

Supporting Information for:

The Next Generation of Phosphodiesterase Inhibitors: Structural Clues to Ligand and Substrate Selectivity of Phosphodiesterases

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Table A. Residue numbering for the 12 amino acids used in superimposition routines.

Residue	PDE1 1TAZ	PDE3 1SO2 1SOJ	PDE4 1PTW 1OYN 1Q9M 1RKO 1TB7 1TBB	PDE4 1MKD	PDE4 1TB5 1FOJ	PDE5 1UDT 1UDU 1UHO 1TBF 1RKP 1T9R 1T9S	PDE9 1TBM
H	223	737	160	257	234	613	252
N	224	738	161	258	235	614	253
H	227	741	164	261	238	617	256
H	263	821	200	297	274	653	292
D	264	822	201	298	275	654	293
H	267	825	204	301	278	657	296
H	296	854	233	330	307	685	325
T	334	893	271	368	345	723	363
D	370	937	318	415	392	764	402
E	391	958	339	436	413	785	423
Q	421	988	369	466	443	817	453
F	424	991	372	469	446	820	456

The numbering corresponds to that provided in the PDB files.

Table B. Sequence alignment of the catalytic domain of human PDEs.

	*h1*****	*h2****		
CN1A_HUMAN	RKTYHMGVLAYPAAVIVTLKDVKWSFDVFALNEASG---EH	173		
CN1C_HUMAN	RRTSNMVGLSYPPAVIEALKDVKWSFDVFSNLNEASG---DH	183		
CN1B_HUMAN	RRTYTSGPTYSTAVLNCLKNLDLWCFDVFSLNQAAD---DH	178		
CN3A_HUMAN	DKPILAPEPLVMNDNLDSEIMEQLNTWNFPIFDLVENIGRKCGR	709		
CN3B_HUMAN	IEQEVSLLDLILVEEYDSLIEKMSNWNPPIFELVEKMGEKSGR	694		
CN4A_HUMAN	NIPRGVKTQDQEELLAQELENLNKWGLNIFCVSDYAG---GR	389		
CN4B_HUMAN	SISRGVNTEDEDHLAKELEDLNWKGLNIFNVAGYSH---NR	362		
CN4C_HUMAN	TVPRFGVQTQDQEELLAKELEDTNWKGLDFVKVAELSG---NR	344		
CN4D_HUMAN	SIPRGVKTQEEDVLAKELEDVNWKGLHVFRIAELSG---NR	418		
CN8A_HUMAN	NIITPISLDDVVPPRIARAMENEYWDFFDIFELEAATH---NR	512		
CN8B_HUMAN	HLAMPITINDVPPCISQLLDNEESWDFNIFELEAITH---KR	571		
CN7A_HUMAN	SNSLNILDDDYNGQAKCMLEKVGWNWFIDIFLFDRLTN---GN	168		
CN7B_HUMAN	QAPLHLLDEDYLQARHMLSKVGMWDFDIFLFDRLTN---GN	129		
CN5A_HUMAN	EETRELQSLAAAVVPSAQTLKITDFSFSDFELS-----DL	569		
CN11_HUMAN	SKAEVDKFK-AANIPLVSELAIDDIHFDDFLSD-----VD	675		
CN6A_HUMAN	KEPWCEEEEELAEILQAELPDADKYEINKFHFSSDLPLT--EL	515		
CN6B_HUMAN	KEPADCDEDELGEILKEELPGPTTFDIFYEFHFSDECT--EL	514		
CN6C_HUMAN	DVIDDCEEKQLVAILKEDLPDPRSAELYEFRFSDFPLT--EH	519		
CN10_HUMAN	SYHSICTSEEWQGLMQFTLPVRLCKEIELHFHDIGPF---EN	474		
CN2A_HUMAN	HMKVSDEYTKLHDGIQPVAIDSNFASFTYTPRSLP--ED	613		
CN9A_HUMAN	TPRRDVPTYPKYLSPETIEALRKPTFDVWLWE-----PN	269		
	:	:		
	*h3*****	*h4**	*h5*****	*h6*****
CN1A_HUMAN	SLKFMIYELFTRYDLINRFKIPVSCLITFAEALEVGYSKYKNPYHNLIHAADVTVHYIM			234
CN1C_HUMAN	ALKFIFIYELLTRYDLISRFKIPIVASLVSFVEALELEVGYSKHKNPYHNLMHAADVTVHYLL			244
CN1B_HUMAN	ALRTIVFELLTRHNLISRFKIPTVFLMSFLDALETGYGKYKNPYHNQIHAADVTVHCFL			239
CN3A_HUMAN	ILSQVSYRLFEDMGLFEAFKIPIREFMNYFHALEIGYR--DIPYHNRIHATDVLHAVWYLT			768
CN3B_HUMAN	ILSQVMYTLFQDTGLLEIFKIPITQQFMNYFRALENGYR--DIPYHNRIHATDVLHAVWYLT			753
CN4A_HUMAN	SLTCIMYMFQERDLLKKFRIPVDTMTYMLTLEDHYH-ADVAYHNSLHAADVLQSTHVL			449
CN4B_HUMAN	PLTCIMYAIQFERDLLKTFRISSDTFITYMMTLEDHYH-SDVAYHNSLHAADVAQSTHVL			422
CN4C_HUMAN	PLTAIIFSIFQERDLLKTFQIPADTLATYLLMLEGHYH-ANVAYHNSLHAADVAQSTHVL			404
CN4D_HUMAN	PLTVIMHTIFQERDLLKTFKIPVDTLITYLMTLEDHYH-ADVAYHNNIHAADVQSTHVL			478
CN8A_HUMAN	PLIYLGLKMFARFGICEFLHCSESTLRSWLQIIEANYH-SSNPYHNSTSADVLHATAYFL			572
CN8B_HUMAN	PLVYLGLKVFSRGVCEFLNCSETTLRAWFQVIEANYH-SSNAYHNSTHAADVLHATAFFL			631
CN7A_HUMAN	SLVSLTFHLFSLHGLIEYFHLDMMKLRRFLVMIQEDYH-SQNPYHNAVAADVTQAMHCYL			228
CN7B_HUMAN	SLVTLCHLFNTHGLIHHFKLDMVTLHRFLVMQEDYH-SQNPYHNAVAADVTQAMHCYL			189
CN5A_HUMAN	ETALCTIRMFTDNLVQNFQMKHEVLCRWLISVKKNYRK-NVAYHNWRHAFNTAQCMFAAL			629
CN11_HUMAN	AMITAALRMFMELGMVQKFIDYETLCRLLTVRKNYR--MVLYHNWRHAFNVQCLMFAML			734
CN6A_HUMAN	ELVKCGIQMYEYLVVDFKHIPQEALVRFMYSLSKGYR--KITYHNWRHGFNVQGQTMFSLL			574
CN6B_HUMAN	DLVKCGIQMYEYLGVVRKFQIPQEVLVRLFLFSISKGYR--RITYHNWRHGFNVQGQTMFTLL			573
CN6C_HUMAN	GLIKCGIRLFFEINVVEKFVVPVEVLTRWMYTVRKGYR--AVTYHNWQHGFGNVQGQTMFTLL			578
CN10_HUMAN	MWPGIFVYMHVRSCGTSCFEL--EKLCRFIMSVKKNYR--RVPYHNWKHAVTVAHCMYAIL			531
CN2A_HUMAN	DTSMAILSMLQDMNFINNYKIDCPTLARFCLMVKKGYR--DPPYHNWMHAFSVSHFCYLLY			672
CN9A_HUMAN	EMLSCLEHMYHDLGLVRDFSINPVTLRRWLFCVHDNYR--NNPFHNFRHCFCAQMMYSMV			328
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			**	*
			AA	A

*h7***

CN1A_HUMAN	LHTG-----	-IMHWLTELEILA	250
CN1C_HUMAN	YKTG-----	-VANWLTELEIFA	260
CN1B_HUMAN	LRTG-----	-MVHCLSEIELLA	255
CN3A_HUMAN	TQPIPGLSTVINDHGSTSDDSGFTGHMGYVFSKTYNVTDDKYGCLSGNIPALELMA	828	
CN3B_HUMAN	TRPVPGLQQIHNGCGTGNETSDGRINHGRIAYISSKCSNPDESYGCLSSNIPALELMA	813	
CN4A_HUMAN	ATPA-----	-LDAVFTDLEILA	465
CN4B_HUMAN	STPA-----	-LDAVFTDLEILA	438
CN4C_HUMAN	ATPA-----	-LEAVFTDLEILA	420
CN4D_HUMAN	STPA-----	-LEAVFTDLEILA	494
CN8A_HUMAN	SKER-----	-IKETLDPIDEVA	588
CN8B_HUMAN	GKER-----	-VKGSILDQLDEVA	647
CN7A_HUMAN	KEPK-----	-LANSVTPWDILL	244
CN7B_HUMAN	KEPK-----	-LASFLTPLDIML	205
CN5A_HUMAN	KAGK-----	-IQNKLTDLIELA	645
CN11_HUMAN	TTAG-----	-FQDILTEVEILA	750
CN6A_HUMAN	VTGK-----	-LKRYFTDLEALA	590
CN6B_HUMAN	MTGK-----	-LKSYYTDLEAFA	589
CN6C_HUMAN	MTGR-----	-LKKYYTDLEAFA	594
CN10_HUMAN	QNNH-----	-TLFTDLERKG	545
CN2A_HUMAN	KNLE-----	-LTNYLEDIEIFA	688
CN9A_HUMAN	WLCS-----	-LQEKFQSQTDLI	344

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*h7**** *h8**** *h9**** *h10*****

CN1A_HUMAN	MVFAAAIHDYEHTGTTNNFHIQTRS DVA ILYNDR-SVLENHHVSAAYR LMQE-EEMN ILI	308
CN1C_HUMAN	IIFSAAIHDYEHTGTTNNFHIQTRS DP A ILYNDR-SVLENHHL SAAYR LLQDDEM NILI	319
CN1B_HUMAN	IIFAAAIHDYEHTGTTNSFHIQTKSECAIVY NDR-SVLENH HISSVFR LMQD-DEM NI FI	313
CN3A_HUMAN	LYVAAAMHDYDH PGRTNAFLVAT SAPQAVLY NDR-SVLENH HAAA A WNL FMS RPEYNFL I	887
CN3B_HUMAN	LYVAAAMHDYDH PGRTNAFLVATNAPQAVLY NDR-SVLENH HAASAWN LYLSRPEYNFL L	872
CN4A_HUMAN	ALFAAAIHDVDHPGVSNQFLINTNSEL ALM YNDE-SVLENHHL AVGF KLL QE-DNC DIF Q	523
CN4B_HUMAN	AIFAAAIHDVDHPGVSNQFLINTNSEL ALM YNDE-SVLENHHL AVGF KLL QE-EHCD IF M	496
CN4C_HUMAN	ALFASAIHDVDHPGVSNQFLINTNSEL ALM YNDA-SVLENHHL AVGF KLL QA-ENCD IF Q	478
CN4D_HUMAN	AIFASAIHDVDHPGVSNQFLINTNSEL ALM YNDS-SVLENHHL AVGF KLL QE-ENCD IF Q	552
CN8A_HUMAN	ALIAATIHDVDHPGRTNSFLCNAGSEL A ILYNDT-AVLES HHA ALA FQL T GDD K CNI FK	647
CN8B_HUMAN	ALIAATVHDVDHPGRTNSFLCNAGSEL A LVLYNDT-AVLES HHTA LA FQL TVKD TK CNI FK	706
CN7A_HUMAN	SLIAAATHDLDHPGVNQPFLIKTNH YLATL TYKNT-SVLENH HWRSAV GLL R--ESGL FS	300
CN7B_HUMAN	GLLAAAHDVDHPGVNQPFLIKTNH H LANLYQNM-SVLENH HWRSTIGMLR--ESRL LA	261
CN5A_HUMAN	LLIAALSHDLDH RG VNN SYIQRSEHPLA QLYCH--SIMEHH FDQCLMILNS-PGNQ ILS	702
CN11_HUMAN	VIVGCLCHDLDH RG TNNAFQAKSGS ALA QLYGTS-ATLEHHHF NHAVM I LQS-EGHN I FA	808
CN6A_HUMAN	MVTAAFC HDIDH RG TN NL YQM KS QNPLA KLH GS--SILER HHLEFGK TLL RD-ESLN IF Q	647
CN6B_HUMAN	MVTAGLCHDIDH RG TN NL YQM KS QNPLA KLH GS--SILER HHLEFGK FLL SE-ETLN IY Q	646
CN6C_HUMAN	MLAAAFCHDIDH RG TN NL YQM KS STSP LARL H GS--SILER HHLEY SKT LL QD-ESLN IF Q	651
CN10_HUMAN	LLIA CLCHDLDH RG FSNSY LQKFDH PLA ALYST--STMEQHH FS QTV S I L QL-EGHN I F S	602
CN2A_HUMAN	LFISCMCHDLDH RG TN NSFQV ASK SVLA ALYS SSEG SVM ERHH FAQA I AIL NT-HGC NIF D	747
CN9A_HUMAN	LMTAAICHDLDHPGYNN TYQINARTELAVRYNDI-SPLENH HCAV AFQ I LAE-PECN I F S	402

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	*h11*****	*h12*****	
CN1A_HUMAN	NLSKDDWRDLRNLVIEMLSTDMSGHFQQIKNIRNSLQQ-----		347
CN1C_HUMAN	NLSKDDWREFRTLVIEMVATDMSCHFQQIKAMKTALQQ-----		358
CN1B_HUMAN	NLTKEFVELRALVIEMVLATDMSCHFQQVKTMKTALQQ-----		352
CN3A_HUMAN	NLDHVEFKHFRFLVIEAILATDLKKHDFVAKFNGKVNDVG-----	D	931
CN3B_HUMAN	HLDHVEFKRFRFLVIEAILATDLKKHDFLAEFNAKANDVNSNGI-----	E	918
CN4A_HUMAN	NLSKRQRQSLRKVIDMVLATDMSKHMTLLADLKTMVETKVTSSG-----	VLL	572
CN4B_HUMAN	NLTKKQRQTLRKVIDMVLATDMSKHMSSLADLKTMVETKVTSSG-----	VLL	545
CN4C_HUMAN	NLSAKQRSLRRMVIDMVLATDMSKHMNLLADLKTMVETKVTSLG-----	VLL	527
CN4D_HUMAN	NLTKKQRQSLRKVIDIVLATDMSKHMNLLADLKTMVETKVTSSG-----	VLL	601
CN8A_HUMAN	NMERNDYRTLRQGIIDMVLATEMTKHFHVNVKFNNSINKPLATLEENGETDKNQEINTM		707
CN8B_HUMAN	NIDRNHYRTLRQAIIDMVLATEMTKHFHVNVKFNNSINKPM--AAEIEGSDCECNPAG--		762
CN7A_HUMAN	HPLESRQQMETOQIGALILATDISRQNEYLSLFRSHLDRG-----	DLC	343
CN7B_HUMAN	HLPKEMTQDIEQLQLSLLATDINRQNEFLTRLKAHLHNK-----	DLR	304
CN5A_HUMAN	GLSIEEYKTTLKIIKQAILATDLALYIKRGEFFELIRKN-----	QFN	745
CN11_HUMAN	NLSSKEYSDLMQLLKQSILATDLTLYFERRTEFFELVSKG-----	EYD	851
CN6A_HUMAN	NLNRRQHEHAIHMDIAIIATDLALYFKKRTMFQKIVDQSKEYSE-----	QEWTQYM	700
CN6B_HUMAN	NLNRRQHEHVIHMDIAIIATDLALYFKKRAMFQKIVDESKNYQDK-----	KSWVEYL	699
CN6C_HUMAN	NLNKRQFETVIHLFEVAAIIATDLALYFKKRTMFQKIVDACEQMTE-----	EEAIKYV	704
CN10_HUMAN	TLSSEYEQVLEITRKAAIIATDLALYFGNRQLEEMYQT-----	GSLN	645
CN2A_HUMAN	HFSRKDYQRMLDLMRDIIILATDLAHLRIFKDLQKMAEV-----	GYD	789
CN9A_HUMAN	NIPPDGFKQIRQGMITLILATDMARHAEIMDSFKEKMEN-----	FD	443
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	A		

	*h13*****	*h14*****	
CN1A_HUMAN	-PEGIDRAKTMSSLILHAADISHPAKSWKLHYRWTMALKMEEFFLQGDKEAELGLPFS-PLC	405	
CN1C_HUMAN	-PEAIEKPALKSMLHTADISHPAKAWDLHHRWTMSLLEEFFRQGDREAEGLPFPS-PLC	416	
CN1B_HUMAN	-LERIDKPALKSLLHAADISHPTKQWLVHSRWTKALMEEFFRQGDKEAELGLPFS-PLC	410	
CN3A_HUMAN	WTNENDRLLVCQMCIKLADINGPAKCKELHLQWTDGIVNEFYEQGDEEASLGLPIS-PFM	990	
CN3B_HUMAN	WSNENDRLLVCQVCIKLADINGPAKVRDLHLKWTEGIVNEFYEQGDEEANLGLPIS-PFM	977	
CN4A_HUMAN	LDNYSDRIQVLRNMVHCADLSNPTKPLELYRQWTDRIMAEFFQQGDRERERGMEIS-PMC	631	
CN4B_HUMAN	LDNYTDRIQVLRNMVHCADLSNPTKSLEYRQWTDRIMAEFFQQGDRERERGMEIS-PMC	604	
CN4C_HUMAN	LDNYSDRIQVQLQNLVHCADLSNPTKPKPLYRQWTDRIMAEFFQQGDRERESGLDIS-PMC	586	
CN4D_HUMAN	LDNYSDRIQVQLQNMVHCADLSNPTKPKQLYRQWTDRIMAEFFQQGDRERERGMEIS-PMC	660	
CN8A_HUMAN	LRTPENRTLICKRLIKCADVSNCRPLQYCIEWAARISEEEYSQTDEEKQQGLPVVMPVF	767	
CN8B_HUMAN	KNFPENQILIKRMMIKCADVANPCRPLDLCIEWAGRISSEYFAQTDEEKQQGLPVVMPVF	822	
CN7A_HUMAN	LEDTRRHLVLQMALKCADCNCPCRTWELSKQWSKVEKTYEFFHQGDIKKYHLGVS-PLC	402	
CN7B_HUMAN	LEDAQDRHFMLQIALKCADCNCPCRIEMSKQWSERVCEEFYRQGELEQKFELEIS-PLC	363	
CN5A_HUMAN	LEDPHQKELFLAMLMTACDSLAITKPWPIQQRIAELVATEFFDQGDRERKELNIEPTDLM	805	
CN11_HUMAN	WNKHNHDIFRSMLMTACDLGAVTPWEISRQVAELVTSEFFEQQGDRERLELKLTPSAIF	911	
CN6A_HUMAN	MLEQTRKEIVMAMMMTACDSLAITKPWEVQSQVALVAAEFWEQGDLERTVLQQNPIPMM	760	
CN6B_HUMAN	SLETTTRKEIVMAMMMTACDSLAITKPWEVQSKVALVAAEFWEQGDLERTVLDQQPIPMM	759	
CN6C_HUMAN	TVDPKKEIIMAMMMTACDSLAITKPWEVQSQVALMVANEFWEQGDLERTVLQQPIPMM	764	
CN10_HUMAN	LNNQSHRDRVIGLMMTACDLCsvTKLWPVTKLTANDIYAEFWAEGDEMKKLG-IQPIPMM	704	
CN2A_HUMAN	RNNKQHHRLLLCLLMTCSDLSDQTKGWKTTRKIAELIYKEFFSQGDLEKAMG-NRPMEMM	848	
CN9A_HUMAN	YSNEEHMTLLKAMILIKCCDISNEVRPMEVAEPWVDCLLEEFYFMQSDREKSEGLPVA-PFM	502	
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*h15a***** *h15b*****

CN1A_HUMAN	DRKST-MVAQSQIGFIDFIVEPTFSLLTDSTEKIVI	440
CN1C_HUMAN	DRKST-MVAQSQVGFIDFIVEPTFTVLTDMDTEKIVS	451
CN1B_HUMAN	DRTST-LVAQSQIGFIDFIVEPTFSVLTDAEKSVQ	446
CN3A_HUMAN	DRSAP-QLANLQESFISHIVGPLCNSYDSAGLMPGK	1026
CN3B_HUMAN	DRSSP-QLAKLQESFITHIVGPLCNSYDAAGLLPGQ	1012
CN4A_HUMAN	DKHTA-SVEKSQVGIFDYIVHPLWETWADLVHPD--	664
CN4B_HUMAN	DKHTA-SVEKSQVGIFDYIVHPLWETWADLVQPD--	637
CN4C_HUMAN	DKHTA-SVEKSQVGIFDYIAHPLWETWADLVHPD--	619
CN4D_HUMAN	DKHNA-SVEKSQVGIFDYIVHPLWETWADLVHPD--	693
CN8A_HUMAN	DRNTC-SIPKSQISFIDYFITDMFDAWDAFVD----	798
CN8B_HUMAN	DRNTC-SIPKSQISFIDYFITDMFDAWDAFAH----	853
CN7A_HUMAN	DRHTE-SIANIQIGFMTYLVEPLFTEWARFS-NTRL	436
CN7B_HUMAN	NQQKD-SIPSIQIGFMSYIVEPLFREWAHFTGNSTL	398
CN5A_HUMAN	NREKKNKIPSMQVGFDIAICLQLYEALTHVSED---	838
CN11_HUMAN	DRNRKDELPRQLQLEWIDSICMPLYQALVKVNVK---	944
CN6A_HUMAN	DRNKAELPKLQVGFIDFVCTFVYKEFSRFHEE--	793
CN6B_HUMAN	DRNKAELPKLQVGFIDFVCTFVYKEFSRFHEE--	792
CN6C_HUMAN	DRNKRDELPKLQVGFIDFVCTFVYKEFSRFHKE--	797
CN10_HUMAN	DRDKKDEVPPQQLGFYNAVAIPCYTTLTQILPP--	737
CN2A_HUMAN	DREKA-YIPELQISFMEMHIAMPIYKLQLQDLFPK--	880
CN9A_HUMAN	DRDKV-TKATAQIGFIKFVLIPMFETVTKLFPMV--	535

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CN1A_HUMAN	PLIEEASKAETSSYVASSSTTIVGLHIADALRRSNTKGMSMDGSYSPDYSLAAVDLKSF	499
CN1C_HUMAN	PLIDETSQTGGTGQRRSSLNSISSSDAKRSGVKTSSEGSAPIINNSVISVDYKSF----	506
CN1B_HUMAN	PLADEDSKSKNQPSFQWRQPSLDVEVGDPNPDVVSF-----	482
CN3A_HUMAN	WVEDSDESGDTDDPEEEEEEAPAPNEEETCENNESPKKKTFK-RRKI-----	1072
CN3B_HUMAN	WLEAEEDN-DTESGDDEDGEE-LDTEDEEMENNLPKPPRKSRRRI-----	1057
CN4A_HUMAN	-----	
CN4B_HUMAN	-----	
CN4C_HUMAN	-----	
CN4D_HUMAN	-----	
CN8A_HUMAN	-----	
CN8B_HUMAN	-----	
CN7A_HUMAN	-----	
CN7B_HUMAN	-----	
CN5A_HUMAN	-----	
CN11_HUMAN	-----	
CN6A_HUMAN	-----	
CN6B_HUMAN	-----	
CN6C_HUMAN	-----	
CN10_HUMAN	-----	
CN2A_HUMAN	-----	
CN9A_HUMAN	-----	

*h16*****

CN1A_HUMAN	KNNLVVDIIQQNKERWKELAAQEARTSSQKCE	530
CN1C_HUMAN	KATWTEVVHINRERWRAKVPKEEKAKKEAEE	537
CN1B_HUMAN	RSTWVKRIQENKQKWKERAASGITNQMSIDE	513
CN3A_HUMAN	YCQITQHLLQNHKMWKKVIEEEQRLAGIENQ	1103
CN3B_HUMAN	FCQLMHHLTENHKIWEIVEEECKCADGNK	1088
CN4A_HUMAN	AQEILD TLEDNRDWYYSAIRQSPSPPPEES	695
CN4B_HUMAN	AQDILD TLEDNRNWYQSMIPQSPSPPLDEQN	668
CN4C_HUMAN	AQDLLDTLEDNRREWYQSKI PRSPS DLTNPER	650
CN4D_HUMAN	AQDILD TLEDNRREWYQSTIPQSPSPAPDDPE	724
CN8A_HUMAN	LPDLMQHLDNNFKYWKG LDEM KLRNL RPPPE	829
CN8B_HUMAN	LPALM QH LA DNYKHWKTLDDLCKSLRLPSD	884
CN7A_HUMAN	SQTM LGHVGLNKA SWKG LQRE QSS SEDTDAA	467
CN7B_HUMAN	SENMLGHLAHNKAQWK SLLPRQHRSRGSSGS	429
CN5A_HUMAN	CFPLLDGCRKNRQK WQALAEQQEKMLINGES	869
CN11_HUMAN	LKPM L DSVATNR SKWEELHQKRLLA STASSS	975
CN6A_HUMAN	ITPMLDG ITNNR KEW KALADEYDAKM KVQEE	824
CN6B_HUMAN	IILPMFDRLQNNR KEW KALADEYEAKV KALEE	823
CN6C_HUMAN	ITPMLSGLQNNR VEW KSLADEYDAKM KVIEE	828
CN10_HUMAN	TEPLLKACRDNL SQWEKV IRGEETATWISSP	768
CN2A_HUMAN	AAELYERV ASNGE HWT KVSHKFTIRGIPSNN	911
CN9A_HUMAN	EEIMLQPLWESRDR YEE ELKRIDDAM KELQKK	566

The numbering scheme has been taken from the SwissProt database.

Helix numbering is intended as a guide and refers to the PDE4 structure 1PTW. Differences occur in overall structure between PDE isoforms such as helices 1, 8 and 9.

"*" indicates that the residues in that column are identical in all sequences in the alignment

" :" indicates that conserved substitutions have been observed

" ." indicates that semi-conserved substitutions are observed

"A" indicates that this residue is in the active site

The multiple sequence alignment was conducted using Clustal W (1.82) on the <http://www.ebi.ac.uk/clustalw/> web site²⁴. Modifications to the Clustal W alignment were required at the start of the sequences shown here, between helices 2 and 3, and between helices 15 and 16. In some cases further manual changes to the alignment were needed to coincide with alignments observed in the crystal structures themselves.

Table C. Residue numbers of the amino acids involved in cyclic nucleotide selectivity

Position Figure 5	PDE1 1TAZ	PDE3 1SO2 1SOJ	PDE4 1PTW 1OYN 1Q9M 1RKO 1TB7 1TBB	PDE4 1MKD	PDE4 1TB5 1FOJ	PDE5 1UDT 1UDU 1UHO 1TBF 1RKP 1T9R 1T9S	PDE9 1TBM
R1	H373	G940	N321	N418	N395	A767	N405
R2	H381	H948	Y329	Y426	Y403	Q775	A413
R3	Y222	Y736	Y159	Y256	Y233	Y612	F251
R4	T385	T952	T333	T430	T407	A779	V417
R5	L388	I955	I336	I433	I410	V782	L420
R6	Q421	Q988	Q369	Q466	Q443	Q817	Q453
R7	W496	W1072	Y406	Y503	Y480	W853	Y490
R8	P374	P941	P322	P419	P396	I768	E406

The numbering corresponds to that provided in the PDB file. The R9 position is asparagine in all PDEs except PDE9.

Table D. Amino acid identities for the amino acids involved in cyclic nucleotide selectivity.

PDE gene	R1	R2	R3	R4	R5	R7	R8
1A	H	H	Y	T	L	W	P
1B	H	H	Y	T	L	W	P
1C	H	H	Y	T	L	W	P
2A	D	T	Y	A	I	W	Q
3A	G	H	Y	T	I	W	P
3B	G	H	Y	T	I	W	P
4A	N	Y	Y	T	I	Y	P
4B	N	Y	Y	T	I	Y	P
4C	N	Y	Y	T	I	Y	P
4D	N	Y	Y	T	I	Y	P
5A	A	Q	Y	A	V	W	I
6A	A	Q	Y	A	V	W	I
6B	A	Q	Y	A	V	W	I
6C	A	Q	Y	A	V	W	I
7A	N	S	Y	S	V	W	P
7B	N	S	Y	S	V	W	P
8A	N	C	Y	A	I	W	P
8B	N	C	Y	A	I	W	P
9A	N	A	F	V	L	Y	E
10A	S	T	Y	A	I	W	V
11A	A	S	Y	A	V	W	V

R6 represents the conserved glutamine (e.g. Q369 of 1PTW) in the active site. The R9 position is asparagine in all PDEs except PDE9.

Table E Amino acid residues corresponding to the metal (M), core (Q), hydrophobic (H) and lid (L) binding regions

[1PTW]	Region	1(A,B,C)	2	3	4	5	6	7(A,B)	8	9	10	11
200 (i)	M	H	H	H	H	H	H	H	H	H	H	H
201 (i)	M	D	D	D	D	D	D	D	D	D	D	D
204 (i)	M	H	H	H	H	H	H	H	H	H	H	H
230 (i)	M	E	E	E	E	E	E	E	E	E	E	E
233 (i)	M	H	H	H	H	H	H	H	H	H	H	H
318 (i)	M	D	D	D	D	D	D	D	D	D	D	D
159 (R3)	Q	Y	Y	Y	Y	Y	Y	Y	F	Y	Y	Y
321 (R1)	Q	H	D	G	N	A	A	N	N	S	A	
329 (R2)	Q	H	T	H	Y	Q	Q	S	C	A	T	S
336 (R5)	Q + H	L	I	I	I	V	V	V	I	L	I	V
339 (i)	Q	E	E	E	E	E	E	E	E	E	E	E
369 (R6) (i)	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q	Q
372	Q	F	F	F	F	F	F	F	F	F	F	W
406 (R7)	Q	W	W	W	Y	W	W	W	W	Y	W	W
337	H	M,L,M	Y	V	M	A	A	T,C	S	L	Y	T
340	H	F	F	F	F	F	F	F	Y	Y	F	F
357	H	L (ii)	M	F	M	L	M	L	V	F	M	I
209	L	N	N	N	N	N	N	Q	N	N	N	N
210	L	N,N,S	S	A	Q	S	L	P	S	T	S	A
211	L	F	F	F	Y	Y	F	F	Y	Y	Y	Y
368	L	S	L	L	S	M	L	I	S	A	G	L
371	L	G	S	S	G	G	G	G	S	G	G	E
375	L	F	H	H	Y	A	F	Y	Y	F	A	S
322 (R8)	(iii)	P	Q	P	P	I	I	P	P	E	V	V
333 (R4)	(iv)	T	A	T	T	A	A	S	A	V	A	A

The R9 position is asparagine in all PDEs except PDE9.

(i) indicates an invariant residue.

(ii) While a list of amino acids have been compiled for this hydrophobic region position, this amino acid falls in a loop region. Each of the PDEs have differing lengths for this loop and thus in a 3 dimensional sense it may be difficult to make comparisons. Having said that, PDEs 3, 4, 5 and 9 overlay quite well while PDE1 is distorted at this point. Any future homology modelling of this region should take care in addressing this loop region.

(iii) no region assigned to this residue (see R8, Figure 5)

(iv) no region assigned to this residue (see R4, Figure 5)