

# Supplemental Materials

*Molecular Biology of the Cell*

Dou et al.

**Supplemental Materials for:**

**Type II transmembrane domain hydrophobicity dictates the co-translational dependence for inversion**

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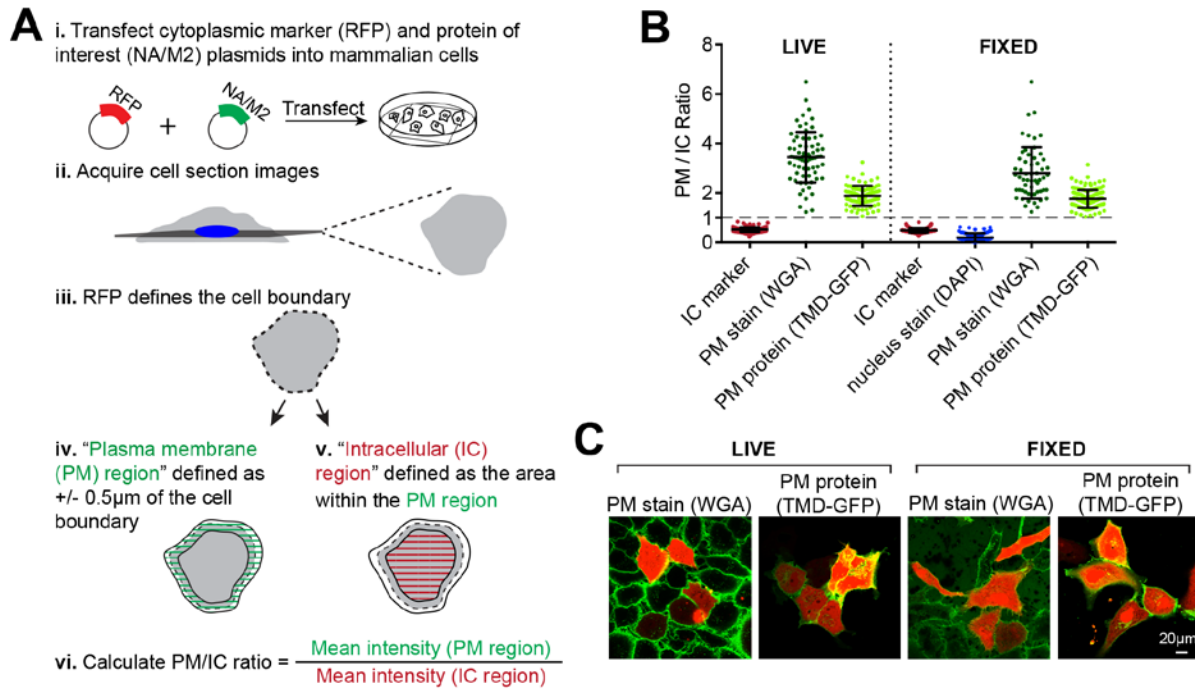
### **Single-cell approach to monitor plasma membrane localization**

To monitor the plasma membrane (PM) localization of a protein in an unbiased manner, we established a semi-automated approach that compares the mean intensity of a substrate at the PM to the intracellular (IC) region at the single-cell level (Supplemental Fig. 1A). As a control for the approach, Vero cells were co-transfected with plasmids expressing GFP with an N-terminal TMD (TMD-GFP) and RFP for a cellular marker (step -i-). At 48 h post-transfection, live and fixed cells were prepared for confocal microscopy and multiple cell section images were taken (step -ii-). Each image was then processed using Cell profiler 2.0 software (Kamentsky et al., 2011) with the following parameters. The RFP signal identified each 'cell' boundary  $\geq 350 \mu\text{m}^2$ , the lower edge of the normal cell size distribution (step -iii-). The PM region was defined as the peripheral area  $0.5 \mu\text{m}$  in-and-out from the RFP boundary (step -iv-). The intracellular (IC) region was defined as the area surrounded by the PM region (step -v-). The ratio of the mean fluorescent signal for the PM and IC regions were then calculated for each cell (step -vi-).

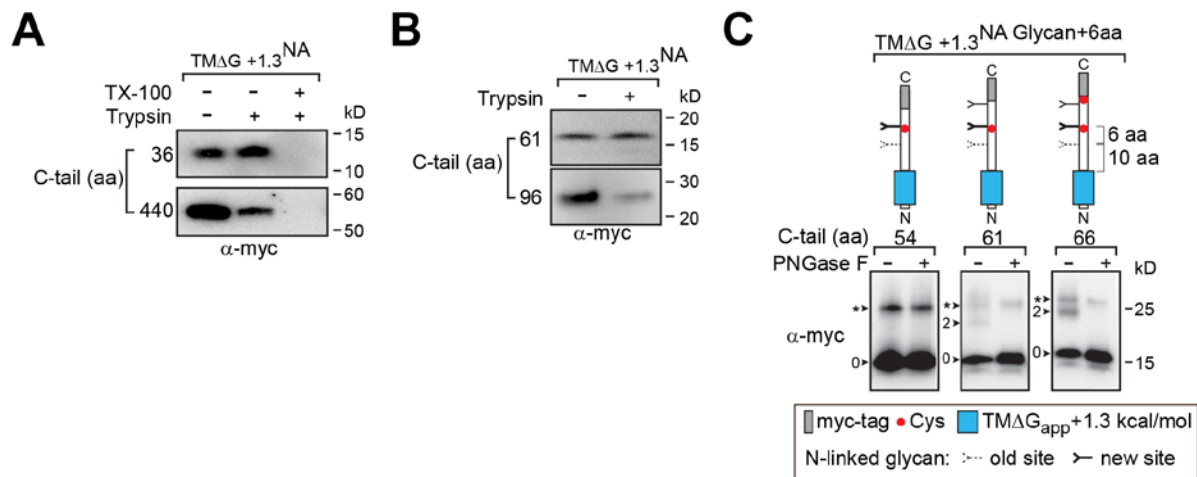
The signal from the RFP and the DAPI stained nucleus were used to determine the threshold IC value, and the signal from wheat germ agglutinin (WGA) and the protein marker TMD-GFP were used to establish the PM values. Analysis of the different cell populations gave average PM/IC ratios of  $\sim 0.5$  for the IC localized targets and the ratios for the individual cells were all  $< 1$  (Supplemental Fig. 1B and C). For the PM controls WGA and TMD-GFP, the average PM/IC ratios were  $\sim 3$  and  $\sim 2$ , respectively for 100-200 cells, and all the individual cell PM/IC ratios were  $> 1$ . Both live and fixed cells showed cell-to-cell variation in the PM/IC ratios, but this was most notable for WGA, which was extremely sensitive to the image acquisition time after fixation or the staining time for the live cell analysis. Based on these results, PM localized substrates should have a PM/IC ratio  $> 1$  and IC localized substrates should have a ratio  $< 1$  using this approach.

### **SUPPLEMENTAL MATERIALS REFERENCE**

Kamentsky, L., Jones, T.R., Fraser, A., Bray, M.A., Logan, D.J., Madden, K.L., Ljosa, V., Rueden, C., Eliceiri, K.W., and Carpenter, A.E. (2011). Improved structure, function and compatibility for CellProfiler: modular high-throughput image analysis software. *Bioinformatics* 27, 1179-1180.



**SUPPLEMENTAL FIGURE 1: Single-cell microscopy assay to monitor PM localization.** (A) Outline of the experimental approach. Cells were transfected with a cytoplasmic marker (RFP) and the TMD construct of interest, and image sections were obtained by confocal microscopy. The images were then processed using Cell profiler 2.0 where RFP defines the cell boundary. The mean intensity of the TMD construct in the plasma membrane (PM) region, defined as 0.5 µm in-and-out from the cell boundary, was calculated. Finally, the PM mean intensity was normalized to that of the intracellular region (IC), defined as the area surrounded by the PM region. (B) The PM/IC values obtained from fixed and live cell controls are displayed with the IC marker RFP. (C) Representative images of fixed and live cell sections are shown for samples where the PM was labeled with WGA, or TMD-GFP, and the IC region with RFP.



**SUPPLEMENTAL FIGURE 2: NA trypsin protection and glycosylation analysis of constructs with a shifted glycosylation site.** (A) Immunoblots of lysates obtained from cells expressing the indicated constructs. Prior to harvesting, the cells were incubated in the absence and presence of trypsin to determine if the myc epitope was properly positioned on the extracellular side of the plasma membrane. The digestion was performed on detergent (TX-100) solubilized cells to confirm the epitope is sensitive to trypsin. Note the decrease in the signal on the full-length construct with the 440 aa C-tail, but not with the 36 aa C-tail. (B) Similar analysis using the PM localized, unglycosylated, NA construct with a 61 aa C-tail and the glycosylated construct with a 96 aa C-tail. The glycosylated construct (C-tail 96 aa) is properly orientated and trypsin sensitive, whereas the unglycosylated construct (C-tail 61 aa) is not properly inverted even though it localizes to the PM. (C) Glycosylation site positioning is not responsible for the low glycosylation efficiency of the NA constructs with a marginally hydrophobic TMD. Transfected cell lysates expressing the indicated constructs with the natural glycosylation site moved 6 aa further from the membrane were either untreated, or treated, with PNGase F, separated by SDS-PAGE, and immunoblotted. The number of N-linked glycans is indicated, and the SDS resistant TMD dimer is depicted with an asterisk.



sp Q5VSG8 MANEL_HUMAN	457	27	428	RRRRRACIALFLVLLFAFGTLMGLRTL	-3.88
sp Q9UL52 TM11E_HUMAN	423	27	378	WVIGLVFISLIVLAVCIGLTVHYVRY	-3.88
sp O14638 ENPP3_HUMAN	875	31	827	LKKYKIACIVLLALLVMSLGLGLGLRKL	-3.87
sp Q92935 EXTL1_HUMAN	676	25	642	LWLALSASVLLVLLGGFSLRLAL	-3.87
sp Q9H4F1 SIA7D_HUMAN	302	33	264	RLVLILCSVVFSAVYLLCCWAGLPLCLATCL	-3.86
sp Q9UH99 SUN2_HUMAN	717	25	463	LKTFWFLPLLLLTLCTYGAWYFY	-3.86
sp Q9UIQ6 LCAP_HUMAN	1025	29	889	ARTMVVCAFVIVAVSVIMVYLLPRCTF	-3.85
sp Q9H3R1 NDST4_HUMAN	872	28	838	LRRSFRTLIVLATFCLVSVISAYFLY	-3.83
sp Q96JF0 SIAT2_HUMAN	529	24	498	WRQRMLFGIFAWGLLFLFIYFT	-3.83
sp Q9NRS4 TMPS4_HUMAN	437	32	374	VGPIIIALLSLASIIIVVLIKVILDKYYFL	-3.75
sp Q9H0X4 ITFG3_HUMAN	552	27	480	RCRAAAFFLSLFLCLFVVFVSVFVPC	-3.73
sp Q5KU26 COL12_HUMAN	742	29	680	WALKFSIILLYLCALLTITVAILGYKVV	-3.71
sp Q6ZMR5 TM11A_HUMAN	421	30	377	LKPWMAVILVLSLTVAVTIGLLVHFLVF	-3.71
sp O60476 MA1A2_HUMAN	641	28	584	TLRLSEKIFILLILSAFITLFCGAFFFL	-3.68
sp Q07837 SLC31_HUMAN	685	28	570	ILFWLTVASVVLVIAATIIAIALSPKCL	-3.68
sp Q6P4E1 CAS4_HUMAN	433	28	398	RRAGRLPSLVLLVLLVVIVLAFNYWSI	-3.64
sp Q16842 SIA4B_HUMAN	350	23	323	LRVWFSLVAFLLVFIMSLFFTY	-3.64
sp Q9NQX7 ITM2C_HUMAN	267	28	182	VCYLSMGMVLLMGLVFAVSVIYRYFFL	-3.63
sp Q96E93 KLRG1_HUMAN	195	25	134	CLVAIALGLLTVALLVLLYQWILC	-3.63
sp Q00973 B4GN1_HUMAN	533	28	505	MWLGRRALCALVLLACASLGLLYASTR	-3.62
sp Q6ZS10 CL17A_HUMAN	378	26	185	RWMVYVCLLVVTLFLGCLGLVTLI	-3.62
sp Q2HXU8 CL12B_HUMAN	276	28	211	IWRHAALGLVTLCLMLLIGLVTGMMFL	-3.61
sp A0FJZ3 GLXLT2_HUMAN	443	25	416	LRSKAAALLLALAALLLALLSLRA	-3.6
sp Q43508 TNF12_HUMAN	249	33	199	TALLVPLALGLGLACLGLLAVVLSGRASL	-3.59
sp Q04609 FOLH1_HUMAN	750	25	707	RWLACAGALVLAGGFLLGFLGFWFI	-3.58
sp P51164 ATP4B_HUMAN	291	30	228	LSRWVWISLYYAFYVVMTGTFALCLYVLM	-3.56
sp Q9C0J1 B3GN4_HUMAN	378	28	325	MLCRLCWLVSYSLAVLLGCLFLRKA	-3.55
sp Q8N6G5 CGAT2_HUMAN	542	23	510	TRTWLHLLGLALLCSLFLFMYLL	-3.55
sp Q8N3T1 GLT15_HUMAN	639	22	606	CRLOFLLLLMLGCVLMMVAML	-3.55
sp Q95502 NPTXR_HUMAN	500	28	470	FLAVLLAAGMLAFLGAVICIASVPLAA	-3.55
sp P16442 BGAT_HUMAN	354	22	317	ALRPMILFLIMLVLFVGYGV	-3.54
sp Q9UJ37 SIA7B_HUMAN	374	24	343	FWWLLLLTAACSGLLFALYFSAV	-3.54
sp Q9Y673 ALG5_HUMAN	324	29	292	LLLQAVLGAALAAALVLSIVAFMTAT	-3.53
sp Q9NR71 ASA2_HUMAN	780	26	746	LETFLFLLVMSAITVALLSLLFIT	-3.51
sp P42857 NSG1_HUMAN	185	27	79	RFKVSVLVLFALFLTCVFLVYKYVY	-3.49
sp P28907 CD38_HUMAN	300	24	253	LCLGSVLLVLLVAVVPRWR	-3.46
sp P33908 MA1A1_HUMAN	653	29	591	AALRLTEKFLVLLVFSAFITLFCGAIFFL	-3.45
sp Q8N1L4 CP422_HUMAN	340	25	307	LMAHPLLILLCMSLLLFQVIRLY	-3.44
sp Q86W10 CP421_HUMAN	505	25	472	LMAHPLLILLCMSLLLFQVIRLY	-3.44
sp Q43529 CHSTA_HUMAN	356	25	327	WLLLAACVWFVFMFVASKFITLTF	-3.41
sp P32971 TNFL8_HUMAN	234	25	172	YFLTTLTALCLVFTVATIMVLV	-3.41
sp P26572 MGAT1_HUMAN	445	25	414	AGLVWGALFVAVNALLLFFWTR	-3.4
sp Q96FL9 GLT14_HUMAN	552	25	525	RLTRRLVLPVFGVLWITVFFWVT	-3.38
sp P06734 FCER2_HUMAN	321	30	276	RCRRGTQVLLGLVTAALWAGLLTLLLLW	-3.37
sp Q49576 MMEL1_HUMAN	779	25	731	FLEGGLLLLLVTAALVALGVLYA	-3.37
sp Q96AG4 LRC59_HUMAN	307	27	36	WAVLKLHLLLVFVAGVGLVACRVTEL	-3.34
sp P78380 OLR1_HUMAN	273	32	213	FLYSPWGLAAATGLVCLGLVVTIMVGMQL	-3.34
sp P05026 AT1B1_HUMAN	303	30	242	WFKILLFYVIFYGCLAGIFGTQVMLLTI	-3.33
sp Q6UXB4 CLC4G_HUMAN	293	27	240	WSRRPLFALAVLVTTLVAVLISILL	-3.31
sp P36269 GGT5_HUMAN	586	31	555	MARGYGATVSLVLLGLGLAVIVLAVLSR	-3.31
sp Q8NF37 PCAT1_HUMAN	534	30	457	LMTLTLFPVRLVAAAMMLLAWPLAVASL	-3.31
sp Q11128 FUT5_HUMAN	374	26	338	WLWRRCLAGLLFOLLVAVCFYSYLRV	-3.28
sp O75900 MMP23_HUMAN	390	25	348	LLRVLGAALVALCCLPALVLLARLGA	-3.28
sp Q06643 TNFC_HUMAN	244	29	197	LLAVAGATSLVTLVAVPITVAVLAVLV	-3.27
sp P26718 NKG2D_HUMAN	216	27	137	FFFCFIAVAMGIRFIMVAIVSAVFL	-3.26
sp Q95672 ECEL1_HUMAN	775	26	690	VCLLSGLVFAAGLCAILAAMLALKYL	-3.25
sp P42167 LAP2B_HUMAN	454	22	20	VWIKILLFVVAVFLVLYQAM	-3.23
sp P08473 NEP_HUMAN	750	30	698	WRTPLEISLVLVLLTIAVTMIALYATY	-3.22
sp Q96MM7 H6S2_HUMAN	605	20	432	LLLAVMLFLFAVIVLQYVC	-3.2
sp O75063 XYLK_HUMAN	409	23	384	LKQRVLLAILVIFITKVFLLI	-3.2
sp O15466 SIA8E_HUMAN	376	26	339	LLGSRLLTFIFICAFALVTLQOILY	-3.17
sp P26715 NKG2A_HUMAN	233	28	135	LIVGILGICLIMASVTVIVIPSTLI	-3.16
sp Q9Y3Q0 NALD2_HUMAN	740	28	706	RLYLWMCALAAALASFLMGFMGWFIKPL	-3.14
sp Q6L9W6 B4GN3_HUMAN	998	28	953	LLRRFRLLLALAVVSVGLWTLVLELVA	-3.13
sp Q6ZWK6 TM11F_HUMAN	438	30	385	FWDSVRLALFTLAVIAGIAGIVTHFV	-3.13
sp Q9NR34 MA1C1_HUMAN	630	27	587	LRLPKQFLFLSLGLVTLFCGALFL	-3.07
sp Q9UP52 TFR2_HUMAN	801	27	696	RRAAPYLVLLALIFTGAFLLGYVAFR	-3.06
sp Q8IUN9 CLC10_HUMAN	316	20	256	LLLGLGLLHLLVIGVGF	-3.02
sp Q43736 TM2A_HUMAN	263	28	184	RCMLTLGLSFLIAGLIVGGACIYKYFM	-2.98
sp Q9UJ71 CLC4K_HUMAN	328	26	260	TVRAALICLTLVAVSLLQAVLYPRFM	-2.96
sp Q92838 EDA_HUMAN	391	26	326	FLGFFGFLSLALHLLTCCYLELR	-2.96
sp Q9Y2C3 B3GT5_HUMAN	310	23	282	MLRMVYCLLVGALCLYFMSYSL	-2.91
sp Q6NSJ0 K1161_HUMAN	714	24	634	LIGSAVLGULLVLAAVVAVCYYSV	-2.91
sp P01375 TNFA_HUMAN	233	24	182	RRCLFLSLFSLVAVAGATTLFCLL	-2.9
sp Q6LXN8 CLC9A_HUMAN	241	28	182	ACCLVMVISCVFCMGLLTAISIFGVKLL	-2.89
sp O60909 B4GT2_HUMAN	372	29	336	TLERVCKAVLLCCLHFLVAVILYFDVYA	-2.87
sp Q93063 EXT2_HUMAN	718	24	672	RIYYITLFSIVLLGIATGMFQFV	-2.85
sp Q43173 SIA8C_HUMAN	380	28	347	MARVASVLGLVMSVALLLILSIVSL	-2.84
sp Q6EIG7 CLC6A_HUMAN	209	33	162	WLSRLRWSVAGISIALSACFIVSVCVYHYFTY	-2.82
sp Q43825 B3GT2_HUMAN	422	22	379	LFRTHLIGVLSLVLFAMFLFF	-2.81
sp P02786 TFR1_HUMAN	760	24	671	ICYGTIAVIVFFLGFIMGYLYGC	-2.81
sp Q8WXI8 CLC4D_HUMAN	215	25	173	LIPSVIAVVFILLSVCFIASCLVT	-2.77
sp Q07108 CD69_HUMAN	199	24	135	VLCAMNVVFTITLIALIALSVG	-2.76
sp P21217 FUT3_HUMAN	361	24	325	WRRCLAALLFOLLVAVCFYSYLRV	-2.75
sp Q9ULY5 CLC4E_HUMAN	219	29	170	MFLWTVAGIPIFLSACFITRCVVFTRIF	-2.74
sp O60344 ECE2_HUMAN	883	27	680	LELVLAGASLLAALLGCLVALGVQY	-2.73
sp Q9NXG6 P4HTM_HUMAN	502	28	420	ICSRAYFLVLMVVFHLYGNVLALLLV	-2.73
sp Q9NP25 B3GA2_HUMAN	323	26	294	ALFTRFFILLPVILVIMLDVDTTR	-2.72
sp Q6IAN0 DRS7B_HUMAN	325	28	280	FITSTAILPLFGCLGVGLFRLLQWVR	-2.7
sp Q3V5L5 MGTS5_HUMAN	792	26	751	LVTLRPPRFLVIGIGFTLCLFMTSL	-2.67
sp Q9NUM4 T106B_HUMAN	274	25	157	RTKLYMVASVFCVLLSGLAVFFLF	-2.67
sp Q13061 TRDN_HUMAN	729	24	658	AAWLLVIALIITVSAVAIVMFDLV	-2.67
sp Q9HCO5 GALT9_HUMAN	603	24	573	IRTLTVNIVFVGVILFVSYCR	-2.65
sp Q76KP1 B4GN4_HUMAN	1039	19	1008	MKLLLLLLLLSCAAWLTYY	-2.64
sp P07306 ASGR1_HUMAN	291	22	230	RILLSLGLSLLLVVVCVIGS	-2.62
sp Q7L1S5 CHST9_HUMAN	443	23	408	VFLSVLIFVAGLHLLFMVLOVWI	-2.59
sp Q7Z7M9 GALT5_HUMAN	940	29	905	FFRSGRVLAFIFAVSVIILLFDMAALRL	-2.59
sp Q43557 TNF14_HUMAN	240	27	179	VARVGLGLLLLLMGAGLAVQGWFLQL	-2.58
sp Q95150 TNF15_HUMAN	251	24	195	ARWALTCLVLPFLAGLTYLLV	-2.58
sp Q9H2S6 TNMD_HUMAN	317	22	267	LKICGLVFGILALTLVILVFGS	-2.58
sp P07307 ASGR2_HUMAN	311	29	234	LAQRCLSMVCFSLALSFNILLVVICVT	-2.57
sp Q9Y644 RFNG_HUMAN	331	23	302	ALCRACLALAAALALLPLPL	-2.57
sp O15393 TMPS2_HUMAN	492	26	383	ALCITLTLGTFVGAALAGLLWKFM	-2.57
sp Q96SE0 ABHD1_HUMAN	405	26	368	TWDTFSLLLAVALYLYGYWACVL	-2.56
sp O75752 B3GL1_HUMAN	331	24	293	LRSLKWSLLLLSLLSFFVMWVYLSL	-2.55

sp Q11206 SIA4C_HUMAN	333	20	308	RWKLAMLALVLMVWYSI	-2.55
sp Q8NFS9 GNT2C_HUMAN	402	23	377	FWRYCFATLLSVIVFRFYSS	-2.54
sp Q9Y275 TN13B_HUMAN	285	26	213	LLAATLLALLSCCLTVVSYQVAAL	-2.53
sp Q6UVW9 CLC2A_HUMAN	174	24	125	WKIGLMCLFSIIITVCIIMATW	-2.51
sp Q14435 GALT3_HUMAN	633	20	594	FWKLGAVFFFIIVLVMQR	-2.51
sp Q58DX5 NADL2_HUMAN	795	22	649	VLKILGTATILFIFIGLIGYV	-2.5
sp Q11201 SIA4A_HUMAN	340	20	314	RTLKVLTLVFLFSLTFFL	-2.5
sp Q95803 NDST3_HUMAN	873	25	838	FORTVILLATFCMVSIISAYLLYS	-2.48
sp Q09327 MGAT3_HUMAN	533	27	504	MRRYKFLMFCMAGLCLISLHFFKTL	-2.46
sp P19526 FUT1_HUMAN	365	19	338	LCLAFLLVCVLSVIFLHI	-2.45
sp Q8NES3 LFNG_HUMAN	379	23	353	RCGRRLLLALAGALLACLLVLT	-2.45
sp Q9H2A9 CHST8_HUMAN	424	25	393	TMRLACMFSSILLFGAAGLLFISL	-2.43
sp P05981 HEPS_HUMAN	417	28	372	VAALTAGTLLLLTAIGASWAIVALLR	-2.43
sp Q6IS24 GLTL3_HUMAN	598	26	569	LRRVKVLVNLNIAVAGFVFLAKCR	-2.42
sp Q9Y662 HS3SB_HUMAN	390	24	338	VRRKLALLFAMLCVWLYMFLYSCA	-2.42
sp Q94901 SUN1_HUMAN	812	30	476	FLLTRCLRNICKFLVLIPLFLLAGLSLR	-2.42
sp P42892 ECE1_HUMAN	770	24	679	RLVVVLVLAAGLVAGLAALGIQY	-2.39
sp Q96L58 B3GT6_HUMAN	329	22	295	LLRRWRRRRAALGLTLALCGAALLYLARCAA	-2.36
sp Q8IXA5 SACA3_HUMAN	215	23	133	LRRRWCPAGIMLLALVCLLSCLL	-2.34
sp Q92186 SIA8B_HUMAN	375	22	349	FRSWMLAALLTLLVFLIFADIS	-2.34
sp P51993 FUT6_HUMAN	359	26	323	WSWRCCLTLLFQLLMAVCFSSYLRV	-2.33
sp Q12918 KLRB1_HUMAN	225	28	159	FALKSCAGIILLVVTGLSVSVTSLI	-2.3
sp Q9UEW3 MARCO_HUMAN	520	32	443	LAVVVIYLLITAGAGLLVQVQLNQAARLVL	-2.29
sp Q12797 ASPH_HUMAN	758	27	680	TSFFTFWFIALLGVWTSVAVWFDLV	-2.28
sp Q43286 B4GT5_HUMAN	388	29	353	LLRPRRSLAALFFSLSSSLLYFYVA	-2.28
sp Q9H1B5 XYLT2_HUMAN	865	29	827	LVRRYKLAIAIALALLLQGLVWWSFGL	-2.27
sp Q6ICH7 ASPH2_HUMAN	369	19	293	TTAVITVACLLVLFVWYCY	-2.26
sp Q86SF2 GALT7_HUMAN	657	28	628	RLKIGFILRSLLVGSGFLGVVWSSLT	-2.22
sp Q86SR1 GLT10_HUMAN	603	26	571	RLQOVALVLAALVLLPNVGLWALYR	-2.22
sp Q9H3S3 TMP55_HUMAN	457	29	384	MRRGCVALGALGLAGAGVGSWLLVLYLC	-2.22
sp P00461 GOL4_HUMAN	696	25	660	RIFQTLTLLTVFGLFYGAMLYYEL	-2.2
sp Q9BYC5 FUT8_HUMAN	575	19	550	SWRWIMLILFAWGTLFYI	-2.19
sp P29965 CD40L_HUMAN	261	28	211	IFMYLLTVLITQMIGSALFVYLHRRRL	-2.18
sp Q9H1C3 GL8D2_HUMAN	349	25	322	LLRKINQVLLFLVILTCVILYKVV	-2.18
sp Q8IXK2 GLT12_HUMAN	581	26	542	LRRGREALVLLALLALAGLSVLR	-2.18
sp Q5H943 KKLC1_HUMAN	113	22	89	FYLLASSILCALIVFWKYRRF	-2.16
sp Q5U4P2 ASPH1_HUMAN	390	33	292	LIMLPWPLPASSALLTLLFGALTSFLWYCYRL	-2.14
sp Q8IUS5 EPHX4_HUMAN	362	33	319	LMLTRSLFVSLVYCYGCLCASIHLLKLLWSL	-2.09
sp Q16706 MA2A1_HUMAN	1144	20	1118	FTVFGSAIFCVIFSLYMLM	-2.09
sp Q9NSB4 CHST7_HUMAN	486	22	454	YCKFALLLVYTLVLLVPSVL	-2.07
sp P52849 NDST2_HUMAN	843	25	845	LELHRLLIILLIIFSLGSMGFLAYVV	-2.07
sp P78562 PHEX_HUMAN	789	24	709	RGTRIALVVFVGGTLVGLTFLV	-2.07
sp Q8NSD6 GBGT1_HUMAN	347	25	320	RRRLALGLGFCLLAGTSLVSLVWYL	-2.04
sp Q4G148 GXL1_HUMAN	440	23	417	MRRYLRVVLVACVAGFCSLLYAF	-2.04
sp Q9BUJ0 ABHEA_HUMAN	271	19	219	QVALLGLSLLMLLVLVYGL	-2.02
sp Q9UBM8 MG24C_HUMAN	478	24	435	LRKRSTVSVFLGVVIFLLFMNLYI	-2.02
sp Q10472 GALT1_HUMAN	559	25	531	FAYCKVVLATSLVWLLDMFLLYF	-2.01
sp P00587 MFNG_HUMAN	321	26	293	CRLPRGLGALLTLLCMGLLCLRYHL	-2.01
sp Q969X2 SIA7F_HUMAN	333	19	273	RSVAVFVILFALITILILYS	-2.01
sp Q9H2X3 CLC4M_HUMAN	399	21	329	ALVLQLLSFMLLAGVLAIVL	-1.99
sp P27487 DPP4_HUMAN	766	24	738	WKVLLGLLGAAALVITITVPVLL	-1.99
sp Q9NSC7 SIA7A_HUMAN	600	21	565	VQVSLLLAVLFFLFFALPFSFI	-1.98
sp Q5T4J0 GCNT6_HUMAN	391	21	368	WVSCYCIITLQIIIIVTVLY	-1.97
sp Q8NBJ4 GOLM1_HUMAN	401	20	366	LVLAALVACIIVLGFNYWIA	-1.96
sp Q94923 GLCE_HUMAN	617	19	589	YKTLIICALFTLVTLVLLW	-1.95
sp Q8NDV1 SIA7C_HUMAN	305	25	277	ILKRKSVIAVSFIAFLFLLVVRV	-1.94
sp Q69513 B4GT4_HUMAN	344	22	314	LSYKFRLLLLTCLTVVGGWAT	-1.93
sp Q7Z410 TMS9_HUMAN	1059	28	1009	AACCRASIGVATSLVLLTGLVLLAF	-1.92
sp P26717 NKG2C_HUMAN	231	27	134	LTAEVLGIICVIMATVKTIVLIPFL	-1.88
sp Q07444 NKG2E_HUMAN	240	27	143	LTAEVLGIICVIMATVKTIVLIPFL	-1.88
sp Q8NET6 CHSTD_HUMAN	341	27	310	CCRRRVLAAACLGAALLLCAAPRSLR	-1.87
sp Q92185 SIA8A_HUMAN	356	27	305	TRRLPMGASALCVVLCWVIFPVYRL	-1.87
sp Q8NB16 XLT1_HUMAN	393	19	351	YCALLLAAALVAFVYLYG	-1.87
sp Q13241 KLRD1_HUMAN	179	26	148	TTLWRISGLTGIICLSMSTLGILL	-1.86
sp Q8NCH0 CHSTE_HUMAN	376	28	310	LLLPSMLMFAVIVASSGLLMIERGILA	-1.85
sp Q9Y274 SIA10_HUMAN	331	26	305	MRYGLVAIFLSAVFLYYLHCLWGT	-1.84
sp P15291 B4GT1_HUMAN	363	26	332	WLNFLTFLCGSAGFLCSQLFSILL	-1.83
sp Q9NS00 C1GLT_HUMAN	398	25	365	LQRAGRLLVAVCALHLGVTLVYYLA	-1.82
sp Q9P2E5 CHPF2_HUMAN	772	29	738	LLALLRALPILGLSLGCSLLRVSWI	-1.81
sp Q92611 EDEM1_HUMAN	657	27	630	MQWRALVGLVLLRGLHGLVWLVFGL	-1.78
sp Q49A17 GLTL6_HUMAN	601	25	570	FLQOMTLFTVALIFPNVGLWLSLY	-1.78
sp Q9P2W7 B3GA1_HUMAN	334	21	310	RDRILAVLIVLPWTLITVW	-1.77
sp Q6Y288 B3GLT_HUMAN	498	24	470	ACWVLLAPPALLTCLSLAFGLA	-1.77
sp Q06430 GNT2B_HUMAN	400	23	375	LSMRYLFIISVSVIIVFVSFV	-1.77
sp Q8NCL4 GALT6_HUMAN	622	19	593	LRLAMVGCFAVFLFLLHR	-1.74
sp P32970 CD70_HUMAN	193	28	151	YGCVLRALVPLVAGLVICLVQIRFA	-1.73
sp P22413 ENPP1_HUMAN	925	24	828	TYKVLVLSVCVLTTLGCIFGL	-1.73
sp P23510 TNFL4_HUMAN	183	27	133	LLVAVSVIQLGLLLCFTYICLHFSAL	-1.73
sp Q9HDC9 AFMAP_HUMAN	416	25	355	RVRVFTFLMLAVSLTVPLLGAAMLL	-1.72
sp Q9Y2A9 B3GN3_HUMAN	372	19	343	ATLILAGAFLLLSL	-1.71
sp Q6UWU4 CF089_HUMAN	347	23	268	LLVIVYKVLATLGLLITAYFVI	-1.71
sp Q8NFL0 B3GN7_HUMAN	401	24	371	TVYRSLCLALALLAVTVFQRLT	-1.7
sp Q9Y4C5 CHST2_HUMAN	530	25	456	VFRKALVLCAGYALLVTLMLNLL	-1.68
sp Q43908 NKG2F_HUMAN	158	24	64	LTAEVLGIICVIMATVKTIVLI	-1.66
sp P50591 TNF10_HUMAN	281	28	242	LGQTCVLIIFTVLLQSLCAVATVYVFT	-1.65
sp Q8VW01 CANT1_HUMAN	401	20	340	RWKVILTFVGAAILWLLCS	-1.62
sp Q8N4A0 GALT4_HUMAN	578	28	549	AVRWTWAGKSCLLAFLTVAYIFVELLV	-1.62
sp Q8N428 GLT16_HUMAN	558	25	533	MRKIRANAILTVAWILGTFYVLYW	-1.61
sp P15907 SIAT1_HUMAN	406	19	379	KFSCCVLFLFVAVICVWK	-1.61
sp Q8WTT0 CLC4C_HUMAN	213	29	171	LWVFLQKVVSMAMVSIILLSCVCTVSSVV	-1.59
sp Q95395 CNT3_HUMAN	438	24	405	LHYLWALGCYMLLATVALKLSFRL	-1.59
sp Q14788 TNF11_HUMAN	317	25	245	MFVALLGLGLQGVVCSVALFFYFRA	-1.58
sp Q8IUC8 GLT13_HUMAN	556	28	528	MRRFYCKVVLATSLMWWLVDFVLLLYF	-1.56
sp P46695 IEX1_HUMAN	156	19	55	AKRLLFLLTIVFCQILMA	-1.54
sp P22760 AAD_HUMAN	399	19	378	RKSLYLLVIGLIAYIYT	-1.53
sp Q0P6D2 FA69C_HUMAN	419	22	374	TLLAFAAWTAGVWVLAALLLRA	-1.52
sp Q9UBV7 B4GT7_HUMAN	327	25	276	RKCSVHFLVACLCLGFFSLLWLQL	-1.51
sp Q6P531 GGT6_HUMAN	493	23	415	TWARVVAALLLAVGCSLAVRQL	-1.5
sp Q07075 AMPE_HUMAN	957	23	916	VAILCAVVGGLVGLVAGLGLTR	-1.49
sp Q9UQQ1 NALDL_HUMAN	740	26	712	WTKVLGLGLGAAALLGLGILGHFAI	-1.49
sp Q96BA8 CR3L1_HUMAN	519	23	125	TGTCMLVAALCFVVLGSLVPCL	-1.48
sp Q8TEY5 CR3L4_HUMAN	395	28	82	TSTCVLILLFSLAILIPSF	-1.48
sp P55073 IOD3_HUMAN	278	24	236	CLVLFPRFLGTAFMLWLLDFLCIR	-1.47
sp Q95461 LARGE_HUMAN	756	23	726	RRKFLAASLLCIPAITWYLF	-1.47
sp P09172 DOPO_HUMAN	617	20	580	AAMFYSTAVAILVILVAAL	-1.46





sp Q96A11 G3ST3_HUMAN	431	19	397	SRRKILLVLGCSTVSLLI	0,54
sp P08910 ABHD2_HUMAN	425	22	392	AVFDGVKLAAVAALYVIVRCL	0,57
sp P67812 SC11A_HUMAN	179	26	136	LYYQVLNFGMIVSSALMIWKLMVIT	0,57
sp O75356 ENTP5_HUMAN	428	20	407	ATSWGTVFFMLVVCVCSAV	0,59
sp Q10981 FUT2_HUMAN	343	19	317	FSFPMAHFILFVFTVSTIF	0,87
sp O00468 AGRIN_HUMAN	2067	22	2036	LRPLLPLVVAACVLPGAGGTC	0,97
sp Q14494 NF2L1_HUMAN	772	26	740	YLTEGLLQFTILLSLIGVRVDVDTYL	0,98
sp Q9UIR0 BTNL2_HUMAN	455	21	431	FPGYNLGSAVASFLFILLTMK	1,19
sp Q94905 ERLN2_HUMAN	339	20	316	LGAVVAVASSFFCASLFSAV	1,38
sp Q9NST1 PLPL3_HUMAN	481	24	452	RGWLSLFAAGCGFLGFYHVGATRCL	1,47
sp Q96AD5 PLPL2_HUMAN	504	24	474	TWNISFAGCGFLGVYVGVASCLR	1,51
sp Q6P179 ERAP2_HUMAN	960	21	920	RGFYCLTAILPQICICSQFSV	2,19