Electronic Supplementary Material (ESI) for Molecular BioSystems.

Supplementary Material

A microscopic insight from conformational thermodynamics to functional ligand binding in proteins

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Supplementary Material:

Supplementary Tables S1-S3

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Table S1. Conformational thermodynamic changes (kJ/mol) of all the dihedrals of the metal binding loop aspartates upon Ca^{2+} binding to native apo-aLA. The ΔG_i^{conf} and $T\Delta S_i^{conf}$ in the last two columns indicate the total change for each residue by adding individual dihedral contributions.

	ΔG_{ϕ}^{conf}	$T\Delta S_{\phi}^{conf}$	ΔG_{ψ}^{conf}	$T\Delta S_{\psi}^{conf}$	$\Delta G_{\chi 1}^{conf}$	$T\Delta S_{\chi 1}^{conf}$	$\Delta G_{\chi 2}^{conf}$	$T\Delta S_{\chi 2}^{conf}$	ΔG_i^{conf}	$T\Delta S_i^{conf}$
D82	-0.75	-1.25	2.0	-0.2	-0.5	-1.1	-0.05	-1.0	0.7	-3.5
D84	0.5	0.1	-0.1	1.65	1.5	-0.2	0.25	2.75	2.15	4.3
D87	-0.4	-0.3	-0.3	-0.3	-0.25	-0.3	0.25	2.2	-0.7	1.3
D88	-0.1	-0.05	0	0	0.2	0.35	0.2	1.9	0.3	2.2

Table S2. Conformational thermodynamic changes (kJ/mol) in residues from A1 and A2 helices upon Ca^{2+}/ Mg^{2+} binding to native apo-aLA.

Residue #	Residue	ΔG_i^{conf} (Ca^{2+})	$T\Delta S_i^{conf}$ (Ca^{2+})	ΔG_i^{conf} (Mg^{2+})	$T\Delta S_i^{conf}$ (Mg^{2+})
5	LYS	1.0	6.4	0.3	-0.8
6	CYS	0.3	0.0	0.3	0.2
7	GLU	0.6	4.9	0.9	3.7
8	VAL	-0.3	-0.9	0.2	0.2
9	PHE	1.3	7.0	0.6	2.8
10	ARG	-1.5	-6.2	2.8	2.9
11	GLU	0.6	5.6	0.2	0.2
12	LEU	0.05	-1.1	-1.1	-4.9
13	LYS	0.12	-4.0	-0.8	-3.5
14	ASP	-0.6	-1.4	-2.8	-2.3
15	LEU	-0.3	-3.1	0.1	-1.6
16	LYS	-0.7	-5.3	0.3	2.3
17	GLY	0.0	-0.3	0.7	1.0
18	TYR	-0.6	-4.2	1.0	1.2
19	GLY	-0.3	-0.2	-0.5	-0.4
20	GLY	0.2	0.2	0.3	0.5
21	VAL	-0.9	-1.7	-0.4	-3.4
22	SER	-0.3	-5.0	0.0	0.0
23	LEU	-0.5	-0.5	-0.5	-3.5
24	PRO	0.1	1.1	0.9	1.3
25	GLU	-0.9	-2.0	-0.8	-4.1
26	TRP	0.9	0.1	-0.4	-0.7
27	VAL	-0.4	-3.0	-0.3	-3.3
28	CYS	0.1	0.0	0.0	-0.1
29	THR	0.0	-1.5	-0.1	-0.7
30	THR	-0.2	0.6	-1.3	-2.8
31	PHE	-0.6	-4.9	-0.4	-2.1
32	HSE	0.2	2.1	0.2	0.6
33	THR	0.1	-0.4	-0.9	-2.1
34	SER	-0.8	2.2	-0.6	-2.0

Table S3. Conformational thermodynamic changes (kJ/mol) at the interfacial residues upon Ca^{2+}/ Mg^{2+} binding to native apo-aLA

Residue #	Residue	ΔG_i^{conf} (Ca^{2+})	$T\Delta S_i^{conf}$ (Ca^{2+})	ΔG_i^{conf} (Mg^{2+})	$T\Delta S_i^{conf}$ (Mg^{2+})
50	TYR	-0.6	1.6	-1.1	-4.0
51	GLY	0.0	-0.7	0.1	-0.7
52	LEU	-0.4	-0.5	-1.1	0.4
53	PHE	0.0	1.4	-0.6	-3.2
54	GLN	-0.4	-5.7	-0.5	-3.5
55	ILE	0.2	-1.4	0.5	-0.4
56	ASN	-0.5	-5.2	0.3	2.6
57	ASN	0.0	1.5	0.0	-1.9
58	LYS	-0.7	-0.7	-1.3	-2.4
59	ILE	0.9	0.8	0.6	-0.5
60	TRP	1.4	-0.4	3.6	2.3
86	THR	-0.8	-1.7	-0.9	-1.9
87	ASP	-0.7	1.3	-1.3	-1.4
88	ASP	0.3	2.2	-0.7	-1.6
89	ILE	-0.6	-1.5	0.0	2.3
90	MET	0.0	-0.1	0.1	0.5
91	CYS	0.1	-0.1	0.0	0.0
92	VAL	-1.0	-3.6	-1.2	-5.2
93	LYS	-1.2	-2.8	-0.2	-0.1
94	LYS	-0.9	-4.0	-0.4	0.1
95	ILE	3.4	2.9	-1.0	-2.7
96	LEU	-0.5	-5.3	-0.5	-3.7
97	ASP	0.1	-0.2	-0.2	-0.7
98	LYS	-0.5	-2.8	-0.6	-1.1
99	VAL	-0.6	-2.4	-0.8	-1.4
100	GLY	-0.7	-1.1	-0.8	-0.7
101	ILE	-1.0	-5.7	-0.2	0.2
102	ASN	2.3	5.7	1.2	0.5
103	TYR	0.3	5.0	0.0	-0.1
104	TRP	-1.0	1.2	-1.9	-1.3

Figure Captions:

Fig. S1. The Ramachandran plot of simulated Mg^{2+}-aLA complex.

Fig. S2. The convergence of equilibrium dihedral distributions corresponding to five samples generated randomly from the 80-100 ns trajectory, for a few representative dihedrals in apo-, Ca^{2+}-, Mg^{2+}-aLA.

Fig. S3. The conformational thermodynamic changes (in kJ/mol) at the residues of the MBL upon metal binding are illustrated. (a) $\Delta G^{conf}_{MBL,i}$, (b) $T\Delta S^{conf}_{MBL,i}$ of Ca^{2+}-aLA (c) $\Delta G^{conf}_{MBL,i}$ and (d) $T\Delta S^{conf}_{MBL,i}$ upon Mg^{2+} binding.

Fig. S4. Representative equilibrium dihedral distributions in native-apo, Ca^{2+}- and Mg^{2+}-aLA: (a) ψ of K79. (b) ψ of D82. (c) ψ of D84. (d) ϕ of D87. (e) ψ of F80. (f) ϕ of L81.

Fig. S5. Representative equilibrium dihedral distributions in native-apo, Ca^{2+}- and Mg^{2+}-aLA: (a) χ_1 and (b) χ_2 of K79. (c) χ_1 of D82. (d) χ_2 of D84. (e) χ_2 of D87. (f) χ_1 of D88.

Fig. S6. Multiple Sequence Alignment (MSA) of aLA sequences from different species. The predicted oleic acid binding residues, H32, W60, I95 and W104 are conserved; however, I59 shows weak conservation across the sequences.

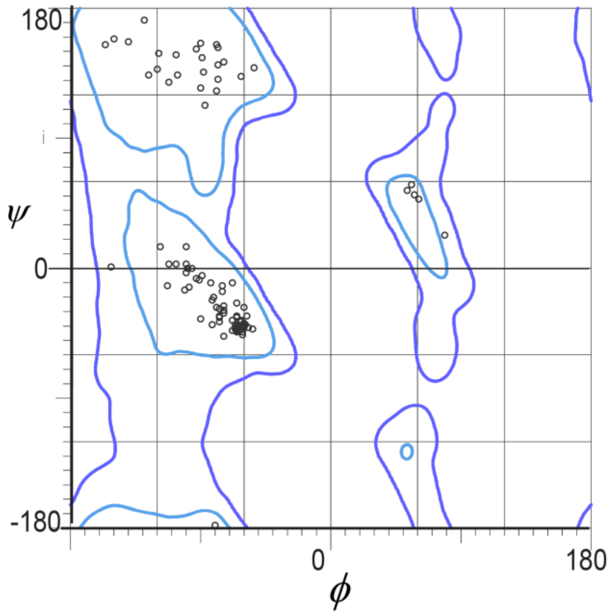

180
0
-180
ψ
0
180
ϕ

Fig S1

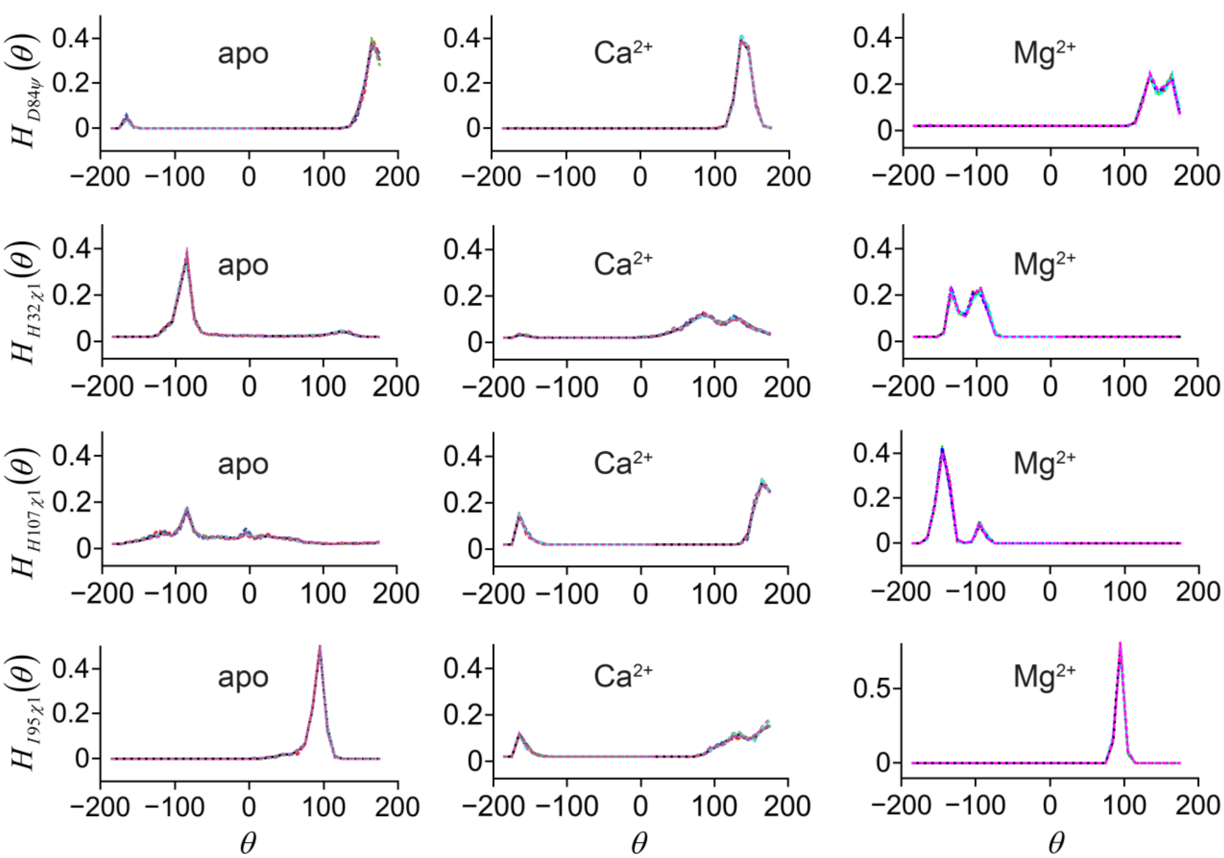
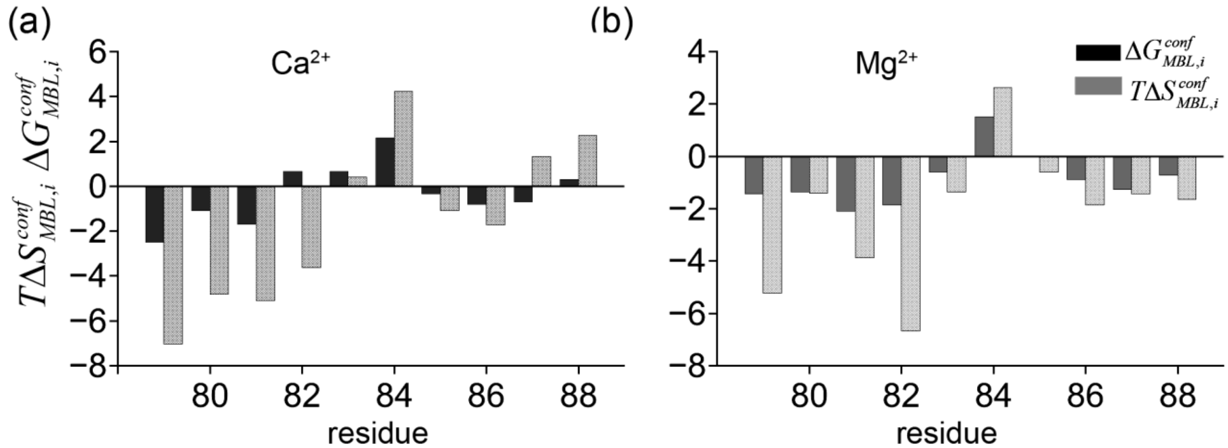

apo
Ca2+
Mg2+
$H_{D84\psi}(\theta)$
$H_{H32\chi1}(\theta)$
$H_{H107\chi1}(\theta)$
$H_{I95\chi1}(\theta)$
θ

Fig S2



(a)
(b)
Ca2+
Mg2+
$T\Delta S^{conf}_{MBL,i}$ $\Delta G^{conf}_{MBL,i}$
$\Delta G^{conf}_{MBL,i}$
$T\Delta S^{conf}_{MBL,i}$
residue
residue

Fig S3

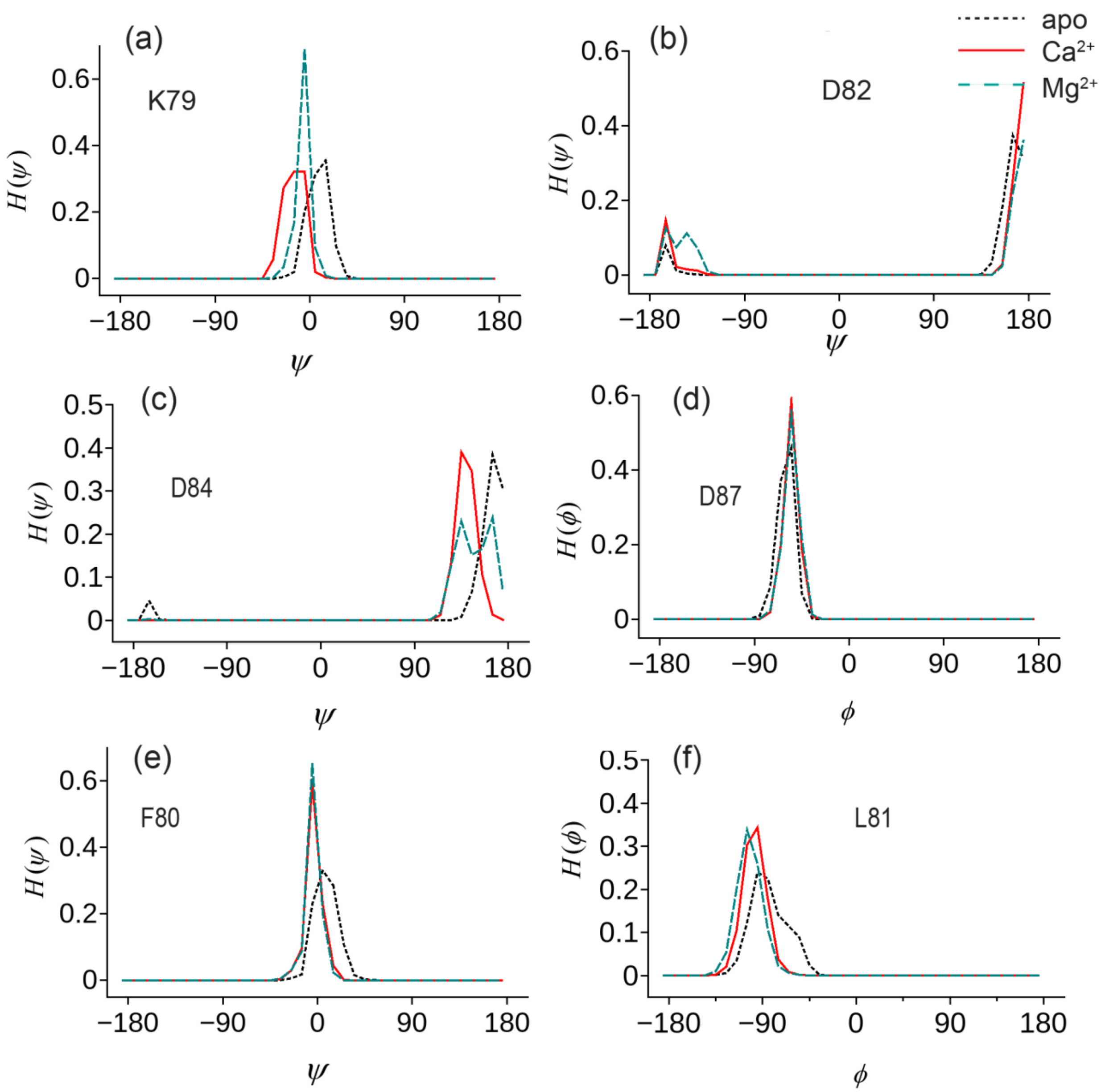

(a)
K79
0.6
0.4
0.2
0
$H(\psi)$
−180
−90
0
90
180
ψ
(b)
D82
apo
Ca^{2+}
Mg^{2+}
0.6
0.4
0.2
0
$H(\psi)$
ψ
(c)
D84
0.5
0.4
0.3
0.2
0.1
0
$H(\psi)$
ψ
(d)
D87
$H(\phi)$
ϕ
(e)
F80
$H(\psi)$
ψ
(f)
L81
$H(\phi)$
ϕ

Fig S4

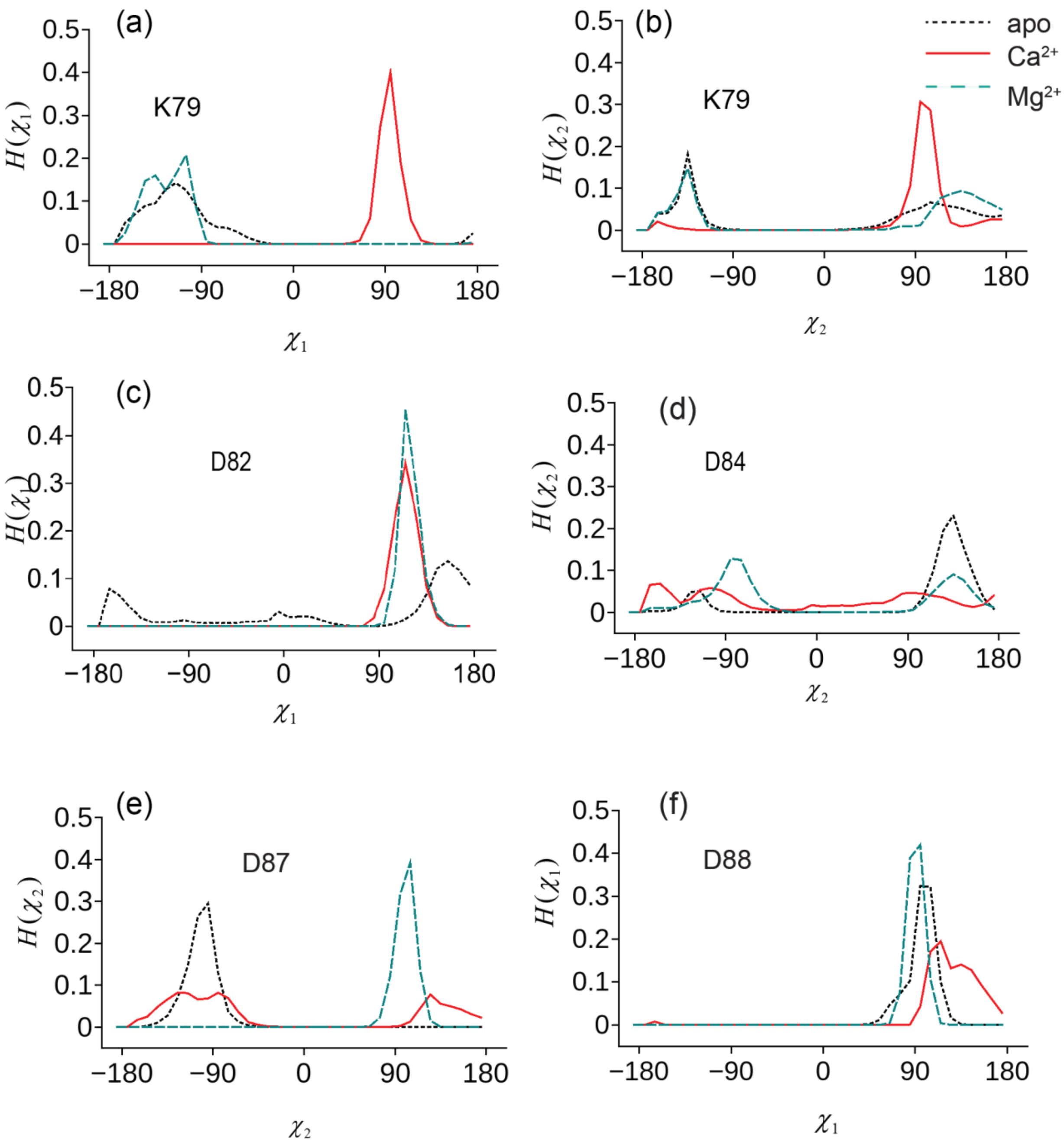

(a)
K79
$H(\chi_1)$
χ_1
(b)
K79
$H(\chi_2)$
χ_2
apo
Ca^{2+}
Mg^{2+}
(c)
D82
$H(\chi_1)$
χ_1
(d)
D84
$H(\chi_2)$
χ_2
(e)
D87
$H(\chi_2)$
χ_2
(f)
D88
$H(\chi_1)$
χ_1

Fig S5

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Bos taurus              --MMSFVSLLLVGIL-FHATQAEQLTKCEVFRELKDLKGYGGVSLPEWVCTTFHTSGYDTQAIVQNNDSTEYGLFQINNKIWCKDDQNPHSSNICNISCDKFLDDDLTDDIMCVKKILDKVGINYWLAHKALCSEKLDQWLCEKL
Bos mutus               --MMSFVSLLLVGIL-FHATQAEQLTKCEVFRELKDLKGYGGVSLPEWVCTTFHTSGYDTQAIVQNNDSTEYGLFQINNKIWCKDDQNPHSSNICNISCDKFLDDDLTDDIMCVKKILDKVGINYWLAHKALCSEKLDQWLCEKL
Bos grunniens           --MMSFVSLLLVGIL-FHATQAEQLTKCEVFRELKDLKGYGGVSLPEWVCTTFHTSGYDTQAIVQNNDSTEYGLFQINNKIWCKDDQNPHSSNICNISCDKFLDDDLTDDIMCVKKILDKVGINYWLAHKALCSEKLDQWLCEKL
Bos indicus             --MMSFVSLLLVGIL-FHATQAEQLTKCEVFRELKDLKGYGGVSLPEWVCTTFHTSGYDTQAIVQNNDSTEYGLFQINNKIWCKDDQNPHSSNICNISCDKFLDDDLTDDIMCVKKILDKVGINYWLAHKALCSEKLDQWLCERL
Bubalus bubalis         --MMSFVSLLLVGIL-FHATQAEQLTKCEVFRELKDLKDYGGVSLPEWVCTAFHTSGYDTQAIVQNNDSTEYGLFQINNKIWCKDDQNPHSSNICNISCDKFLDDDLTDDIMCVKKILDKVGINYWLAHKALCSEKLDQWLCEKL
Ovis aries              --MMSFVSLLLVGIL-FHATQAEQLTKCEVFQELKDLKDYGGVSLPEWVCTAFHTSGYDTQAIVQNNDSTEYGLFQINNKIWCKDDQNPHSRNICNISCDKFLDDDLTDDIMCVKKILDKVGINYWLAHKALCSEKLDQWLCEKL
Capra hircus            --MMSFVSLLLVGIL-FHATQAEQLTKCEVFQKLKDLKDYGGVSLPEWVCTAFHTSGYDTQAIVQNNDSTEYGLFQINNKIWCKDDQNPHSRNICNISCDKFLDDDLTDDIVCAKKILDKVGINYWLAHKALCSEKLDQWLCEKL
Cannis familiaris       --MMSFVSLLLVSIL-FPAIQAKQFTKCELSQVLKDMDGFGGIALPEWICTIFHTSGYDTQTIVNNNGGTDYGLFQISNKFWCKDDQNLQSRNICDISCDKFLDDDLTDDMICAKKILDKEGIDYWLAHKPLCSEKLEQWRCEKL
Mustelo furo            --MMSFISLLLVGIM-FPAIQAKQFTKCELSQVLKDMDGFGGIALPEWICTIFHTSGYDTQTIVNNNGSTEYGLFQINNKVWCKDNKILQSRNICNISCDKFLDDDLTDDMTCAKKILNKEGIDYWLAHKPLCSEKLEQWHCEKL
Pteropus alecto         --MMSFLSLLLVGIL-FPATQAKQFTKCELSQVLKDMDGYGGITLPEWICTIFHSSGYDTETIINNNGKREYGLFQINNKLWCRDNRKLQSRNICDISCDKFLDDDLTDDVICAKKILDSEGIDYWLAHKPLCSEKLEQWRCEKL
Sus scrofa              --MMSFVSLLVVGIL-FPAIQAKQFTKCELSQVLKDMDGYGDITLPEWICTIFHISGYDTKTIVHDNGSTEYGLFQINNKLWCRDNQ-IQSKNICGISCDKFLDDDLTDDMMCAKKILDNEGIDYWLAHKALCSEKLDQWLCEKM
Equus caballus          -KMMSFASLLLVGIL-FSATQAKQFTKCELSQVLKSMDGYKGVTLPEWICTIFHSSGYDTQTIVKNNGKTEYGLFQINNKMWCRDNQILPSRNICGISCNKFLDDDLTDDVMCAKKILDSEGIDYWLAHKPLCSEKLEQWLCEEL
Macaca fascicularis     --MRSFVPLFLVGIL-FPAIPAKQFTKCELSQLLKDIDGYGGIALPEFICTMFHTSGYDTQAIVESNGSTEYGLFQISNKLWCKSSQVPQSRNICDISCDKFLDDDITDDIMCAKKILDIKGIDYWLAHKALCTEKLEQWLCEKL
Macaca mulatta          --MRSFVPLFLVGIL-FPAIPAKQFTKCELSQLLKDIDGYGGIALPEFICTMFHTSGYDTQAIVESNGSTEYGLFQISNKLWCKSSQVPQSRNICDISCDKFLDDDITDDIMCAKKILDIKGIDYWLAHKALCTEKLEQWLCEKL
Gorilla gorilla         --MRFFVPLFLVGIL-FPAILAKQFTKCELSQLLKDIDGYGGIALPELICTMFHTSGYDTQAIVENNESTEYGLFQISNKLWCKSSQVPQSRNICDISCDKFLDDDITDDIMCAKKILDIKGIDYWLAHKALCTEKLEQWLCEKL
Pusa hispida            --MMSFVSLLLVGIM-FPAIQAKQFRKCELSQVLKDMDGFRGIALPKWICTIFHTSGYDTQTIVSNNGSTEYGLFQINNKFWCRDNQILQSRNICDISCDKFLDDDLTDDMICAKKILDKEGIDYWLAHKPLCSEKLEQWHCEKL
Homo sapiens            --MRFFVPLFLVGIL-FPAILAKQFTKCELSQLLKDIDGYGGIALPELICTMFHTSGYDTQAIVENNESTEYGLFQISNKLWCKSSQVPQSRNICDISCDKFLDDDITDDIMCAKKILDIKGIDYWLAHKALCTEKLEQWLCEKL
Myotis brandtii         --MMSFLSLLLVGIL-FPALEAKQFTKCELSQVLKDMDGYGGVTLPEWICNIFHVSGYDTQTMVSNNGKTEYGLFQINNKLWCRDNQ-IQSRNICDISCDKFLDDDLTDDMMCAKKILDSEGIDYWLPHKPLCSEKLEQWLCEKL
Ailuropoda melanoleuca  --MMFFVSLLLVGIM-CPAIQAKQFTKCELSQVLKDMDGFGGIALSEWICTIFHTSGYDTQTIVNNNGSTEYGLFQINNKFWCRDNQILQSRNICDISCDKFLDDDLTDDMICAKKILDKEGIDYWLAHKPLCSEKLEQWHCEKL
Camelus ferus           --MMSLVSLLLVGIL-FPTIQAKQFTKCKLSDELKDMNGHGGITLAEWICIIFHNSGYDTETVVSNNGNREYGLFQINNKIWCRDNENLQSRNICDISCDKFLDDDLTDDKMCAKKILDKEGIDYWLAHKPLCSEKLEQWQCEKW
Cervus elaphus          ------------------------------------------------VCTAFHTSGYDTQAIVQNDDSTEYGLFQINNKIWCKDDQNPHSSNICNISCDKFLDDDLTDDIMCVKKILDKVGINYWLAHKALCSEKLDQWLCEKL
Balaenoptera musculus   ---MSFVSLLLVGNL-FHAIQAEQLTKCEVFQRLKDLDGYGGVTLPEWVCTVFHTSGCDTQTVVNNNGSTEYGLFQINNKIWCRDNHIPHSRDICXISCDKFLDDDLTDDIMCVKKILDNV------------------------
Cavia porcellus         --MMSFFPLLLVGIL-FPAVQAKQLTKCALSHELNDLAGYRDITLPEWLCIIFHISGYDTQAIVKNSDHKEYGLFQINDKDFCESSTTVQSRNICDISCDKLLDDDLTDDIMCVKKILDIKGIDYWLAHKPLCSDKLEQWYCEAQ
Sotalia fluviatilis     --------LLLVGIL-FHAVQAEQLTKCELFQRLKDLDGYGGVTLPEWVCTVFHTSGCDTQTIVNNSDSTEYGLFQINNKIWCRDNQIPHSRDICGISCDKFLDDDLTDDIMCVKKILDNV------------------------
Heterocephalus glaber   ----------------------KQFTKCELSQNLYDIDGYGRIALPELICTMFHTSGYDTQAIVENNESTEYGLFQISNALWCKSSQSPQSRNICDITCDKFLDDDITDDIMCAKKILDIKGIDYWIAHKALCTEKLEQWLCEKE
Papio cynocephalus      --MMSPFPLLLVSIL-FSALQAKQFTKCSLSQELNDLAGYRNITLPEWICIIFHISGYDTQTIIRNNGNTEYGLFQINDKDFCDSSQNLQSRNICDISCDKFLDDDLTDDIMCAKKILDIKGIDHWLAHKPLCSDKLEQWYCKVL
Delphinus delphis       ---------LLVGIL-FHAVQAEQXTKCELFQRLKDLDGYGGVTLPEWVCTVFHTSGCDTQTIVNNNDSTEYGLFQINNKIWCRDNQIPHSRDICGISCDKFLDDDLTDDIMCVKKILDNV------------------------
Equus asinus            ----------------------KQFTKCELSQVLKSMDGYKGVTLPEWICTIFHSSGYDTQTIVKNNGKTEYGLFQINNKMWCRDNQILPSRNICGISCNKFLDDDLTDDVMCAKKILDS-------------------------
Camelus dromedarius     ----------------------KQFTKCKLSDELKDMNGHGGITLAEWICIIFHNSGYDTETVVSNNGNREYGLFQINNKIWCRDNENLQSRNICDISCDKFLDDDLTDDKMCAKKILDK-------------------------
Odobenus rosmarus       --MMSFVSLLLVSIM-FPAIQAKQFTKCELSQVLNDMDGFGGIALPEWICTVFHTSGYDTQTIVSNNGSTEYGLFQINNKFWCRDNQILQSRNICDISCDKFLDNDLTDDMICAKKILDKV------------------------
Oryctolagus cuniculus   --MMPLVPLLLVSIV-FPGIQATQLTRCELTEKLKELDGYRDISMSEWICTLFHTSGLDTKITVNNNGSTEYGIFQISDKLWCVSKQNPQSKNICDTPCENFLDDNLTDDVKCAMKILDKEGIDHWLAHKPLCSENLEQWVCKKL
Rattus norvegicus       --MMRFVPLFLACI-SLPAFQATEFTKCEVSHAIEDMDGYQGISLLEWTCVLFHTSGYDSQAIVKNNGSTEYGLFQISNRNWCKSSEFPESENICDISCDKFLDDELADDIVCAKKIVAIKGIDYWKAHKPMCSEKLEQWRCEKP
Loxodonta africana      AKMMSFVPLLLVGIL-FPAIQAKQFTKCELSQVLKDIDGYAGITLPEFTCTIFHISGYDTQTIVNNNGSTEYGLFQISNKYWCRDHQIPQSRNICDISCDKFLDDDLTDDMMCAKKILDSKGIDYW-------------------
Mus musculus            --MMHFVPLFLVCILSLPAFQATELTKCKVSHAIKDIDGYQGISLLEWACVLFHTSGYDTQAVVNDNGSTEYGLFQISDRFWCKSSEFPESENICGISCDKLLDDELDDDIACAKKILAIKGIDYWKAYKPMCSEKLEQWRCEKP
Phocoenoides phocoena   --------------------QAEQLTKCELFQRLKDLDGYGGVTLPEWVCTVFHTSGCDTQTIVNNNDSTEYGLFQINNKIWCRDNQIPHSRDICGISCDKFLDDDLTDDIM---------------------------------
Cricetulus griseus      --MMPFIPLILVCIL-FPAIQATQLTKCEVYQAMRDMDGHEGISSLEWTCIIFHSSGCDTQATVKNNGSTEYGLFQISNKHWCESSEIPESENICGISCDKFLDDDLTDDKMCAKKILAIKGIDYW-------------------
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Fig S6