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Nomenclature for factors of the SLA class-I system, 2004

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Abstract: A systematic nomenclature for the genes and alleles of the swine major histocompatibility complex (MHC) is essential to the development and communication of research in swine immunology. The Swine Leucocyte Antigen (SLA) Nomenclature Committee of the International Society for Animal Genetics has reviewed all of the DNA sequence information for MHC class-I genes, available in GenBank/EMBL/DDBJ databases, and the associated published reports in order to develop such a systematic nomenclature. This report summarizes the proposed nomenclature, which parallels the World Health Organization's nomenclature for factors of the human MHC. The classical class-I SLA genes are designated as SLA-1, SLA-2 and SLA-3; the non-classical as SLA-6, SLA-7 and SLA-8. Nomenclature assignments for all SLA class-I GenBank sequences are now noted. The Committee will add new SLA class-I allele designations, as they are discovered, and will maintain a publicly available list of all recognized genes and alleles by using the International ImMunoGeneTics Project and its Immuno Polymorphism Database/MHC (IPD/MHC) sequence database for MHC sequences in veterinary species.

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Over the last two decades, numerous studies have documented the influence of swine leucocyte antigen (SLA) genes on immune responses, organ and cell transplantation success and disease resistance. Swine with various SLA haplotypes have been shown to develop SLA-dependent titres of complement and antibodies to defined antigens and vaccines (1–4). SLA alleles have been associated with an array of production traits, as reviewed by Vaiman et al. (5). Only a limited number of SLA-based controlled disease-challenge studies have been performed (6). Both primary and secondary responses to the food-borne helminth parasite *Trichinella spiralis* are regulated by SLA-associated genes (7–9), whereas responses to the food-borne protozoan parasite *Toxoplasma gondii* show no SLA association (10). *In vitro* studies of SLA control of anti-bacterial (4, 11) or anti-viral responses (12) need to be confirmed *in vivo* by challenges of SLA-defined pigs.

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SLA association studies have been made possible by the well-characterized panels of SLA typing reagents produced by Vaiman and his colleagues for outbred pigs (13, 14), the availability of the major histocompatibility complex (MHC) inbred lines of national institutes of health (NIH) minipigs produced by Sachs et al. (15), the early sequencing of swine MHC class-I genes by Singer et al. (16) and monoclonal antibodies to SLA antigens (17, 18). DNA-based SLA typing methods, such as restriction fragment length polymorphism (RFLP), polymerase chain reaction (PCR)-RFLP, PCR with site-specific primers (PCR-SSP) and PCR with site-specific oligonucleotide probes (PCR-SSOP), have improved the ability to perform such SLA association studies in outbred pig populations (19–22). High-resolution MHC typing requires DNA cloning and sequencing, which can quickly become labour-intensive and cost-prohibitive. Genomic approaches have resulted in extensive SLA maps and, most recently, complete sequencing of the SLA class-I region (23–27).

Nomenclature Committee of the International Society for Animal Genetics (ISAG)

The ISAG Nomenclature Committee for Factors of the SLA System met at the 28th Annual ISAG Conference in Goettingen, Germany, on 12 August 2002 in order to establish the principles of a systematic nomenclature system for SLA alleles that have been defined by means of DNA sequencing. The meeting has been followed by continued e-mail communications. This report summarizes the results of the Committee's activities for SLA class-I gene nomenclature. The main objectives discussed were:

- The naming of genes within the SLA region.
- The numbering principles for new alleles.
- The DNA sequence requirements for naming a new allele.
- The reference material requirement for naming a new allele.
- The naming of extended SLA haplotypes.
- Submission of DNA sequence data to the IPD/MHC sequence database.

Owing to the efforts of many investigators, there is now sufficient DNA sequence information on the genes and alleles of the swine MHC to propose a DNA-sequence-based nomenclature for *SLA* genes.

Naming of SLA class-I loci

The SLA class-I region has been mapped and completely sequenced by the researchers at INRA, Jouy-en-Josas cedex, France, and Tokai University, Kanagawa, Japan (Fig. 1) (24, 25, 27). This map is based on a single SLA haplotype (H01) and there is evidence that some haplotypes may differ in the number of expressed SLA class-I genes.

A number of SLA class-I genes have been sequenced, most as cDNA. Several SLA class-I haplotypes have been characterized from cDNA by means of reverse transcription (RT-PCR) and sequencing (Smith et al., unpublished data). Most haplotypes have three constitutively expressed classical SLA class-I loci, which correspond to SLA-1, SLA-2 and SLA-3 in the INRA map. Extensive comparisons of SLA class-I and human leukocyte antigen (HLA) class-I sequences have concluded that there is more sequence homology between these loci (SLA-1, SLA-2 and SLA-3) than that between any of these loci and any HLA class-I gene (28) (supplemental materials). Moreover, the order of the SLA class-I genes on the chromosome is not the same as the order of HLA class-I genes (in swine, the non-classical genes are located closer to the class-II region than the classical genes, whereas the non-classical genes are interspersed in the human class-I region) (5, 6, 23, 27, 29). Therefore, some or all of the gene duplications that gave rise to the HLA and SLA class-I genes may have occurred after the divergence of pigs and humans.

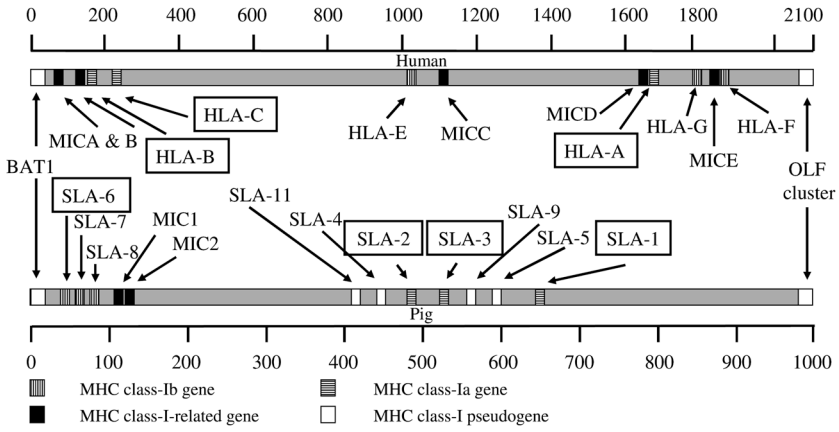

Fig. 1. **Comparative map of the pig and human major histocompatibility complex (MHC) class-I region based on Renard et al. (24).**

The SLA-1 and SLA-3 genes show very strong similarity in their signal peptides and in their 3′ untranslated regions and are quite different from the SLA-2 gene; thus, they probably arose from a more recent gene duplication (30). Three SLA haplotypes have been described that appear to have two loci that are indistinguishable from SLA-1 and may have arisen from another recent gene duplication. Because these loci are currently indistinguishable, these haplotypes will be designated as having two SLA-1 alleles. The same situation was faced in the early development of the HLA nomenclature system, where HLA-A and HLA-B antigens were originally thought to belong to a single locus. When these were recognized as two distinct loci, it was decided not to renumber the antigens, but to make the antigen numbers for the A and B locus non-overlapping (i.e., there is an HLA-A1, but no HLA-B1). When we are able to distinguish the alleles that belong to the SLA-1-like loci, we probably adopt a similar numbering system.

The non-classical HLA class-I genes – HLA-E, HLA-F and HLA-G – show specialized expression patterns that are probably related to specialized functions. There also appear to be one or more non-classical SLA class-I genes, such as SLA-6. It is likely that these genes also play specialized roles, but we do not yet have sufficient information to relate them to the HLA-E, HLA-F and HLA-G genes.

The serologic nomenclature for SLA antigens designates A, B and C loci; however, assigning names based on the serologic loci may imply a homologous relationship with the human HLA locus of the same name. Therefore, we propose to number the SLA class-I loci.

Naming convention 1

The SLA class-I loci will be named after the loci identified in the map of the SLA class-I region published by Renard et al. (24) (Fig. 1), until and unless there is a demonstrated sequence or functional homology to an HLA locus.

Genes of the swine MHC class-I region considered by the Nomenclature Committee

The genes considered by the Committee have been listed in Table 1.

Numbering of SLA class-I alleles

In the HLA nomenclature, the first two digits are used in order to group alleles. Usually, this corresponds to the serologic group to which the allele belongs. Although these groups are composed of alleles with similar sequences, most of the allelic differences can still be recognized by T-cells. This can be observed in the effects of HLA mismatching on the incidence and severity of graft-*vs*-host disease in unrelated donor bone marrow transplants (31). The SLA Nomenclature Committee assigns allele groups based on similarities in sequences. By contrast, the Canine DLA Nomenclature Committee has decided to give each allele a new group number, if any of the amino acid differences are found in regions that are expected to affect the peptide-binding groove. This allele assignment rationale makes good sense concerning the function of MHC proteins; however, it causes practical problems and has not been adopted by the SLA Nomenclature Committee, because it would require high-resolution typing of all individuals.

Low-resolution or medium-resolution MHC typing is often sufficient for many experiments – for example, if one is typing littermates to identify which SLA haplotypes have been inherited. Low/medium-resolution typing might also be sufficient in an initial screen for SLA haplotypes that are linked to genetic traits or to immune responses. However, it is very difficult to talk about the results of low/medium-resolution typing, if alleles are not grouped according to DNA sequence similarities. It is also difficult to talk about the evolutionary relationships of SLA alleles, when a single-base pair change may mean that the new allele has a completely different number. Low/medium-resolution typing can be performed by means of PCR-SSP, PCR-RFLP or PCR-SSOP assays that are cheap and easy.

Names for genes in the SLA class-I region considered by the SLA Nomenclature Committee

Name	Previous equivalents	Molecular characteristics
SLA-1	PD1	Class-I α-chain
SLA-2	PD14	Class-I α-chain
SLA-3	PD7	Class-I α-chain
SLA-4		Class-I α-chain pseudogene
SLA-5		Class-I α-chain possible pseudogene
SLA-6	PD6	Non-classical class-I α-chain
SLA-7		Non-classical class-I α-chain
SLA-8		Non-classical class-I α-chain
SLA-9		Class-I α-chain pseudogene
SLA-11		Class-I α-chain pseudogene
MIC-1		Class-I chain-related pseudogene
MIC-2		Class-I chain-related gene

Gene names are derived principally from the INRA map and the published sequencing of the MHC region of the H01 haplotype (24). Previous equivalents refer to cloned SLA class-I genes from Singer et al. (16). MHC, major histocompatibility complex; SLA, swine leucocyte antigen.

Table 1

High-resolution typing requires cloning and DNA sequencing, which can quickly become labour-intensive and cost-prohibitive.

Naming convention 2

The first two digits will be used in order to designate groups of alleles that have similar DNA sequences. Group names will be based on phylogenetic analysis and the identification of DNA sequence motifs that can be used in order to identify groups with the help of PCR methods. Groups that have at least one confirmed allele will receive a permanent number. If a group does not contain any confirmed alleles, it will be designated with a lower case 'w' in order to indicate a tentative (workshop) designation. Sequences that do not contain the full exon 2 and 3 region and do not match another full-length sequence will not be assigned a group number. Table 2.

Naming convention 3

The third and fourth digits will be used in order to designate alleles that differ in amino acid sequence, with the fifth and sixth digits being used in order to designate alleles that differ only by synonymous substitutions. Table 2.

Naming convention 4

The capital letters N or L will be used in order to designate alleles that have no expression or a low level of expression as protein. If the mutation causing this altered expression occurs outside the protein-coding region of the gene, the allele will be named by using the seventh and eighth digits. Table 2.

The DNA sequence requirements for naming a new allele

Strict quality standards for DNA sequencing are essential to prevent the creation of a large number of non-existent alleles. The following quality standards are adopted.

- Where a sequence is obtained from cDNA or where PCR products are subcloned before sequencing, several clones should have been sequenced.
- Sequencing should always be performed in both forward and reverse directions.
- If direct sequencing of PCR-amplified material is performed, products from at least two separate PCR reactions should have been sequenced.
- In individuals, who are heterozygous for a locus, where one of the alleles is novel, the novel allele must be sequenced in isolation from the second allele. Thus, an allele sequence, where both alleles of a heterozygous individual are sequenced together, is insufficient evidence for the assignment of an official allele designation.
- Sequence derived solely from the primers used for amplifying an allele should not be included in the sequence that is submitted.
- Where possible, a novel sequence should be confirmed by means of a DNA typing method, such as PCR-SSOP, PCR-SSP or PCR-RFLP.
- An accession number in a databank should have been obtained. Sequences may be submitted to these databases online at the following addresses: EMBL: http://www.ebi.ac.uk/submissions/index.html GenBank: http://www.ncbi.nlm.nih.gov/genbank/index.html DDBJ: http://www.ddbj.nig.ac.jp/sub-e.html
- Full-length sequences are preferable, though not essential; the minimum requirements are exons 2 and 3 for an SLA class-I sequence.
- Where possible, a paper should have been submitted for publication.
- DNA or other material – in particular, cell lines – should be made available in a publicly accessible repository or at least in the originating laboratory. The ISAG SLA Nomenclature Committee will maintain documentation on this. The chair of this Committee will serve as curator of this documentation.
- Until an online submission tool is available through the IPD/MHC website, submission of new sequence information should be performed through the chair of the ISAG SLA Nomenclature Committee. Current contact information will be available on the website. Researchers will be expected to complete a question-

Proposed assignment of names and numbers for SLA class-I alleles

Nomenclature	Indicates
SLA	The SLA region and prefix for an SLA gene
SLA-1	A particular SLA locus, i.e. SLA-1
SLA-1*01	A group of SLA alleles (based on DNA sequence similarity)
SLA-1*0101	A specific SLA allele
SLA-1*0101N	A null allele (L = a low-expression allele)
SLA-1*010102	An allele which differs by a synonymous mutation
SLA-1*01010102	An allele which contains a mutation outside the coding region
SLA-1*01010102N	A null allele which contains a mutation outside the coding region

This convention draws heavily on the nomenclature system used for the naming of human MHC genes (32, 33). SLA, swine leucocyte antigen.

Table 2

naire relating to the sequence and to provide a comparison of their new sequence with known related alleles.

Naming convention 5

Sequences that do not meet all of these criteria may be accepted, but will be given a provisional alphanumeric allele name containing two lower-case letters (except L) and two numerals (e.g. SLA-1*an01) (Table 3). If the allele can be assigned to an existing group, then the first two digits will be assigned for that group followed by a provisional alphanumeric allele name (e.g. SLA-1*04gz01). Some groups of sequences may be assigned a provisional (workshop or w) group number, if none of the sequences has been independently confirmed (e.g. SLA-1*w08sz01). Previously published allele sequences will be given a provisional alphanumeric allele name, unless the sequence has been confirmed by more than one laboratory, in more than one breed or by a unique PCR-SSP or PCR-RFLP pattern.

Issues regarding the quality of published DNA sequences

Previously published SLA sequences may not meet all of the quality standards that will be required for prospective submissions; however, these sequences are the basis of this nomenclature system. Nevertheless, it is very important that allele sequences that do not, in fact, exist are not validated, because they will unnecessarily complicate future SLA typing. It is also important to researchers to realize that some of the GenBank/EMBL/DDBJ entries may contain invalid sequences. All of the available information in the GenBank/EMBL/DDBJ entries and in the associated publications has been scrutinized in order to detect any likely sources of sequencing artefacts. The following are some examples of potential sources of sequencing artefacts.

Some GenBank/EMBL/DDBJ entries contain DNA sequence from the primers used for amplifying the genes. One case in which this resulted in an incorrect published sequence is the PC1 allele from the 'c' haplotype in the NIH pigs (AF014003) (Table 4). This sequence contains three bp discrepancies with another GenBank/EMBL/DDBJ entry for the SLA-3 allele of the 'c' haplotype in the NIH pigs (AF464018) at positions +9, 10 and 22 (Table 4). Examination of the associated publication (34) shows that the primer used in order to amplify AF014003 includes the DNA sequence up to position +24. It should also be observed that the PC1 allele was incorrectly described as a homologue of PD1 (SLA-1), but phylogenetic analyses have shown that it is actually a homologue of PD7 (SLA-3). Where it has been determined that a GenBank/EMBL/DDBJ entry has PCR primer sequences included (PSI), this is indicated in the summary tables.

DNA sequences derived from genomic DNA may be susceptible to certain artefacts. The original description of the PD7 coding sequence was incorrect because of the incorrect prediction of an intron/exon splice site (35). The correct coding sequence was determined once the same allele was sequenced from a cDNA library (AF464011). The original description of the PD6 coding sequence was also incorrect (M17014) (Table 5) because of an incorrect sequence near the intron/exon splice site, which caused a frame shift and a second downstream sequencing error that compensated for the frame shift (36). These two errors caused alterations in the amino acid sequence of a section of the third exon, which included the substitution of a critical cysteine residue that is involved in a disulfide bond. The correct sequence was determined from cDNA sequence (AF464020) (Table 5).

Some GenBank entries are determined from partial genomic sequences, in which case it may be difficult to tell whether they are derived from non-expressed pseudogenes. This is particularly important for MHC class-I sequences, because there are a number of closely related pseudogenes. Therefore, any method for DNA-sequence-based typing of SLA class-I genes from genomic DNA would need extensive validation of the locus specificity of the PCR primers used. For this reason, we have not assigned permanent allele names to any SLA class-I alleles derived entirely from partial genomic DNA sequence. Validation of locus-specific primers is of high priority for the Committee, because sequence-based typing is much easier to perform from genomic DNA than to perform from RNA.

DNA sequences that have one or two unique nucleotides at positions constant in all other known alleles have a greater chance of being the result of sequencing artefacts, especially when the DNA sequence matches a previously identified allele except at those one or two positions. The GenBank/EMBL/DDBJ entry for the PA1 allele from the 'a' haplotype in the NIH pigs (AF014002) has two bp discrepancies with another GenBank/EMBL/DDBJ entry for the SLA-1 allele of the 'a' haplotype (AF100665) and an identical sequence from the Sinclair pigs (AY135592) at positions +129 and 130 (Table 3). No other SLA class-I allele matches the AF014002 entry for these two bases. In addition, the PD14 allele from the 'd' haplotype in the NIH pigs (M21058) has 14 bp discrepancies with two other GenBank/EMBL/DDBJ entries for the SLA-2 allele in the 'd' haplotype in the NIH pigs (AF014006 and AF464023) and 11 of these

SLA-1 sequence comparisons and allele assignments

Group	Allele	Previous designation	Comment	Breed	Accession number	Submitter
SLA-1*01	0101	m6		Sinclair	AF464045	Martens et al. (21)
		m6		Unknown	AY135600	Martens et al. (21)
		H01	Genomic	Large White	AJ251829	Renard et al. (24)
	01rh28	H01/28			AF074424	Velten et al. (23)
	0102	H34			AF074433	Velten et al. (23)
		H12			AF074430	Velten et al. (23)
SLA-1*02	0201	a		NIH	AF100665	Smith et al.
		a		Sinclair	AY135592	Martens et al. (21)
		a	PSA: 2 bp ≠ 0101; PSI	NIH	AF014002	Sullivan et al. (34)
	02we02	We2		Westran	AY247765	Lee et al.
SLA-1*03						
SLA-1*04	0401	d	PSI	NIH	AF014005	Sullivan et al. (34)
		wxd		Yucatan	AF464002	Martens et al. (21)
		wxd		Yucatan	AF464016	Martens et al. (21)
		PD1		M. Hairless	AB185316	Ando et al. (37)
		PD1		Clawn	AB185317	Ando et al. (37)
		wxd		Meishan	AY459306	Ho et al.
		PD1		NIH	AF380372	Oleksiewicz et al. (40)
		PD1	PSA: 8 bp ≠ 0301	NIH	M21057	Satz et al. (41)
	04gz01	gz		Guizhou Xiang	AY102468	Chen et al. (42)
	04we01	We1		Westran	AY247764	Lee et al.
	04gx01	gx		Bama	AY102467	Chen et al. (42)
SLA-1*05	0501	m7		Sinclair	AF464044	Martens et al. (21)
		m7		M. Hairless	AB185318	Ando et al. (37)
SLA-1*06	0601	m11		Sinclair	AY135593	Martens et al. (21)
		m11		Hanford	AY135591	Martens et al. (21)
		v472			AY135595	Martens et al. (21)
SLA-1*07	0701	m10		Sinclair	AF464036	Martens et al. (21)
		m10		Hanford	AY459298	Martens et al. (21)
	07ce08	C25E08	EST sequence		AY135587	Martens et al. (21)
SLA-1*w08	w08sz01	z		Yucatan	AF464013	Martens et al. (21)
	w08Lw02	LW2		Large White	AY247767	Lee et al.
	w08sy01	y		Yucatan	AF464015	Martens et al. (21)
	w08sm08	m8		Hanford	AF464043	Martens et al. (21)
	w08ms05	ms5		Meishan	AY459299	Ho et al.
SLA-1*w09	w09sm09	m9		Hanford	AY135594	Martens et al. (21)
SLA-1*w10	w10sm21	m21		Hanford	AY135589	Martens et al. (21)
SLA-1*w11	w11yn01	yn		Yunnan Banna	AY102469	Chen et al. (42)
SLA-1*w12	w12Lw01	LW1		Large White	AY247766	Lee et al.
SLA-1*w13	w13ms21	ms21		Meishan	AY459297	Ho et al.
SLA-1*	–rh03	H03			AF074427	Velten et al. (23)
SLA-1*	–an01	01		M. Hairless	AB105379	Ando et al. (37)
SLA-1*	–an02	02		Goettingen	AB105380	Ando et al. (37)

SLA, swine leucocyte antigen.

Table 3

SLA-3 sequence comparisons and allele assignments

Group	Allele	Previous designation	Comment	Breed	Accession number	Submitter
SLA-3*01	0101	H01	Genomic	Large White	AJ131112	Renard et al. (24)
		H01			AB185320	Ando et al. (37)
	01rh28	H01/28	PSA: 2 bp ≠ 0601		AF074426	Velten et al. (23)
	01ev04	TP1.2w_Pig7_E4			AJ581582	Eiz-Vesper et al.
	01rh12	H12			AF074432	Velten et al. (23)
SLA-3*02						
SLA-3*03	0301	c		NIH	AF464018	Martens et al. (21)
		c		Goettingen	AB185321	Ando et al. (37)
		PC1	PSI, PSA: 3 bp in primer ≠ 0101	NIH	AF014003	Sullivan et al. (34)
	0302	We1		Westran	AY247768	Lee et al.
		01		M. Hairless	AB105387	Ando et al. (37)
	03an02	02		Clawn	AB105388	Ando et al. (37)
	03an04	04		M. Hairless	AB105390	Ando et al. (37)
	03an05	05		Goettingen	AB105391	Ando et al. (37)
SLA-3*04	0401	xd		Yucatan	AF464011	Martens et al. (21)
		xd		NIH	AF464017	Martens et al. (21)
		xd		Hanford	AY135602	Martens et al. (21)
		PD7	Genomic	NIH	M59750	Frels et al. (35)
	04sc19	C19H09	EST clone		AY135586	Martens et al. (21)
		allele 12	exon 2; 1 bp ≠ 02c19		AF000073	Gaycken et al. (43)
			exon 3			
			exon 3			
					AF000074	Gaycken et al. (43)
SLA-3*05	0501	m13		Sinclair	AF464042	Martens et al. (21)
		m13		Hanford	AY135601	Martens et al. (21)
	05sm14	m14		Hanford	AF464041	Martens et al. (21)
	05sw01	w		Yucatan	AF464012	Martens et al. (21)
SLA-3*06	0601	y		Yucatan	AF464010	Martens et al. (21)
		H03			AF074429	Velten et al. (23)
	06an03	03		Clawn	AB105389	Ando et al. (37)
SLA-3*07	0701	z	3 aa insertion	Yucatan	AF464009	Martens et al. (21)
		LW1		Large White	AY247769	Lee et al.
	0701sm19	m19	3 aa insertion	Sinclair	AF464040	Martens et al. (21)
	07Lw02	LW2	3 aa insertion	Large White	AY247770	Lee et al.
	07rh34	H34	3 aa insertion		AF074435	Velten et al. (23)

SLA, swine leucocyte antigen.

Table 4

discrepancies are unique to the M21058 entry (Table 6). The PD1 allele from the 'd' haplotype in the NIH pigs (M21057) has eight bp discrepancies with another GenBank/EMBL/DDBJ entries for the SLA-1 allele in the 'd' haplotype in the NIH pigs (AF014005) and three identical alleles from Yucatan and Landrace pigs (AF464002, AF464016 and AF380372) and six of these discrepancies are unique to the M21057 entry (Table 3). It is, therefore, particularly important to have confirmatory evidence of a unique polymorphic position, such as a unique PCR-RFLP or PCR-SSP pattern.

SLA-6 sequence comparisons and allele assignments

Group	Allele	Previous designation	Comment	Breed	Accession number	Submitter
SLA-6*01	0101	z		Yucatan	AF464006	Martens et al. (21)
		H01		Large White	AJ251914	Chardon et al. (25)
			Endothelial cell line (AOC) (44)	Large White	AY463540	Crew et al. (38)
	0102	d		NIH	AF464020	Martens et al. (21)
		LW1		Large White	AY247772	Lee et al.
		PD6	PSA	NIH	M17014	Ehrlich et al. (36)
	01we01	We1		Westran	AY247771	Lee et al.
	01sc01	c		NIH	AY459304	Martens et al. (21)
	01sx01	x		Yucatan	AF464008	Martens et al. (21)
SLA-6*w02	w02sa01	a		NIH	AF464019	Martens et al. (21)
SLA-6*w03	w03sy01	y		Yucatan	AF464007	Martens et al. (21)

SLA, swine leucocyte antigen.

Table 5

Summary of published SLA class-I DNA sequence data and assignment of allele names

SLA-1 and SLA-3 alleles (Tables 3 and 4)

Both the SLA-1 and SLA-3 alleles have a leader peptide of 21 amino acids, in contrast to the SLA-2 alleles that have a leader peptide of 24 amino acids. They also have a high degree of similarity in their 5′ and 3′ untranslated regions. This sequence similarity likely reflects a derivation of these loci from relatively recent gene duplication. This sequence similarity makes it difficult to design locus-specific primers that would amplify the entire class-I coding sequence. A relatively successful locus-specific primer has been designed that is located more than 400 bp downstream of the stop codon; however, it may not work with all SLA-3 alleles (28). The alternative is to design a primer within the coding sequence, such as that used by Ando et al. (37). The disadvantage of either of these approaches is that the entire coding sequence cannot be determined from a single PCR product. The four sites that appear to be locus-specific for the SLA-3 alleles are the CC at positions +9 and 10, a T at position +51 in exon 1, an A at position +118 in exon 2 and a GC at position +559 and 560 in exon 3. Phylogenetic analysis of the published SLA-3 sequences shows a clustering of SLA-3 sequences that is fairly separate from the SLA-1 sequences.

Three haplotypes have been identified that appear to contain a duplicated SLA-1 locus (Table 7) (28). In each case, both SLA-1 alleles have been found by means of RT-PCR and DNA sequencing, which indicates that they are expressed at least at the mRNA level. In each case, the alleles have had their entire coding region sequenced and they appear to be full-length transcripts without deletions. Their inheritance pattern has been documented by means of PCR-SSP or PCR-RFLP. Using phylogenetic analysis, they are more closely related to other SLA-1 alleles than to the other SLA class-I loci. It has not yet been possible to separate these loci by means of analysis of their DNA sequences.

The SLA-1 locus is highly polymorphic. There are 42 published full or partial DNA sequences, of which 26 are unique (Table 3). There are seven groups with multiple alleles, and eight additional unique sequences, of which five have full exon 2 and 3 sequences. One haplotype has been identified that does not appear to have an expressed SLA-1 locus (the NIH minipig haplotype ‘c’). There are also three sequences that have not yet been assigned to a group. The SLA-1*0401 allele appears to be very common. It has been found in five breeds.

The SLA-3 locus is highly polymorphic. There are 32 published full or partial DNA sequences, of which 20 are unique (Table 4). There are six groups, all with multiple alleles. One haplotype has been identified that does not appear to have an expressed SLA-3 locus (the NIH minipig haplotype ‘a’). The SLA-3*06 alleles have a distinctive insertion of nine bp, which results from a duplication of the preceding nine bp. This creates an insertion of three amino acids at the end of the first alpha helical domain. These alleles are expressed at least at the mRNA level, but there is no direct proof of protein expression. However, a computer model of the three dimensional structure predicts that the three extra amino acids could be accommodated in a functional protein (Smith et al., unpublished data).

SLA-2 sequence comparisons and allele assignments

Group	Allele	Previous designation	Comment	Breed	Accession number	Submitter
SLA-2*01	0101	H01	Genomic	Large White	AJ131112	Renard et al. (24)
		H01/28			AF074425	Velten et al. (23)
	01an06	06			AB105386	Ando et al. (37)
SLA-2*02	0201	a		NIH	AF464021	Martens et al. (21)
		a		Sinclair	AY126719	Martens et al. (21)
		a		Hanford	AY126720	Martens et al. (21)
		a	PSA: 0301 $\neq$ 1 bp	NIH	AF014001	Sullivan et al. (34)
	02Lw02	LW2		Large White	AY247775	Lee et al.
SLA-2*03	0301	c		NIH	AF014004	Sullivan et al. (34)
		c		NIH	AF464022	Martens et al. (21)
	03sp01	p		Sinclair	AF464057	Martens et al. (21)
	03gz01	gz		Guizhou Xiang	AY102471	Chen et al. (42)
SLA-2*04	0401	d		NIH	AF014006	Sullivan et al. (34)
		d		NIH	AF464023	Martens et al. (21)
		PD14	PSA: 14 bp $\neq$ 0101	NIH	M21058	Satz et al. (41)
		d		M. Hairless	AB185319	Ando et al. (37)
	04sx01	x		Yucatan	AF464004	Martens et al. (21)
SLA-2*05	0501	t		Sinclair	AF464054	Martens et al. (21)
		t-1		Sinclair	AY135597	Martens et al. (21)
	05rh34	H34			AF074434	Velten et al. (23)
	05sy01	y		Yucatan	AF464005	Martens et al. (21)
	05rh07	H07		Land $\times$ York	AF380371	Oleksiewicz et al. (40)
	05sz01	z		Yucatan	AF464003	Martens et al. (21)
	05rh03	H03			AF074428	Velten et al. (23)
SLA-2*06	0601	u		Sinclair	AF464053	Martens et al. (21)
		u-1		Sinclair	AY135596	Martens et al. (21)
		u		Hanford	AY135598	Martens et al. (21)
	06sr01	r		Sinclair	AF464055	Martens et al. (21)
	06sv01	v		Sinclair	AF464052	Martens et al. (21)
	06an03	03		Clawn	AB105383	Ando et al. (37)
	06me01	m		Meishan	AF464049	Martens et al. (21)
SLA-2*w07	w07ss01	s		Sinclair	AF464056	Martens et al. (21)
	w07we01	We1		Westran	AY247773	Lee et al.
	w07rh12	H12	short exon 2 sequence		AF074431	Velten et al. (23)
	w07an05	05		M. Hairless	AB105385	Ando et al. (37)
SLA-2*w08	w08sw01	w		Yucatan	AF464014	Martens et al. (21)
	w08gx01	gx		Bama	AY102470	Chen et al. (42)
SLA-2*w09	w09sn01	n		Meishan	AF464059	Martens et al. (21)
	w09an02	02		Clawn	AB105382	Ando et al. (37)
SLA-2*w10	w10sm01	m1		Hanford	AF464039	Martens et al. (21)
	w10an01	01		M. Hairless	AB105381	Ando et al. (37)
SLA-2*w11	w11so01	o		Sinclair	AF464058	Martens et al. (21)
SLA-2*w12	w12Lw01	LW1		Large White	AY247774	Lee et al.
SLA-2*w13	w13sm20	m20		Hanford	AY135599	Martens et al. (21)
SLA-2*w14	w14yn01	yn		Yunnan Banna	AY102472	Chen et al. (42)
SLA-2*	an04	04		Goettingen	AB105384	Ando et al. (37)

SLA, swine leucocyte antigen.

Table 6

SLA class-I haplotype assignments

Hp-	Breed	Previous designation	SLA-1	SLA-3	SLA-2	SLA-6
1a.0	Large White	H01	0101	0101	0101	0101
1b.0		H28	01rh28	01rh28	0101	
2.0	NIH	a (NIH)	0201, 0701	null	0201	02sa01
	Sinclair	b (Sinclair)				
	Hanford					
3.0	NIH	c (NIH)	null	0301	0301	01sc01
4a.0	NIH	d (NIH)	0401	0401	0401	0102
4b.0	Yucatan	x (Yucatan)	0401	0401	04sx01	01sx01
5.0	Yucatan	w (Yucatan)	0401	05sw01	08sw01	null
6.0	Yucatan	y (Yucatan)	w08sy01	0601	05sy01	03sy01
7.0	Yucatan	z (Yucatan)	w08sz01	0701	05sz01	0101
8.0	Westran		02we02, 04we01	0302	07we01	01we01
9.0	Sinclair	a (Sinclair)	0601	0501	0601	
	Hanford					
10.0	Sinclair	c (Sinclair)	0501	?	?	
11.0	Sinclair	d (Sinclair)	0101	0701sm19	0501	
12.0	Hanford	e (Hanford)	w08sm08, w09sm09	05sm14	10sm01	
13.0	Hanford	f (Hanford)	w10sm21	0401	w13sm20	
14.0		H12	0102	01rh12	07rh12	
15.0		H34	0102	07rh34	05rh34	

All class-II haplotypes have been given the provisional number 0 designation. The SLA Nomenclature Committee will next address class-II nomenclature and assign class-II haplotypes at that time; ? = specific allele at this locus not yet defined.

Table 7

SLA-5 allele

One genomic sequence of an SLA-5 allele has been described in the H01 haplotype by the INRA researchers (Fig. 2 and supplemental material). This allele appears to code for a full-length mRNA that is consistent with a functional MHC class-I protein, but there has been no proof of mRNA expression. No SLA-5 clones were found in a cDNA library of spleen tissue by using an MHC class-I gene probe (Smith et al., unpublished data) and no EST clones found in GenBank closely match SLA-5. Other studies have used locus-specific PCR primers in order to amplify expressed MHC class-I genes and might, therefore, not have been able to detect SLA-5 mRNA. The promoter region of the SLA-5 gene has several mutations, which may modify or eliminate its expression. One might ask whether the apparently duplicated SLA-1 loci described above represent SLA-5 alleles. This seems unlikely, because the SLA-5 DNA sequence has some unique features that differ from the unique features of the other SLA-1 alleles, including a mismatch in the 3′ locus-specific primer sequence used for amplifying many of the SLA-1 alleles. In addition, the SLA alleles from haplotypes with a duplicated SLA-1 locus are more closely related to other SLA-1 alleles than to the SLA-5 gene sequence. The SLA-1, SLA-3 and SLA-5 phylogeny (Fig. 2) illustrates these relationships.

SLA-2 alleles (Table 6)

In addition to the difference in the length of the leader peptide, SLA-2 alleles are easily distinguished from SLA-1 and SLA-3 alleles by substantial differences in their 5′ and 3′ untranslated regions. This makes it relatively easy to design locus-specific primers. This is important, because phylogenetic analysis will not reliably distinguish between alleles of SLA-2 and SLA-1 when comparing exon 2 and 3 sequences.

The SLA-2 locus is highly polymorphic. There are 46 published full or partial DNA sequences, of which 35 are unique (Table 3). There are 10 groups with multiple alleles, and five additional unique sequences, of which four have full exon 2 and 3 sequences. The SLA-2 sequence phylogeny has been shown in Fig. 3.

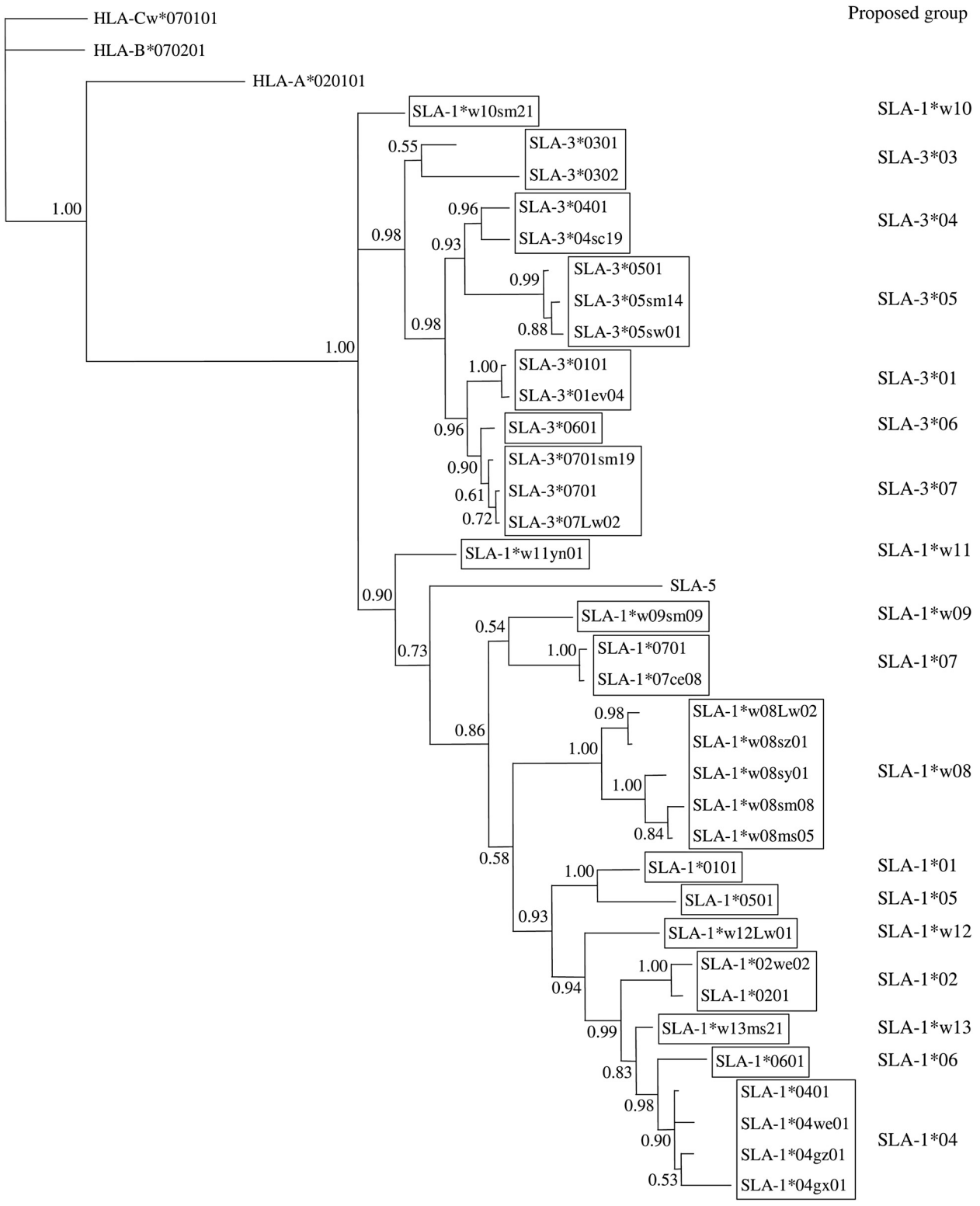

Fig. 2. **SLA-1, SLA-3 and SLA-5 phylogeny.** Only GenBank sequences containing full SLA class-I exon 2 and 3 were considered for establishing SLA class-I gene phylogeny, based on Bayesian analysis (Mr Bayes, http://morphbank.ebc.uu.se/mrbayes/info.php). Detailed sequence data and multiple sequence alignments are based on GenBank/EMBL/DDBJ sequences retrieved as of 20 September 2003. SLA, swine leucocyte antigen.

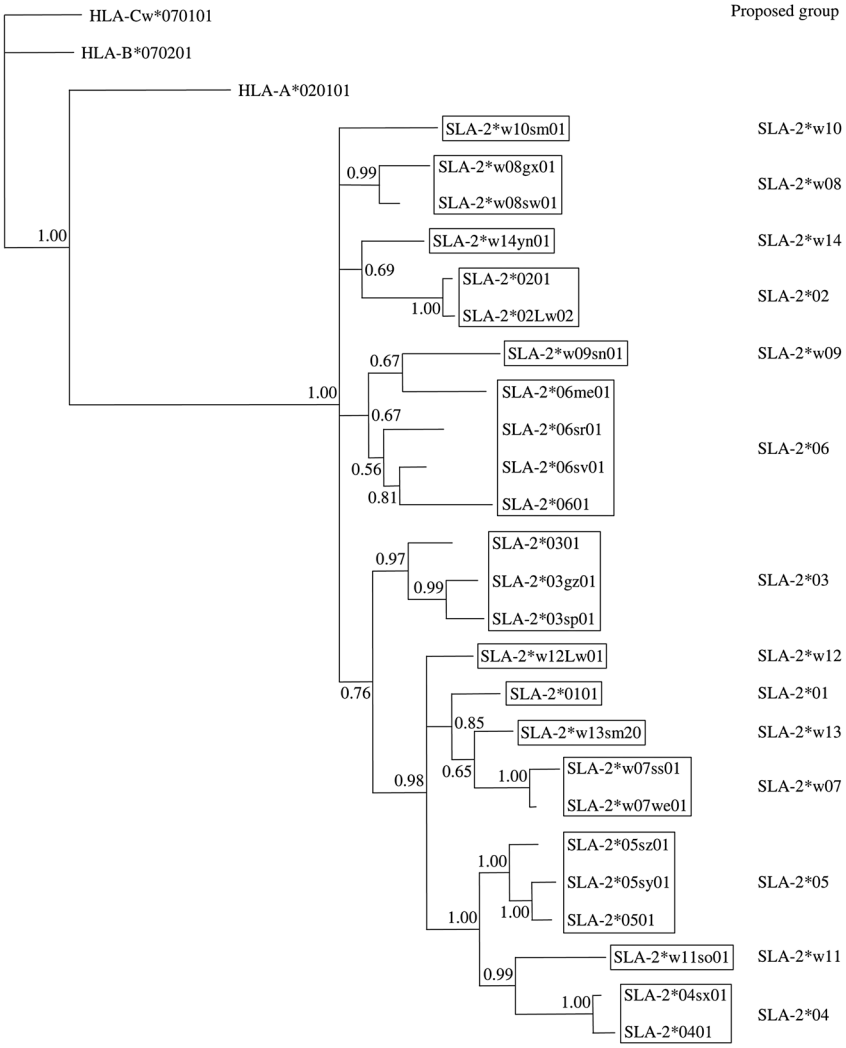

Fig. 3. **SLA-2 phylogeny.** Only GenBank sequences containing full SLA class-I exon 2 and 3 were considered for establishing SLA class-I gene phylogeny, based on Bayesian analysis (Mr Bayes). Detailed sequence data and multiple sequence alignments are based on GenBank sequences retrieved as of 20 September 2003. SLA, swine leucocyte antigen.

SLA-6 alleles (Table 5)

The SLA-6 locus has limited polymorphism. Only two polymorphisms occur within the alpha-1 or alpha-2 domains. One occurs at bp 465, which causes a substitution of Glu for Asp at residue 106 in the 'y' allele, which is a conservative substitution. Another polymorphism occurs at bp 679, which causes a substitution of Met for Val at residue 227 in the NIH minipig 'a' allele, which is a conservative substitution. Therefore, we have assigned alleles to three groups based on these substitutions. This limited polymorphism of SLA-6 is similar to the limited polymorphism of 'non-classical' MHC class loci, such as HLA-E, HLA-F and HLA-G in humans. SLA-6 mRNA has been found in many tissues with the highest level found in lymphoid tissue, which is a pattern of expression, which is more similar to HLA-E than to HLA-F or HLA-G (36).

SLA-7 and SLA-8 alleles

Very little sequence data has been published for SLA-7 or SLA-8 alleles. Genomic DNA sequences from the H01 haplotype have been published (AJ251914) (25). Crew et al. (38) have cloned a single SLA-7 and SLA-8 allele from an immortalized porcine aortic endothelial cell line (AY463541 and AY463542). Their cDNA clone of SLA-7 differed from the predicted cDNA sequence from genomic DNA, because exon 7 began 4 bp upstream from the predicted splice site. These investigators also found that SLA-8 was the most abundant transcript found in embryonic and placental tissue.

Naming of SLA haplotypes (Table 7)

We expect that many haplotypes that have been defined by using serologic typing will be divisible into various haplotypes based on high-resolution typing with the help of DNA sequencing. In one

example, the class-I haplotype of the 'd' haplotype of NIH minipigs is almost identical to the class-I haplotype of the 'x' haplotype of Yucatan miniature pigs. For the 'x' haplotype, the SLA-1 and SLA-3 alleles are identical and the SLA-2 gene has a few base pair differences confined to a single small region from the 'd' haplotype (supplemental materials). The 'd' haplotype types serologically as H04. Unless the small change in the SLA-2 gene changes its serologic type, the 'x' haplotype would also type as H04. In addition, the H04 haplotype is found frequently in the Danish population of Duroc pigs; however, we have found an SLA-2 allele in a Duroc pig that differs from the SLA-2 allele of the H04 haplotype in the 'd' line of NIH miniature pigs (Smith et al., unpublished data).

The haplotypes that have been defined by means of serologic typing almost exclusively take into consideration the SLA class-I genes. Because crossovers between the class-I and class-II regions occur relatively frequently (29, 39), it is necessary to have a nomenclature system that considers both class-I and class-II genes. We propose that the first number represents the class-I haplotype and the second number represents the class-II haplotype.

Naming convention 6

Haplotypes defined by means of high-resolution DNA sequencing will be named with a prefix Hp- and a number for the class-I haplotype followed by a number for the class-II haplotype separated by a period (i.e., Hp-1.1). If no typing for the associated class-I or class-II alleles is available, this will be indicated by using the number 0 (i.e. Hp-1.0). Because the ISAG SLA Nomenclature Committee will next address SLA class-II nomenclature, all class-II haplotypes are currently assigned the number 0.

Although crossovers within the class-I region occur infrequently, they have been documented (29, 39). In addition, it would be helpful in the future to be able to denote the similarity of haplotypes that have arisen from the mutation of a single gene. Therefore, it would be useful to have a modifier that would denote haplotypes with very similar SLA alleles.

Naming convention 7

The number designation of an SLA haplotype may be modified with a lower-case letter (i.e. 1a.1 *vs* 1b.1) in order to designate a second haplotype that is closely related to the original haplotype.

SLA class-I haplotypes that have been characterized by DNA-sequence-based typing and have been assigned a haplotype number have been listed in Table 7.

Discussion

A systematic nomenclature for SLA class-I alleles is critical of the further development of research in swine immunology and disease research and for the use of the swine as a transplantation model and xenotransplantation donor. It allows the investigators to communicate more easily about SLA alleles and haplotypes, particularly in outbred pigs, where there are few molecularly defined SLA haplotypes. MHC proteins play a central role in the presentation of antigenic peptides to $CD8^+$ T-cells; powerful new technologies, such as MHC/peptide tetramers, will be extremely useful in the study of cell-mediated immunity to pathogens and vaccine responses. Full sequence comparisons will further the definition of the peptide-binding motifs of individual SLA alleles. This will inform the discovery of T-cell epitopes in viral or bacterial proteins, particularly those that derive from conserved portions of viral genomes. Overall, such information will be very useful for designing vaccines that produce effective protective immunity for infectious diseases in pigs.

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