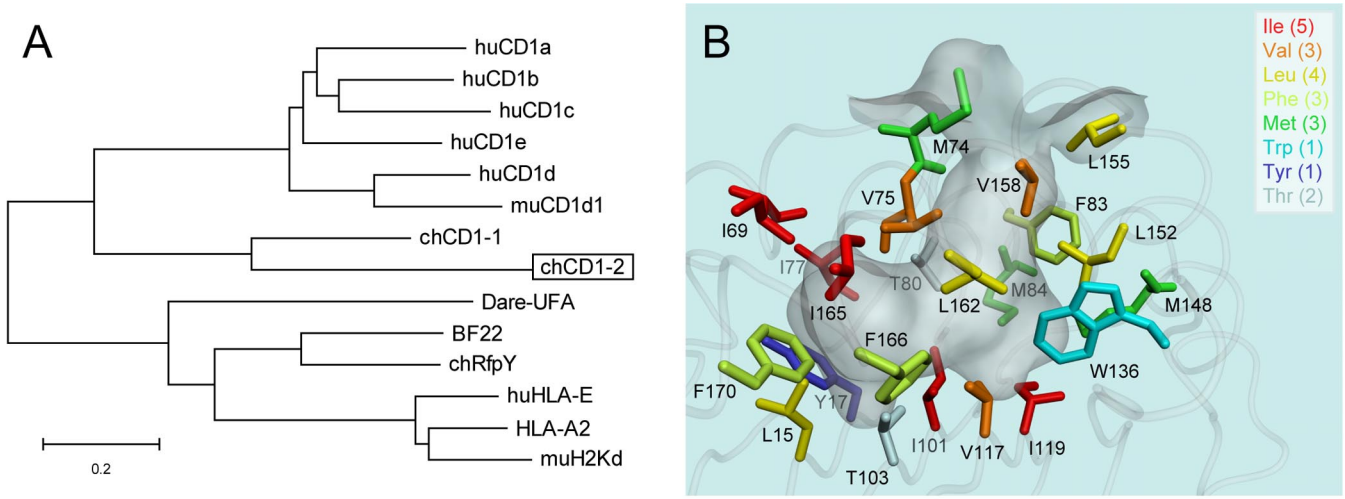
Supporting Information

Zajonc *et al.* 10.1073/pnas.0809814105



C

α1 α A' loop α

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chCD1-2 : ETSCPPPEESQFFQLFYTLLLGNVSSTELTGMALLADVPIMVLDPHTWNLNICRPWVQEITAETEVKKILSFSMVGIRNTIRFMHEMTAKAGLD-- : 94
chCD1-1 : PGTTAEPEGSHMLKLLHFATFQNSTSVLVGGLGLLGDVKMGSLDSRTGNIRYYRPWLRPSLPKGDWDVIESSIKSYVRDFSRLVQMYT---TVP-- : 91
huCD1a  : DGNADGLKEPLSFHVIWIASFYNHSWKQNLVSGWLSDLQTHTWDSNSSTIVFLWPWSRGNFSNEEWKELETLFRIRTIRSFEGIRRYAHELQFE-- : 94
huCD1b  : GNSEHAFQGPTSFHVIQTSSFTNSTWAQTQGSGWLDDLQIHGWDSDSGTAIFLKPWSKGNFSDKEVAELEEIFRVYIFGFARELVQDFAGDFQMK-- : 94
huCD1c  : GDNADASQEHVSFHVIQIFSFVNQSWARGQGSGWLDELQTHGWDSESGTIIFLHNWSKGNFSNEELSDLELLFRFYLFGLTREIQDHASQDYSK-- : 94
huCD1d  : WGSAEVPQRLFPLRCLQISSFANSSWTRTDGLAWLGELQTHSWSNDSDTVRSLKPWSQGTFSDQQWETLQHIFRVYRSSFTRDVKEFAKMLRLS-- : 94
muCD1d1 : WGQSEAQQKNYTFRCLQMSSFANRSWSRTDSVVWLGDLQTHRWSNDSATISFTKPWSQGKLSNQQWEKLQHMFQVYRVSFTRDIQELVKMMSPKED : 96
huCD1e  : GENTAAAEEQLSFRMLQTSSFANHSWAHSEGSGWLGDLQTHGWDTVLGTIRFLKPWSHGNFSKQELKNLQSLFQLYFHSFIQIVQASAGQFQLE-- : 94
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α2 α

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chCD1-2 : YPRVFQIHTGCKLYTNGTRWSFVNIGEGGRDLVTYELSRERWVPQRSTLLAKVMSNTLTDLRAVSGFLEHIFSSSFPNYILMLHEEGRTDLERRVPP : 191
chCD1-1 : YPFVFQSSIGCELQSNGTIRTFFDIAYEGQNFLRFNLDAGTWDQMQHNQLSAKAEHLMANASTLNEVIQVLLNDTCVDILRLFIQAGKADLERQVPP : 188
huCD1a  : YPFEIQVTGGCELHSGKVSGSFLQLAYQGSDFVSFQNNSWLPYPVAGNMAKHFCKVL-NQNQHENDITHNLLSDTCPRFILGLLDAGKAHLQRQVKP : 190
huCD1b  : YPFEIQGIAGCELHSGGAIVSFLRGALGGLDFLSVKNASCVPSPEGGSRAQKFCALI-IQYQGIMETVRILLYETCPRYLLGVLNAGKADLQRQVKP : 190
huCD1c  : YPFEVQVKAGCELHSGKSPEGFFQVAFNGLDLLSFQNTTWVPSPGCGSLAQSVCHLLNHQYEGVTETVYNLIRSTCPRFLLGLLDAGKMYVHRQVRP : 191
huCD1d  : YPLELQVSAGCEVHPGNASNNFFHVAFQGKDILSFQGTSWEPTQEAPLWVNLAIQVL-NQDKWTRETVQWLLNGTCPQFVSGLLESGKSELKKQVKP : 190
muCD1d1 : YPIEIQLSAGCEMYPGNASESFLHVAFQGKYVVRFWGTSWQTVPGAPSWLDLPIKVL-NADQGTSATVQMLLNDTCPLFVRGLLEAGKSDLEKQEKP : 192
huCD1e  : YPFEIQILAGCRMN---APQIFLNMAYQGSDFLSFQGISWEPSPGAGIRAQNICKVL-NRYLDIKEILQSLLGHTCPRFLAGLMEAGESELKRKVKP : 187
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Fig. S1. CD1 sequence alignment and groove amino acid composition. (*A*) Relationship of chCD1 proteins to avian and mammalian non-classical and classical MHC molecules. (*B*) Chicken CD1–2-binding groove close-up (*gray*) with groove-forming residues colored by amino acid type as shown in the inset. The outer molecular surface of CD1–2 is removed for clarity. (*C*) Sequence alignment between chicken, mouse, and human CD1s. Identical or highly similar residues are indicated by black bars. Residues that form the chCD1–2-binding groove are colored as in *B*. The black arrows indicate chCD1–2 residues that were mutated to form the CD1a-like binding groove illustrated in Fig. 5 *A* and *B*. Red arrows indicate mutations resulting in the CD1d-like groove.

Table S1. Data collection and refinement statistics for chCD1–2

Data collection	
Resolution range (Å)	30.0–2.0 (2.07–2.00)*
Completeness (%)	93.2 (95.6)
Number of unique reflections	26,874
Redundancy	4.1
R_{sym}† (%)	8.2 (64.9)
I/σ_I	23.0 (2.0)
Refinement statistics	
Number of reflections ($F > 0$)	26,843
Maximum resolution (Å)	2.0
R_{cryst}‡ (%)	21.6 (25.0)
R_{free}§ (%)	26.6 (27.0)
Number of atoms	3,214
Protein	3,005
Palmitic acid	18
N-linked carbohydrates	42
Water	149
Ramachandran statistics (%)	
Favored	96.3
Allowed	3.7
Root-mean-square deviation from ideal geometry	
Bond length (Å)	0.015
Bond angles (°)	1.6
Average B values (Å^2)¶	
Protein	52.5
Palmitic acid	66.1
Water molecules	48.7
Carbohydrates	92.4

*, Number in parentheses refer to the highest resolution shell.
†, $R_{sym} = (\Sigma_h\Sigma_i|I_i(h)-<I(h)>|\Sigma_h\Sigma_i I_i(h)) \times 100$, where $< I(h)>$ is the average intensity of i symmetry-related observations for reflections with Bragg index h.
‡, $R_{cryst} = (\Sigma_{hkl}F_o-F_c/\Sigma_{hkl}F_o) \times 100$, where F_o and F_c are the observed and calculated structure factors, respectively, for all data.
§, R_{free} calculated as for R_{cryst}, but on 4% of data excluded from refinement.
¶, B values were calculated with the CCP4 program TLSANL [Howlin B, Butler DS, Moss DS, Harris GW, Driessen HPC (1993) TLSANL: TLS parameter analysis program for segmented anisotropic refinement of macromolecular structures. *J Appl Cryst* 26:622–624.]

Table S2. Percent identity of chCD1–2 to MHC and CD1

Protein (PDB code)	Species	α1-α2	α1–α3
chCD1–1	Chicken	23	48
huCD1a (1ONQ)	Human	16	24
huCD1b (1GZQ)	Human	19	25
muCD1d (1CD1)	Mouse	17	23
chBF2*21 (3BEW)	Chicken	11	22
HLA-E (1MHE)	Human	14	19