B and bonobo (Pan paniscus) Papa-B alleles to 65 HLA-B sequences demonstrated that there were striking similarities between the α domains of HLA-B sequences and their chimpanzee and bonobo orthologues (Fig. 1). For example, Papa-B*01 and Papa-B*04 differed from HLA- B^*0702 by only three and two residues in the $\alpha 1$ domain. respectively. Based on sequence similarity in the α 1 domain and serological cross-reactivity it was possible to identify only four major groups of chimpanzee and bonobo HLA-B homologues: HLA-B15, HLA-B48, HLA-B57/58, and HLA-B27/7.

The α 2 Domain Is Not Conserved Between Chimpanzees and **Bonobos and Humans.** The interspecies similarity of the $\alpha 1$ domains of the B locus alleles was not continued in the rest of the molecule (Figs. 1 and 2B). Unlike the tree of exon 2

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Abbreviations: MHC, major histocompatibility complex; HLA, human leukocyte antigen.

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^{††}The sequences reported in this paper have been deposited in the GenBank data base (accession nos. U05575-U05587).

Table 1. Or	rigin and serologic	al reactivity of	f chimpanzee and	bonobo HLA-B	homologues
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		Colony/ wild born	Clone		Serology		Similarity
Individual	Colony			Allele	Human	Chimp	to al*
Hugo	TNO	СВ	ChLA-B1	Patr-B*01			B17
			ChLA-B2	Patr-B*02	B53	118	B48
Tank	Yerkes	СВ	Ch-18	Patr-B*04	B40		B48
			Ch-39	Patr-B*03	B 17		B 17
Colin	Yerkes	СВ	Ch-11	Patr-B*05			B48
Kasey	Yerkes	СВ	Ch-7	Patr-B*06			B48
Harv	Yerkes	WB		Patr-B*06			B 17
				Patr-B*07			B 17
Teppie	Yerkes	WB		Patr-B*08			B15
••				Patr-B*09			B17
Renee	TNO	WB		Patr-B*10	B 17	125	B17
Victoria	TNO	WB		Patr-B*11	B07	106	B07
				Patr-B*12	B53	118	B07
Toetie	TNO	WB		Patr-B*10	B17	125	B17
Noel	TNO	WB		Patr-B*10		125	B17
Wodka	TNO	WB		Patr-B*14	B38/37	117	B07
				Patr-B*13		106	B07
1028	Duke			Patr-B*15	B40		B48
				Patr-B*09	B27		B17
Lorel	Yerkes	WB		Papa-B*01			B07
				Papa-B*03			B27
Bosondjo	Yerkes	WB		Papa-B*02			B27
				Papa-B*04			B07

*Similarity to $\alpha 1$ domain of B locus alleles indicated.

(Fig. 2A), the gene tree of exons 3-8 resulted in completely different clustering among human, chimpanzee, and bonobo B locus sequences (Fig. 2B). In this tree, the HLA-B homologues of chimpanzees and bonobos clustered separately from their human counterparts. By contrast, in similar trees of A locus alleles, the great ape HLA-A homologues clustered with HLA-A1, -A3, -A11 in trees of exon 2 and exons 3-8 (data not shown). Despite being very similar in the $\alpha 1$ domain, the HLA-B7-like bonobo alleles, Papa-B*01 and Papa-B*04, each differed from HLA-B*0702 in the α 2 domain by more than nine amino acid substitutions. Recombination of motifs between polymorphic B locus alleles and a number of Patr- and Papa-B-specific amino acid substitutions differentiated the chimpanzee and bonobo sequences from their human counterparts. Many of the unique substitutions in the chimpanzee and bonobo B alleles (i.e., substitutions not found in any HLA-B allele) were present in alleles of the HLA-A1/-A3/-A11 lineage (position 114, E and R; 151, H; 152, W and A).

The HLA-A and HLA-B Loci Evolve Differently. Analysis of human and chimpanzee MHC class I alleles reveals significant differences between mechanisms of evolution of the A and B loci. Pairwise comparisons between exons 2 and 3 of HLA-A, Patr-A, and Papa-A alleles demonstrate a correlation between the number of nucleotide substitutions in exons 2 and 3 (Fig. 3A)—that is, if A locus alleles differ by large numbers of substitutions in exon 2 they will also differ by large numbers of substitutions in exon 3, indicating that in most A locus alleles exons 2 and 3 share a common evolutionary history. No such correlation was seen in a similar analysis of HLA-B, Patr-B, and Papa-B alleles (Fig. 3B). This provides additional support for the hypothesis that intralocus recombination has occurred in exon 3 of B locus alleles serving to reassort polymorphic motifs between B locus alleles in exon 3.

DISCUSSION

Comparison of chimpanzee and human B locus genes suggests that B locus alleles may be an exception to the hypoth-

esis of trans-species evolution (12, 13). While homologues of the HLA-A allelic lineages are conserved in chimpanzee, bonobos, and humans (14-16), the HLA-B locus allelic lineages have been scrambled by recombination. Thus, the pattern of evolution observed at the B locus does not support the idea that all MHC loci evolve slowly (12, 13). Although there is evidence that diversity at the class II DRB loci in primates has been enhanced by recombination events, HLA- $DR\beta$ and Patr-DR\beta alleles of the same lineage cluster together in phylogenetic trees (20, 27, 28). Indeed, Patr-DR_{β4*0104} and HLA-DR_{β4*0104} differ at only three residues in the $\alpha 1$ domain (20), and the $\alpha 2$ domains of DR β alleles are conserved between chimpanzees and humans (29). This sharing of trans-species DRB sequence lineages is even observed between humans and rhesus macaques (30). Furthermore, a rhesus monkey Mamu-DRB1*03 allele can present a peptide from the 65-kDa heat shock protein of Mycobacterium tuberculosis/leprae to a human T-cell clone restricted by HLA-DR17 (31). Likewise, homologues of HLA-A (14-16), $-DR\alpha$, (29), $-DR\beta$, (20, 27, 28), $DQ\alpha$ (32, 33), and - $DQ\beta$, (34, 35) alleles in great apes evolve in a trans-specific fashion.

The presence of HLA-A1/-A3/-A11 specific substitutions in the chimpanzee and bonobo HLA-B homologues suggests a role for interlocus as well as intralocus recombination in the generation of diversity at the B locus. Thus, the great ape and human B7-related alleles probably derived from an HLA-B*07-like ancestral gene that gave rise to HLA-B*0702 in humans and Papa-B*01 and Papa-B*04 in bonobos. Subsequent to their divergence from the ancestral gene, both interand intralocus recombination in exon 3 encoding the $\alpha 2$ domain served to create new B locus alleles in humans, chimpanzees, and bonobos. Interestingly, it was sequence changes in exon 3 that were largely responsible for the creation of new HLA-B alleles found in South American tribes (21, 36).

The fact that an unusually high proportion of alleles at the B locus are products of recombination need not imply that this locus is a "hotspot" for recombination. If recombinant alleles have been selectively favored at the B locus to a

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Alpha 1	10	20	30	40	50	60	70	80	90 •
CONSENSUS	GSHSMRYFYTAMSRP	GRGEPRFIT	VGYVDDTOFV	RFDSDAASPRI	EPRAPWIEQEG	PEYWORETO	ISKTNTOT	RENLANLLA	YYNOSEA
HLA-B*5301		A		1		N	- F	IA	
HLA-B*5201 HLA-B*7901		Ä				N	-c	IARG	
HLA-B*3501 HLA-B*1301		A		7	λ	N	-F		
HLA-B*5101 HLA-B*4403		^	L	T				TA	
HLA-B*1302 HLA-B*3701	H-SV	s	L	T	L			DT	
HLA-B*4402 HLA-B*1801	н-sv	s	<u>L</u>	Ť		N			
HLA-B*3801 HLA-B*7801	sv	S		E		N	-C[RG	
HLA-B*4401 HLA-B*0801	p	s	L	E		N	-FI	SRG	
HLA-B*4501 HLA-B*4601		A	<u>-</u>	N	۱ A		KY-RQAI	-VSRG	
HLA-B*1501 Patr-B*08		»		<u>I</u> N	λ		A	SRG	
HLA-B*4101 HLA-B*4001	H		L L	T					
HLA-B*4002 HLA-B*5001	H-SV		L	T T				SRG	
HLA-B*4801	sv	s		E			A	SRG	
Patr-B*05	¥	8		¥1 ¥1			λ		
Patr-B*04			-	¥	I		&	-70	
HLA-B*5701 HLA-B*5801		^ ^		N	λ	GR	NM-λSλ NM-λSλ	IA IA	
Patr-B*01 Patr-B*09	<u>x</u> sv						- AGA	IA	
Patr-B*10 Patr-B*03	b A						й <u>ү</u> -хах:		
Patr-B*06 Patr-B*07	EF-2A					R	NY-ASA		Z D
HLA-B*2702	H-SV	e	L	E	#		-C-AKAD	IA	
Papa-B*03	<u>D</u> A	->			X	#	-C-AQAE	I A	
HLA-B*0702 Papa-B*01	SV SV	s			x	N	-Y-AQAI	SRG	
Papa-B*04 Patr-B*13	87	8			X	-	-Y-AQAI	-78-930	
Patr-B*11 Patr-B*12					X	X	-Y-XQA	-28-230	
Patr-B*14 HLA-B*4201	SV	s		IE		N	-Y-AQAI	SRG	
HLA-B*5401 HLA-B*5502				E		N	-Y-AQAI	SRG	
HLA-B*5602		A				N N	-Y-AQAD	SRG	
HLA-B*3903 HLA-B*1401	sv sv	s		B		N	-C	SRG	
HLA-B*1402 CONSENSUS	GSHSMRYFYTAMSRP	GRGEPRFIT	VGYVDDTQFV	RFDSDAASPRK	EPRAPWIEQEG	PEYWDRETQ	ISKINTOTY	RENLRNLLRY	YNOSEA
Alpha 2	100	110	120	130	140	150	160	170	180
Alpha 2 CONSENSUS	100 GSHTLQRMYGCDVGPE	110 GRLLRGHNC	120 YAYDGKDYIA	130	140 ADTAAQITQRKM	150 AAA · AO · BAARVABOL	160 A. AO RAYLEGLC	170 AO AO- TEWLRRYLEIN	180 GKETLQRA
Alpha 2 CONSENSUS HLA-B*4701 HLA-B*5301 HLA-B*5201	100 GSHTLQRMYGCDVGPC II	110 GRLLRGHNG	120 YAYDGKDYIA	130	140 • 40• ADTAAQITQRKM	150 AAA• AØ• BAARVABQL	160 A. AQ RAYLEGLCV	170 40 40. WEWLRRYLEN	180 GKETLQRA
Alpha 2 CONSENSUS HLA-B*4701 HLA-B*5301 HLA-B*5201 HLA-B*5201 HLA-B*7901	100 GSHTLQRMYGCDVGPE 	110 GRLLRGHNC	120 YAYDGKDYIA D	130	140 • 40• Adtanqitorku	150 AAA AO EAARVABQL 	160 A. AO RAYLEGLCY	170 40 40 - 7EWLRRYLEN	180 GKETLQRA
Alpha 2 CONSENSUS HLA-B*4701 HLA-B*5301 HLA-B*5201 HLA-B*5201 HLA-B*3501 HLA-B*3501 HLA-B*1301	100 GSHTLQRWYCCDVGPT 	110 XGRLLRGHIN YH D	120 	130 LNEDLSSWTA	140 • & & & & & & & & & & & & & & & & & & &	150 AAA • AØ • EAARVAEQI E	160 A. AO RAYLEGLCV E	170 40 40. EWLRRYLEN H	180 GKETLORA
Alpha 2 CONSENSUS HLA-B*4701 HLA-B*5201 HLA-B*5201 HLA-B*5201 HLA-B*3501 HLA-B*3501 HLA-B*1301 HLA-B*1301	100 GSHTLQRWYCCDVGPC 	110 GRLLRGHM YH D D D D D	120 	130 LNEDLSSWTA	140 . 40 . ADTANQITORKM	150 AAA · AO · EAARVAEQI 	160 A. AO RAYLEGLCV E	170 40 40. TEWLRRYLEN H	180 GRETLQRA
Alpha 2 CONSENSUS HLA-B*4701 HLA-B*5301 HLA-B*5201 HLA-B*7901 HLA-B*3501 HLA-B*1301 HLA-B*1301 HLA-B*1302 HLA-B*1302 HLA-B*3901	100 GSHTLORMOCDVOP 	110 XGRLLRGHNG YHD- D- D- D- 	120 	130	140 . 40. ADTAAQITQRKM	150 AAA · AO · EAARVAEQI E EE E E	160 A. AO RAYLEGICY E- E- E- E- E- E- E- E- E- E- E- E- E- E- E- E- E- E- E- E- E- E- E- E- E- E- E- E- E- E- E- E- E- E- E- 	170 A0 A0- FEWLRRYLENN H	180 GKETLORA
Alpha 2 CONSENSUS HLA-B*4701 HLA-B*5201 HLA-B*5201 HLA-B*5201 HLA-B*3501 HLA-B*3501 HLA-B*3100 HLA-B*1302 HLA-B*1302 HLA-B*1301 HLA-B*403 HLA-B*401 HLA-B*401 HLA-B*401 HLA-B*401	100 GSHTLORMYGCDVGPG 	110 CRLLRGHNC 	120 	130 LNEDLSSWTA	140 . <u>40</u> 	150 AAA • 40 • BAARVAEQI 	160 A. AQ RAYLEGICC E	170 Δ0 Δ0. TEWLERYLEN H	180 GRETLQRA
Alpha 2 CONSENSUS HLA-B*4701 HLA-B*5201 HLA-B*5201 HLA-B*5201 HLA-B*5201 HLA-B*1301 HLA-B*1301 HLA-B*1301 HLA-B*1301 HLA-B*1301 HLA-B*4001 HLA-B*1801 HLA-B*1801 HLA-B*1801 HLA-B*1801	100 GSHTLORMYCCDVGPC 	110 XGRLLRGHNG 	120 	130 INEDLSSWTA	140 . <u>40</u> . ADTAAQITORKM	150 <u>AAA. 40</u> . ERARVAEQI 	160 A. 40 RAYLEGLCY E	170 40 20 EWLRRYLEN H	180 GKETLQRA
Alpha 2 CONSENSUS HLA.B * 4701 HLA.B * 5301 HLA.B * 5201 HLA.B * 5201 HLA.B * 5101 HLA.B * 1301 HLA.B * 1301 HLA.B * 1302 HLA.B * 1302 HLA.B * 3011 HLA.B * 4001 HLA.B * 40	100 GSHTLORMYCCDVGPC 	110 GRILRCHWG 	120 	130	140 . 200. ADTAAQITQRKM	150 <u>AAA</u> · AO · EAARVAEQI 	160 A 40 RAYLEGLCV 	170 A0 A0- TBMLRRYLEN 	180 GKETLQRA
Alpha 2 CONSENSUS HLA-B*4701 HLA-B*5201 HLA-B*5201 HLA-B*5201 HLA-B*3501 HLA-B*3501 HLA-B*1301 HLA-B*1301 HLA-B*1301 HLA-B*1301 HLA-B*1301 HLA-B*1301 HLA-B*1801 HLA-B*1801 HLA-B*1801 HLA-B*1801 HLA-B*4501 HLA-B*4501	100 GSHTLORMYCDVGPU 	110 SGRLLRGHW 	120 	130 INEDLISSWTA	140 . 407 ADTRAQITORKM	150 AAA - A0 - EAARVAEQI 	160 A. 40 RAYLEGIC 	170 40 40 . ESVLRYLEN 	180 GRETLQRA
Alpha 2 CONSENSUS HLA-B*4701 HLA-B*5201 HLA-B*5201 HLA-B*5201 HLA-B*3501 HLA-B*3501 HLA-B*1301 HLA-B*1302 HLA-B*1302 HLA-B*1302 HLA-B*1302 HLA-B*1302 HLA-B*1302 HLA-B*1301 HLA-B*401 HLA-B*4501 HLA-B*4501 HLA-B*1501 Par-*000	100 GSHTLORMYCCDVGPC 	110 XGRLLRGHW 	120 	130	140 . 407 ADTRAQITORKM	150 <u>AAA</u> . <u>AO</u> . EAARVAEQI 	160 A . 40 RAYLEGICT 	170 40 40- ESVLRYLEN 	180 GRETLORA
Alpha 2 CONSENSUS HLA-B+4701 HLA-B-5201 HLA-B-5201 HLA-B-5201 HLA-B-7901 HLA-B-7901 HLA-B-1301 HLA-B-1301 HLA-B-1302 HLA-B-1302 HLA-B-14001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-4001 HLA-B-400	100 GSHTLORHYOCDVGPT 	110 SGRLLRGHUNG HH	120 	130 LINEDLSSWTA	140 . 407 ADTANQITORKM	150 <u>AAA</u> . 40. EAARVAEQI 	160 A . 40 RAYLEGIC 	170 40 40- ESVLRYLEN 	180 GRETLQRA
Alpha 2 CONSENSUS HLA-B+4701 HLA-B+201 HLA-B+201 HLA-B+201 HLA-B+201 HLA-B+301 HLA-B+301 HLA-B+301 HLA-B+301 HLA-B+301 HLA-B+301 HLA-B+401 HLA-B+401 HLA-B+1501 Patr=900 HLA-B+1001 HLA-B+1001 HLA-B+1001 HLA-B+1001 HLA-B+1001 HLA-B+2001 HLA-B+2001 HLA-B+2001 HLA-B+2001 HLA-B+2001 HLA-B+2001 HLA-B+2001	100 GSHTLORMYCCDVGPT IIFL W-TL IIL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-TL W-T W-TL W-T W-T	110 SGRLLRGHNG 	120 	130	140 . 60. ADTANQITORKM	150 <u>AAA</u> . 40. EAARVAEQI 	160 A. 40 RAYLBOIC 	170 40 40- HESVLRRYLEN 	180 GRETLQRA
Alpha 2 CONSENSUS HLA-B+4701 HLA-B-5201 HLA-B-5201 HLA-B-5201 HLA-B-5201 HLA-B-5201 HLA-B-501 HLA-B-501 HLA-B-101 HLA-B-101 HLA-B-4001 HLA-B-4001 HLA-B+4001 HLA-B+4001 HLA-B+4001 HLA-B+4001 HLA-B+4001 HLA-B+4001 HLA-B+4001 HLA-B-5001 HLA-B-4001 HLA-B-4001 HLA-B-5001 HLA-B-4001 HLA-B-5001 HLA-B-4001 HLA-B-5001	100 GSHTLQRMYGCDVGPT IIFL W-TL W-T II W-T W-T IIS W-T W-T W-T 	110 SGRLLRGHNG 	120 	130 LINEDLSSWTA	140 . 60. ADTAAQITQRKM	150 AAA . 40 . EAARVAEQL 	160 A. 40 RAYLBOICY 	170 40 40 - HEMLRRYLEN 	180 GRETLQRA
Alpha 2 CONSENSUS HLA-B*4701 HLA-B*5301 HLA-B*5201 HLA-B*5201 HLA-B*5301 HLA-B*5301 HLA-B*1301 HLA-B*1301 HLA-B*1302 HLA-B*3701 HLA-B*3701 HLA-B*4401 HLA-B*4601 HLA-B*4501 HLA-B*4501 HLA-B*4011 HLA-B*4011 HLA-B*4011 HLA-B*5011 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B	100 GSHTLORMYGCDVGPI 	110 SGRLLRGHNG 	120 	130 LINEDLSSWTA	140 . 60. ADTAAQITQRKM	150 AAA - AO - RAARVAEQI 	160 A. 40 RAYLBOLC 	170 <u>A0</u> <u>A0</u> . ESVLREYLEN 	180 GKETLQRA
Alpha 2 CONSENSUS HLA-B*4701 HLA-B*5301 HLA-B*5301 HLA-B*5301 HLA-B*1301 HLA-B*1301 HLA-B*1301 HLA-B*1301 HLA-B*3701 HLA-B*4401 HLA-B*4601 HLA-B*4601 HLA-B*4501 HLA-B*4501 HLA-B*401 HLA-B*401 HLA-B*401 Patr=B*25 Patr=B*25 Patr=B*25 Patr=B*25 Patr=B*26	100 GSHTLORMOCDVOP 	110 SGRLLRGHM 	120 	130 LINEDLSSWTA	140 	150 AAA - 40 - EARVAEQL 	160 A. 40 RAYLEGICY 	170 <u>A0</u> <u>A0</u> . ESVLRRYLEN 	180 GKETLQRA
Alpha 2 CONSENSUS HLA-B-4701 HLA-B-5301 HLA-B-5301 HLA-B-5301 HLA-B-5301 HLA-B-5301 HLA-B-1301 HLA-B-1301 HLA-B-4401 HLA-B-4401 HLA-B-4601 HLA-B-4501 HLA-B-1501 HLA-B-1501 HLA-B-1501 HLA-B-401 Patr-B-00 HLA-B-1501 HLA-B-4501 HLA-B-4501 HLA-B-4501 HLA-B-4501 HLA-B-4501 HLA-B-4501 HLA-B-501 HLA-B-5501 HLA-B-5501 HLA-B-5501	100 GSHTLORMOCDVOP 	110 SGRLLRGHM 	120 	130 LINEDLSSWTA	140 . 40. ADTAAQITQRM 	150 AAA - 40 - EARVAEQL 	160 A . 40 RAYLEGICY 	170 <u>40</u> <u>40</u> . ESVLRYLEN 	180 GKETLQRA GKETLQRA
Alpha 2 CONSENSUS HLA-B'4701 HLA-B'5201 HLA-B'5201 HLA-B'5201 HLA-B'501 HLA-B'1301 HLA-B'1301 HLA-B'1302 HLA-B'3701 HLA-B'4001 HLA-B'4001 HLA-B'4001 HLA-B'4001 HLA-B'5011 HLA-B'5011 HLA-B'401 Patr-B'002 HLA-B'401 Patr-B'002 HLA-B'401 Patr-B'501 Patr-B'501 Patr-B'501 Patr-B'501 Patr-B'501 Patr-B'501 Patr-B'501 Patr-B'00	100 GSHTLORMOCDVAP 	110 SGRLLRGHM 	120 	130 LINEDLSSWTA	140 . 40. ADTAAQITQRM 	150 AAA - 40 - EARVAEQL 	160 A . 40 RAYLEGICY 	170 <u>40</u> <u>40</u> . ESVLRYLEN 	180 GRETLQRA
Alpha 2 CONSENSUS HLA-B'4701 HLA-B'5001 HLA-B'5001 HLA-B'5001 HLA-B'5001 HLA-B'1300 HLA-B'1300 HLA-B'1300 HLA-B'1300 HLA-B'4001 HLA-B'4001 HLA-B'4001 HLA-B'4001 HLA-B'4001 HLA-B'5001 HLA-B'4001 HLA-B'5001 HLA-B'5001 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701 HLA-B'5701	100 GSHTLORMYCCDVCPC 	110 SGRLLRGHM 	120 	130 LINEDL.SSWTA	140 . 40. ADTAAQITQRM 	150 AAA - AO - EARVAEQL 	160 A . 40 RAYLBOLC 	170 <u>40</u> <u>40</u> . ESVLRYLEN 	180 GRETLQRA
Alpha 2 CONSENSUS HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*1301 HLA-B*1301 HLA-B*1301 HLA-B*1301 HLA-B*1301 HLA-B*1301 HLA-B*4001 HLA-B*4001 HLA-B*4001 HLA-B*5011 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA-B*501 HLA	100 GSHTLORMYCOUGPC 	110 CRLLRCHM 	120 	130 LINEDL.SSWTA	140 . 40. 	150 AAA . 40 . EARVAEQL 	160 A. 40 RAYLBOICS 	170 <u>40</u> <u>40</u> . ESVLRYLEN 	180 CRETLQRA CRETLQRA
Alpha 2 CONSENSUS HLL-B-5301 HLA-B-5201 HLA-B-5201 HLA-B-5201 HLA-B-5101 HLA-B-5101 HLA-B-1301 HLA-B-1301 HLA-B-1402 HLA-B-1401 HLA-B-4402 HLA-B-4402 HLA-B-4401 HLA-B-4401 HLA-B-4401 HLA-B-4601 HLA-B-5001 HLA-B-5001 HLA-B-5001 HLA-B-4002 HLA-B-4002 HLA-B-4002 HLA-B-4002 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 HLA-B-501 H	100 GSHTLORMYCCDVGPC 	110 SCRLLRCHM TH 	120 YAYDGKDYIA D	130 LINEDLOSWTA	140 . 40. ADTAAQITQRM 	150 AAA . 40 . EAARVAEQL 	160 A. 40 RAYLEGICY 	170 40 40. ESVLRYLEN 	180 CRETLQRA CRETLQRA
Alpha 2 CONSENSUS HLA = B * 5001 HLA = B * 1002 HLA = B * 1002 HLA = B * 1002 HLA = B * 4002 HLA = B * 4001 HLA = B * 5001 HLA = B *	100 CSHTLORMOCDVGPC 	110 SCRLLRCHM 	120 YAYDGKDYIA D	130 LINEDLOSWTA	140 . 40. ADTAAQITQRM 	150 AAA . 40 . EARVARDI EARVARDI E. E. D. D. D. D. D. D. D. D. D. D	160 A. 40 RAYLEGICY 	170 40 40. ESALARYLEN 	180 GRETLQRA
Alpha 2 CONSENSUS HLA B* 7001 HLA B* 5001 HLA B* 5001 HLA B* 5001 HLA B* 5001 HLA B* 5001 HLA B* 1001 HLA B* 1001 HLA B* 1001 HLA B* 1001 HLA B* 4001 HLA B* 5001 HLA B* 2002 Parr B* 000 Parr B* 000	100 CSHTLOPHYCCDVGPC 	110 CRELEROHM CRELEROHM D D D D D D D D D D D D D	120 YAYDGKDYIA D	130 LINEDLOSWTA	140 . 40. ADTAAQITQRM 	150 AAA . 40 . EARVARDI E. E. E. D. E. D. E. D. E. D. E. D. E. D. E. D. E. D. E. D. E. D. E. D. E. D. D. E. E. D. D. E. E. D. D. E. E. D. D. E. E. D. D. D. E. E. D. D. D. E. E. D. D. D. E. E. D. D. D. E. E. D. D. D. E. E. D. D. D. E. E. D. D. D. D. E. E. D. D. D. D. D. E. D. D. D. E. E. D. D. D. E. E. D. D. D. E. E. D. D. E. E. D. D. E. E. D. D. E. E. D. D. E. E. D. D. E. E. D. D. E. E. E. D. E. E. D. E. E. D. E. E. D. E. E. E. E. E. E. E. E. E. E	160 A. 40 RAYLBOICS 	170 40 40. ESVLRYLEN 	180 GRETLQRA
Alpha 2 CONSENSUS HLA BP *7011 HLA BF 5201 HLA BF 5201 HLA BF 5201 HLA BF 5201 HLA BF 5201 HLA BF 5101 HLA BF 1302 HLA BF 1302 HLA BF 1302 HLA BF 4401 HLA BF 4401 HLA BF 4401 HLA BF 4601 HLA BF 4601 HLA BF 4601 HLA BF 4601 HLA BF 4001 HLA BF 4002 HLA BF 4001 HLA BF 400	100 CSHTLOPHYCCDVGPC 	110 CRELLROHM CRELLROHM D D D D D D D D D D D D D	120 YAYDCKDYIA D	130 LINEDLOSWTA	140 . 40. ADTAAQITQRM 	150 AAA . AO . EAARVAEQL 	160 A. 40 RAYLBOICY 	170 40 40. ESVLRYLEN 	180 GKETLQRA
Alpha 2 CONSENSUS HLA = D = 7001 HLA = D = 5001 HLA = D = 100 HLA = D =	100 CSHTLORMYCCDVGPC 	110 CRILIROHM CRILIROHM D D D D D D D D D D D D D	120 YAYDCKDYIA D	130 LINEDLOSWTA 	140 . 40. ADTAAQITQRM 	150 AAA . AO . EAARVAEQL 	160 A. 40 RAYLEGICY 	170 40 40. TSMLRNYLEN 	180 CKETLQRA CKETLQRA
Alpha 2 CONSENSUS HLA B* 47011 HLA B* 53011 HLA B* 53011 HLA B* 52011 HLA B* 52011 HLA B* 51011 HLA B* 51011 HLA B* 13021 HLA B* 13021 HLA B* 44013 HLA B* 44011 HLA B* 45011 HLA B* 57011 HLA B* 55011 HLA B* 550	100 GSHTLORMYCCDVGPC 	110 CRILIRCHM CRILIRCHM 	120 YAYDCKDYIA D	130 LINEDLOSWTA 	140	150 AAA . AO . RAARVAEQL 	160 A. 40 RAYLBOIC 	170 40 40- ESVLRYLEN 	180 GKETLQRA
Alpha 2 CONSENSUS HLA BP 47011 HLA BP 5301 HLA BP 5201 HLA BP 5201 HLA BP 5201 HLA BP 5201 HLA BP 5101 HLA BP 1301 HLA BP 1301 HLA BP 1301 HLA BP 4401 HLA BP 4401 HLA BP 4401 HLA BP 4401 HLA BP 4401 HLA BP 4401 HLA BP 4501 HLA BP 5501 HLA BP 5501 HLA BP 5701 Patr BP	100 GSHTLORMYCCDVGPC 	110 SCRLLRCHM SCRLLRCHM 	120 YAYDCKDYIA D	130 LINEDLSSWTA 	140 	150 AAA . AO . RAARVAEQL 	160 A. 40 RAYLBOICY 	170 40 40. ESVLRYLEN 	180 CKETLQRA CKETLQRA
Alpha 2 CONSENSUS HLA BP 47011 HLA BP 5301 HLA BP 5201 HLA BP 5201 HLA BP 5201 HLA BP 5201 HLA BP 5101 HLA BP 1301 HLA BP 1301 HLA BP 4401 HLA BP 4501 HLA BP 4501 HLA BP 4501 HLA BP 4501 HLA BP 4501 HLA BP 4501 HLA BP 5701 HLA BP 5701 HLA BP 5501 HLA BP 5701 Patr BP 5701 HLA BP 5701 HLA BP 5501 HLA	100 GSHTLORMYCCDVGPC 	110 SCRLLRCHM SCRLLRCHM 	120 YAYDCKDYIA 9	130 LINEDLSSWTA	140 	150 AAA . 40 . RAARVAEQL 	160 A. 40 RAYLBOLC 	170 40 40. ESVLRYLEN 	180 CKETLQRA CKETLQRA
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FIG. 1. Predicted amino acid sequences of the chimpanzee and bonobo *HLA-B* homologues compared to *HLA-B* sequences. A consensus sequence of the αl and $\alpha 2$ domain is shown and the chimpanzee and bonobo *HLA-B* homologues (in boldface type) and *HLA-B* alleles are compared; identity is indicated with a dash. Any unique amino acid substitutions in the bonobo and chimpanzee *B* alleles not found in previously sequenced human *HLA-B* alleles are indicated by underlining them. Amino acids that point into the Ag-recognition site (\bullet), point both into the Ag-recognition site and up toward the T-cell receptor (\diamond), and point up toward the T-cell receptor (\diamond), and point up toward the B pocket, is also indicated (\Box).



FIG. 2. Phylogenetic trees for exon 2 (A) and exons 3-8 (B) of B locus alleles from human (HLA-), chimpanzee (Patr-), and bonobo (Papa-). Statistically significant internal branches are indicated as follows: *, P < 0.05; **, P < 0.01; ***, P < 0.001. Trees were rooted using HLA-A, Patr-A, and Papa-A sequences.

greater extent than they have at other MHC loci, recombinant alleles are expected to be observed more frequently at



FIG. 3. Number of nucleotide substitutions per site (24) in exons encoding the α^2 domain as a function of that of exons encoding the α 1 domain for all pairwise comparisons among human, chimpanzee, and bonobo A locus alleles (A) and B locus alleles (B); human sequences used are as in Fig. 2. The lines shown are linear regression lines: (A) y = 0.024 + 0.479x; (B) y = 0.057 + 0.050x. Pairwise comparisons of the number of nucleotide substitutions per site, d(24), in exons encoding the α^2 domain were plotted as a function of the number of nucleotide substitutions per site in exon 2 (encoding the α 1 domain). Because pairwise comparisons are not strictly independent, in order to have a conservative test of the null hypothesis that the correlation coefficient is equal to zero, we tested with n-2 df, where n is the number of sequences compared rather than the number of pairwise comparisons. In A, r = 0.612 (P < 0.001); in B, r = 0.094 [not significant (NS)]. Similar values were obtained when the number of synonymous substitutions per site (26) in exons encoding the $\alpha 2$ domain was correlated with that of exons encoding in the α 1 domain—namely, for the A locus r = 0.562 (P < 0.01), and for the B locus r = 0.056 (NS). Similar values were also obtained for the correlation of the number of nonsynonymous nucleotide substitutions per site in exon 2 with that in exon 3-namely, for the A locus $r = 0.551 \ (P < 0.01)$ and for the B locus $r = 0.075 \ (NS)$.

the B locus than at other loci even if the rate of recombination at the B locus is similar to that at other MHC loci. An analogous phenomenon has been observed in bacteria and other microorganisms (37–39), in which recombinants are much more likely to be observed at loci where diversity is selectively favored.

The conservation of the α l domain between chimpanzee, bonobo, and human *B* locus alleles implies that the peptide binding pockets encoded by this domain may be under strong structural and functional constraints. In many of the peptide binding motifs analyzed, it appears that the anchoring amino acid is bound by the second, or B, pocket (40-42). The amino acids lining this pocket are largely made up of residues

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encoded for by exon 2. Thus, the peptide binding motifs of the chimpanzee, bonobo, and human B locus alleles may be very similar, with likely conservation of the anchor residues between these two species. The difference in the $\alpha 2$ domains may, however, change some of the amino acids accommodated by pockets D-F but not change the anchor amino acid of the peptides bound by chimpanzee, bonobo, and human B locus alleles. Perhaps the advantage of being able to adapt rapidly to changing pathogens selects for the binding of slightly different peptides. An example of this may be the differential peptide binding abilities of HLA-B35 and HLA-B53, which differ by only five amino acids at the end of the α 1 domain. The anchor residue of proline is conserved between the peptides bound by these two alleles, yet HLA-B53 does not require a tyrosine residue at position 9 of the bound peptide, unlike HLA-B35, which does. HLA-B53, however, can now bind to a different malarial peptide and confers selective advantage to individuals with this allele (42, 43).

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