CD Antigens 2001

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The Tradition of Human Leukocyte Differentiation Antigen Workshops

The process of categorizing the antigenic molecules and epitopes associated with human white cells, via the collaborative study of monoclonal antibodies, dates back to the early 1980s, when the first HLDA (Human Leukocyte Differentiation Antigen) Workshop was held in Paris. This initial meeting listed only 15 agreed molecular entities, but it created an internationally agreed basis for the nomenclature of leukocyte molecules (the CD scheme) and also provided a forum for reporting studies on their function and practical relevance. A further six HLDA meetings have been held since the first Paris meeting. The most recent of these (HLDA7) took place last year in Harrogate, UK, and the proceedings of the meeting will be published later this year (*Leucocyte Typing VII,* Oxford University Press).

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The Aims and Approaches of the 7th HLDA Workshop

The Limitations of "Blind" Antibody Screening

It was apparent at the previous meeting, HLDA6, held in Kobe, Japan, in 1996, that the technique of detecting molecular entities by screening coded panels of monoclonal antibodies against human cells was becoming obsolescent. Antibodies to the most immunogenic molecules had already been produced, and fewer laboratories than in the early days were prepared to devote resources to raising new antibodies because the probability of finding novel reagents becomes ever less likely. In consequence, many antibodies in the 6th Workshop were reagents (submitted by laboratories that were not equipped to characterize them) that proved to be of known specificity.

Selection of Antibodies

With these considerations in mind, the 7th Workshop adopted a different approach: instead of screening poorly characterized antibodies, reagents were selected (and actively solicited) for which at least some molecular data were already available. A substantial number of monoclonal antibodies reactive with leukocyte-associated molecules exist that do not meet the traditional criterion for establishing a new CD specificity (*i.e.*, the existence of at least two independent antibodies of the same spec-

TABLE 1. New CD Designations

CD Designation	Name	Section	Locus Link
CD15u	Sulphated CD15	Carbohydrate structures	
CD60a	GD3	Carbohydrate structures	
CD60b	9-O-acetyl-GD3	Carbohydrate structures	
CD60c	7-O-acetyl-GD3	Carbohydrate structures	
CD75	Lactosamines	Carbohydrate structures	
CD75s	Alpha-2,6-sialylated lactosamines (formerly CDw75 and CDw76)	Carbohydrate structures	
CD85	ILT/LIR family (see Table 2)	Dendritic cells	
CD110	MPL, TPO R	Platelets	4352
CD111	PRR1/Nectin1	Myeloid cells	5818
CD112	PRR2	Myeloid cells	5819
CD133	AC133	Stem/progenitor cells	8842
CD156b	TACE/ADAM17	Adhesion structures	6868
CD158	KIR family (see Table 2)	NK cells	
CD159a	NKG2A	NK cells	3821
CD160	BY55	T cells	11126
CD162R	PEN5	NK cells	6404
CD167a	Discoidin domain R (DDR1)	Adhesion structures	780
CD168	RHAMM	Adhesion structures	3161
CD169	Sialoadhesin	Adhesion structures	6614
CD170	Siglec-5	Adhesion structures	8778
CD171	L1	Adhesion structures	3897
CD172a	SIRPα	Adhesion structures	8194
CD173	Blood group H type 2	Carbohydrate structures	
CD174	Lewis y	Carbohydrate structures	
CD175	Tn	Carbohydrate structures	
CD175s	Sialyl-Tn	Carbohydrate structures	
CD176	TF	Carbohydrate structures	
CD177	NB1	Myeloid cells	
CD178	Fas ligand	Cytokine/chemokine receptors	356
CD179a	Vpre-B	B cells	7441
CD179b	Lambda 5	B cells	3543
CD180	RP105	B cells	4064
CD183	CXCR3	Cytokine/chemokine receptors	2833
CD184	CXCR4	Cytokine/chemokine receptors	7852
CD195	CCR5	Cytokine/chemokine receptors	1234
CDw197	CCR7	Cytokine/chemokine receptors	1236
CD200	OX2	Nonlineage molecules	4345
CD201	EPC R	Endothelial cells	10544
CD202b	Tie2 (Tek)	Endothelial cells	7010
CD203c	NPP3/PDNP3	Myeloid cells	5169
CD204	Macrophage scavenger R	Myeloid cells	4481
CD205	DEC205	Dendritic cells	4065
CD206	Macrophage mannose R	Dendritic cells	4360
CD207	Langerin	Dendritic cells	50489
CD208	DC-LAMP	Dendritic cells	
CD209	DC-SIGN	Dendritic cells	30385
CDw210	IL-10 R	Cytokine/chemokine receptors	3587; 3588
CD212	IL-12 R	Cytokine/chemokine receptors	3594
CD213a1	IL-13 R α 1	Cytokine/chemokine receptors	3597
CD213a2	IL-13 R α 2	Cytokine/chemokine receptors	3598
CDw217	IL-17 R	Cytokine/chemokine receptors	23765
CD220	Insulin R	Nonlineage molecules	3643
CD221	IGF1 R	Nonlineage molecules	3480
CD222	Mannose-6-phosphate/IGF2 R	Nonlineage molecules	3482
CD223	LAG-3	Nonlineage molecules	3902
CD224	γ-glutamyl transferase	Nonlineage molecules	2678
CD225	Leu13	Nonlineage molecules	8519
CD226	DNAM-1 (PTA1)	T cells	10666
CD227	MUC.1	Nonlineage molecules	4582
CD228	Melanotransferrin	Nonlineage molecules	4241
CD229	Ly9	Nonlineage molecules	4063
CD230	Prion protein	Nonlineage molecules	5621
CD231	TALLA-1/A15	Nonlineage molecules	7102
CD232	VESP R	Nonlineage molecules	10154
CD233	Band 3	Erythroid cells	6521
CD234	Fy-glycoprotein (DARC)	Erythroid cells	2532
CD235a	Glycophorin A	Erythroid cells	2993
CD235b	Glycophorin B	Erythroid cells	2994
CD235ab	Glycophorin A/B crossreactive mabs	Erythroid cells	
CD236	Glycophorin C/D	Erythroid cells	
CD236R	Glycophorin C	Erythroid cells	2995
CD238	Kell	Erythroid cells	3792
CD239	B-CAM	Erythroid cells	4059
CD240CE	Rh30CE	Erythroid cells	6006
CD240D	Rh30D	Erythroid cells	6007
CD240DCE	Rh30D/CE cross-reactive mabs	Erythroid cells	
CD241	RhAg	Erythroid cells	6005
CD242	ICAM-4	Erythroid cells	3386
CD243	MDR-1	Stem/progenitor cells	
CD244	2B4	NK cells	51744
CD245	p220/240	T cells	
CD246	Anaplastic lymphoma kinase	T cells	238
CD247	ζ chain	T cells	919

ificity). This rule dates from the first HLDA Workshop two decades ago: since that time, biochemical and molecular biological techniques for characterizing the targets of new antibodies have come to be widely used. Consequently, it is now considered appropriate to establish a CD designation for a molecule if its gene has been cloned and at least one specific monoclonal antibody has been studied in the Workshop.

New Workshop Sections

Four new sections were introduced in the 7th HLDA Workshop to add to the traditional list from past meetings: namely, Dendritic Cells, Stem/Progenitor Cells, Erythroid Cells, and Carbohydrate Structures. Although it has been recognized for many years that monoclonal antibodies reactive with human leukocytes can be specific for carbohydrate epitopes (*e.g.*, the carbohydrate CD category CD15 was identified at the first Workshop), they had not received specific attention in any Workshop. The inclusion of erythroid molecules, although it may seem out of place in a "Leukocyte Workshop," was justified by the number of molecules shared between white and red cells (*e.g.*, cytokine receptors) that hint at unexplored functions of red cells.

TABLE 2. New CD Nomenclature for ILT/LIR and KIR Molecules

CD Designation	Name
The ILT/LIR family	
CD85a	ILT5/LIR3
CD85b	ILT8
CD85c	LIR8
CD85d	ILT4/LIR2, MIR10
CD85e	ILT6/LIR4
CD85f	ILT11
CD85g	ILT7
CD85h	ILT1/LIR7
CD85i	LIR6
CD85j	ILT2/LIR1, MIR7
CD85k	ILT3/LIR5
CD85l	ILT9
CD85m	ILT10
The KIR family	
CD158z	KIR3DL7/KIRC1
CD158b1 and CD158b2	KIR2DL2/p58.2 and KIR2DL3/p58.3
CD158a	KIR2DL1/p58.1
CD158c	KIR2DS6/KIRX
CD158d	KIR2DL4
CD158e1 and CD158e2	KIR3DL1/p70 and KIR3DS1/p70
CD158f	KIR2DL5
CD158g	KIR2DS5
CD158h	KIR2DS1/p50.1
CD158i	KIR2DS4/p50.3
CD158j	KIR2DS2/p50.2
CD158k	KIR3DL2/p140

For further details of this classification, based on the position of the genes on chromosome 19q;13.4 from centromeric to telomeric loci, see André *et al.* (1).

The Yield of New CD Specificities in the 7th HLDA Workshop

This more active approach to the identification of new CD specificities represented a break with tradition, but the results justified the new approach because a total of well over 80 new entities were added to the list of CD specificities. This compares favorably with previous Workshops (an average of less than 30 CD specificities per Workshop), and it also largely avoided the laborious screening in multiple laboratories of antibodies that prove to be directed against known CD molecules.

Tables 1 and 2 list the new specificities established at the 7th Workshop. Full details will be found in *Leucocyte Typing VII*, and molecular, functional, and other data can be found for many of these new specificities on the Website for "Protein Reviews on the Web" (www.ncbi.nlm.nih.gov/prow/).

The Eighth Workshop

Plans are well advanced for the 8th Workshop, to be organized in Adelaide in 2004 under the aegis of Prof. H. Zola (see www.hlda8.org). It is sometimes assumed that the catalog of surface molecules associated with human hemopoietic cells is now essentially complete, but there is abundant evidence in the literature for novel surface molecules that would merit study at the next Workshop and that could provide the basis for new CD designations. Table 3 comprises a list of potential new molecules reported after the production of monoclonal antibodies and also a more extensive list of surface molecules identified by gene cloning. In most instances, no antibodies are available against the putative new leukocyte/endothelial markers in this latter group. Specific and well-characterized reagents, whether monoclonal or polyclonal, are needed not only for detecting these new "virtual" molecules but also for defining functional domains, for characterizing 3D protein structure, and for analyzing protein–protein interactions. It may be added that cloning of gene sequences often reveals multiple members of new or existing molecular families (*e.g.*, the Toll-like receptors) and may identify surface receptors that bind more than one ligand or *vice versa* (*e.g.*, the TALL-1 and APRIL ligands for TACI and BCMA). Furthermore, a number of leukocyte-associated markers have been cloned from mice and other species, and almost all will have human homologues. The 8th Workshop will provide a forum for a range of antibody-based studies relating to this accumulating corpus of genomic and proteomic data.

As in the 7th Workshop, in which four new sections were added, it may be possible to include neuronal cells in the 8th Workshop. Many neuronal

TABLE 3. Examples of Possible Future CD Specificities

Molecule	Molecule Size	Cell Types	Comments	Reference no.
Identified after antibody production				
AM-3K antigen	70 and 120 kDa	Macrophages		(2)
BDCA-2, BDCA-3, and BDCA-4 antigens		Dendritic cells	Identifies subsets of dendritic cells	(3)
BENE	17 kDa	Endothelium	"Raft-associated" member of MAL family; interacts with caveolin-1	(4)
CMRF-44	?	Dendritic cells	Differentiated/activated	(5, 6)
CMRF-56	95 kDa	Dendritic cells	Differentiated/activated	
H47 antigen	100 kDa (non red.) 120 kDa (red.)	T cells & most NK and B cells and monocytes	? Involved in T-cell activation	(7)
Hal-1	200 kDa (100 kDa)	T cells, EBV-transformed B cells, myelo-monocytic cells, anaplastic large cell lymphoma	? New lymphoma marker	(8)
LAK1 and LAK2 antigens	120 kDa and 110 + 140 kDa, respectively	LGL and LAK cells		(9)
NKp80	80-kDa dimer	NK cells and CD56-positive T cells	Novel member of the killer cell lectin-like receptor gene family, encoded by KLRF1 gene; triggers NK cell cytotoxicity	(10, 11)
VAP-1 (vascular adhesion protein)	90 kDa	Endothelium	Mediates lymphocyte–endothelial adhesion; has monoamine oxidase activity	(12, 13)
Wue-1 antigen	94 kDa	Plasma cells	Stimulates growth of plasma cells	(14)
Identified by gene cloning				
B-cell maturation factor	184 aa	B cells	TNFR family member; receptor for TALL-1 and APRIL	(15, 16)
B7-H2	302 aa	Dendritic cells	New member of B7 family; binds ICOS on activated T cells	(17)
CLEC-1	280 aa	Dendritic cells	Novel C-type lectin-like receptor with cytoplasmic tyrosine-based motif	(18)
CMRF-35A	224 aa	NK cells, neutrophils, monocytes,	Novel Ig superfamily receptors; CMRF-35H contains 3 cytoplasmic tyrosine based motifs	(19, 20)
CMRF-35H	300 aa	dendritic cells, and subset of T lymphocytes		
CS1		NK cells	Novel receptor belonging to CD2 subset of Ig superfamily	(21)
DC-STAMP	470 aa	Dendritic cells	Novel protein containing seven putative transmembrane domains.	(22)
EMR3	652 aa	Mainly leukocyte restricted. Highest levels on neutrophils, monocytes, and macrophages	Novel EGF-TM7 molecule; interacts with a surface ligand on myeloid cells	(23)
Flt-1 (VEGFR-1)		Endothelial cells, monocytes		(24)
GPRv53	390 aa	Leukocytes	Identified by gene cloning; G-protein–coupled histamine receptor	(25)
IRTA1 and IRTA2		Subpopulations of B cells	Homologous to the Fc and inhibitory receptor families	(26)
M160	1453 aa	Macrophages	New member of scavenger receptor cysteine-rich superfamily	(27)
MARCO (macrophage receptor with collagenous structure)	520 aa	Macrophages	Class A scavenger receptor; involved in bacterial clearance *in vivo*	(28, 29)
TACI	293 aa	B cells	TNFR family member; receptor for TALL-1 and APRIL	(30)
TREM-1 and TREM-2 (triggering receptors expressed on myeloid cells)		Neutrophils and subset of monocytes (TREM-1) and macrophages (TREM-2)	Novel Ig superfamily receptors; TREM-1 triggers neutrophil secretion (*e.g.*, IL-8) and degranulation; TREM-2 activates macrophages, and both associate with DAP12	(31, 32)

cells express cell surface proteins found on leukocytes and *vice versa* (*e.g.*, CD56, CD100, CD168, CD171). Furthermore, the guidance cues used by neuronal cells share similarities to those involved in leukocyte extravasation, so the expression of these molecules in common may reflect shared biological processes. It may also be noted that other molecules such as the mucins, thought to be primarily associated with epithelial cells, are now being described on leukocytes.

Finally, it remains to be established how the 8th and subsequent HLDA Workshops should deal with lineage- or stage-restricted leukocyte molecules that are localized within the cell cytoplasm (or nucleus).

Given the importance of many of these molecules in signaling pathways initiated via known surface CD molecules, their identification and study is an inevitable extension of the work of the first seven HLDA Workshops. Whether or not a new "intracellular CD" categorization scheme is devised for such molecules, they are of interest for many laboratories studying human hematopoietic cells, and their investigation will be among the aims of the next Workshop.

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Errata

In the *Modern Pathology* November article "Immunohistochemical Detection of the Alternate *INK4a*-Encoded Tumor Suppressor Protein $p14^{ARF}$ in Archival Human Cancers and Cell Lines Using Commercial Antibodies: Correlation with $p16^{INK4a}$ Expression" by Joseph Geradts, M.D., Robb E. Wilentz, M.D., and Helen Roberts, B.Sc. (Mod Pathol 2001:14(11):1162–1168), the title was misprinted. The error was corrected before the article was posted to the Internet.

In the *Modern Pathology* December article "Immunohistochemical Analysis of RCAS1 in Human Pituitary Adenomas," author Takeshi Watanabe's name was misspelled.