Table S1. Crystallography Data Collection and Refinement Statistics, Related to Figure 2

	WRC/WIRS seleno*
Data collection	
Space group	$P2_12_12_1$
Cell dimensions	
a, b, c (Å)	97.49, 114.75, 323.61
α, β, γ (°)	90, 90, 90
Resolution (Å)	50-2.43 (2.48-2.43)#
R_{merge}	0.086 (0.54)
$I / \sigma I$	26 (1.84)
Completeness (%)	99.6 (98.8)
Redundancy	6.3 (4.4)
Refinement	
Resolution (Å)	19.98-2.43
No. reflections	262,551
R_{work} / R_{free}	18.4% / 20.9%
No. nonhydrogen atoms	
Protein	22,013
Water	746
B-factors	
Protein	60.4
Water	53.6
R.m.s. deviations	
Bond lengths (Å)	0.002
Bond angles (°)	0.589

*One crystal was used for the data collection.

#Values in parentheses are for the highest resolution shell.

Table S3. DNA Constructs and Peptides Used in This Study, Related to the Experimental Procedures

Construct name	**Description***	**Source or reference**
Sra1	His6-Tev-hSra1 (1-1253, full length), His6-Tev finally removed	(Ismail et al., 2009)
Nap1	His6-Tev-hNap1 (1-1128, full length), His6-Tev finally removed	(Ismail et al., 2009)
ΔWAVE1	MBP-Tev-hWAVE1 (1-178), MBP-Tev finally removed	(Ismail et al., 2009)
miniWAVE1	MBP-Tev-hWAVE1 [(1-186)-(GGS)6-(485-559)], MBP-Tev finally removed	(Chen et al., 2010)
WAVE1-H6-FL [#1, 2]	MBP-Tev-hWAVE1 [(1-452)-HHHHHH-(457-559)], MBP-Tev finally removed	This study
VCA	hWAVE1(485-559)	(Ismail et al., 2009)
$WAVE1_{217}$ [#2]	MBP-Tev-hWAVE1 [(1-217)-(GGS)6-(485-559)], MBP-Tev finally removed	This study
ΔAbi2	MBP-Tev-hAbi2 (1-158), MBP-Tev finally removed	(Ismail et al., 2009)
HSPC300	MBP-Tev-hHSPC300 (1-79) (Full length), MBP-Tev finally removed	(Ismail et al., 2009)
2MBP-HSPC300	MBP-MBP-hHSPC300 (1-79) (Full length)	This study
2MBP-ΔWRC	Sra1 + Nap1 + ΔWAVE1 + ΔAbi2 + 2MBP-HSPC300	This study
$2MBP\text{-}\Delta WRC_{Y923A}$ [#3]	Sra1 (Y923A) + Nap1 + ΔWAVE1 + ΔAbi2 + 2MBP-HSPC300	This study
$2MBP\text{-}\Delta WRC_{L1090A}$ [#3]	Sra1 (L1090A) + Nap1 + ΔWAVE1 + ΔAbi2 + 2MBP-HSPC300	This study
$2MBP\text{-}\Delta WRC_{E1084A}$ [#3]	Sra1 (E1084A) + Nap1 + ΔWAVE1 + ΔAbi2 + 2MBP-HSPC300	This study
$2MBP\text{-}\Delta WRC_{R106A}$ [#3]	Sra1 + Nap1 + ΔWAVE1 + ΔAbi2 (R106A) + 2MBP-HSPC300	This study
$2MBP\text{-}\Delta WRC_{R106M}$ [#3]	Sra1 + Nap1 + ΔWAVE1 + ΔAbi2 (R106M) + 2MBP-HSPC300	This study
$2MBP\text{-}\Delta WRC_{R107A}$ [#3]	Sra1 + Nap1 + ΔWAVE1 + ΔAbi2 (R107A) + 2MBP-HSPC300	This study
$2MBP\text{-}\Delta WRC_{G110W}$ [#3]	Sra1 + Nap1 + ΔWAVE1 + ΔAbi2 (G110W) + 2MBP-HSPC300	This study
$2MBP\text{-}\Delta WRC_{AW}$ [#3]	Sra1 + Nap1 + ΔWAVE1 + ΔAbi2 (R106A/G110W) + 2MBP-HSPC300	This study
miniWRC	Sra1 + Nap1 + miniWAVE1 + ΔAbi2 + HSPC300	(Chen et al., 2010)
2MBP-miniWRC	Sra1 + Nap1 + miniWAVE1 + ΔAbi2 + 2MBP-HSPC300	This study
WRC217	Sra1 + Nap1 + $WAVE1_{217}$ + ΔAbi2 + HSPC300	This study
FL-WRC	Sra1 + Nap1 + WAVE1-H6-FL + ΔAbi2 + HSPC300	This study
dSra	His6-Tev-dSra (1-1291) (Full length), His6-Tev finally removed	(Ismail et al., 2009)
dNap	His6-Tev-dNap (1-1126) (Full length), His6-Tev finally removed	(Ismail et al., 2009)
dΔWAVE	MBP-Tev-dWAVE1 (1-181), MBP-Tev finally removed	(Ismail et al., 2009)
dΔAbi	MBP-Tev-dAbi (1-170), MBP-Tev finally removed	(Ismail et al., 2009)
2MBP-dHSPC300	MBP-MBP-dHSPC300 (1-76) (Full length)	(Ismail et al., 2009)
2MBP-dΔWRC	dSra + dNap + dΔWAVE + dΔAbi + 2MBP-dHSPC300	This study

2MBP-dΔWRC$_{AW}$[#2]	dSra + dNap + dΔWAVE + dΔAbi (R118A/G122W) + 2MBP-dHSPC300	This study
Rac1	hRac1 Q61L full length	(Prigmore et al., 1995)
GST-Rac1	GST-Tev-hRac1 Q61L full length	(Prigmore et al., 1995)
GST-PCDH10 WIRS	GST-Tev-MERSFSTFGKE	This study
GST-PCDH10 WIRS $_{T1002A}$	GST-Tev-MERSFSAFGKE	This study
WIRS peptide	WGAERSFSTFGKEKA	Synthesized (Abgent)
WIRS 2Ala peptide	WGAERSFSAAGKEKA	Synthesized (Abgent)
FITC-WIRS peptide	FITC-GAERSFSTFGKEKA	Synthesized (UTSW)
Seleno-WIRS peptide	WGAERSM*STFGKEKA (M* = selenomethionine)	Synthesized (Abgent)
Peptide A	AVEYSDSEDDSEFDEVDWLE	Synthesized (UTSW)
GST-PCDH10 CT	GST-Tev-mPCDH10 (778-1040)	Openbiosystems (BU511004)
GST-PCDH10 CT with point mutations in WIRS [#4]	GST-PCDH10 CT with single point mutations E997A or R998A or S999(A, G, H P) or F1000(A, M, W, Y, S, G, T, N, P, E, V, K) or S1001(A, D, F, K, L, H, G, P) or T1002(A, S, V, G, C) or F1003(A, W, H, Y) or G1004(A, V, L, E, H) or K1005(A, G, R, M, F, E) or E1006A; or with multiple point mutations, with 997ERSFSTFGKE1006 replaced by 997AAAFATFGKA1006, or 997AAAFATFAKA1006, or 997AAAFATFGAA1006, or 997AAAFATFAAA1006, or 3Ala (F1000A/T1002A/F1003A).	This study
GST-PCDH10 (ΔCM1, ΔCM2) [#2]	GST-Tev-mPCDH10 [(778-906)-GGSEGGGSEGGSTGATSG-(925-939)-ASGSGGGSEGGSEGATS-(957-1040)]	This study
GST-PCDH10 CT$_{short}$	GST-Tev-mPCDH10 (879-1040)	This study
GST-hPCDH10 CT	GST-Tev-hPCDH10 (879-1040)	Openbiosystems (BC111560)
GST-hPCDH10 CT$_{short}$ AA[#3]	GST-Tev-hPCDH10 (879-1040), T1002A/F1003A	This study
GST-PCDH17 CT	GST-Tev-hPCDH17 (857-1159)	Openbiosystems (BC028165)
GST-PCDH17 CT AA[#3]	GST-Tev-hPCDH17 (857-1159), T1000A/F1001A	This study
GST-PCDH18 CT	GST-Tev-hPCDH18 (861-1135)	Openbiosystems (BC093815)
GST-PCDH19 CT	GST-Tev-mPCDH19 (875-1145)	Openbiosystems (BC118529)
GST-PCDH12 CT	GST-Tev-hPCDH12 (960-1184)	Openbiosystems (BC052973)
GST-PCDHα6 CT	GST-Tev-hPCDHa6 (728-919)	Openbiosystems (BC036674)
GST-PCDH8 CT	GST-Tev-hPCDH8 (915-1070)	Openbiosystems (BC036025)
GST-FAT1 CT	GST-Tev-mFAT1 (4215-4436)	Openbiosystems (BC049872)
GST-FAT3 CT[#5]	GST-Tev-mFAT3 (4185-4401)	Openbiosystems (CA318408)
GST-FAT3 CT$_{mh}$[#6]	GST-Tev-mFAT3 (4185-4386)-hFAT3 (4421-4589)	Openbiosystems (clone # LIFESEQ720863)
GST-LRIG3 CT	GST-Tev-hLRIG3 (841-1119)	Openbiosystems (BC126171)

GST-ROBO1 CT	GST-Tev-hROBO1 (886-1078)	Openbiosystems (BC115021)
GST-Neuroligin-1 CT	GST-Tev-hNeuroligin-1 (725-840)	Openbiosystems (BC032555)
GST-Neuroligin-1 CT AA[#3]	GST-Tev-hNeuroligin-1 (725-840), T816A/F817A	This study
NusA-Neuroligin-1 CT	NusA-Tev-hNeuroligin-1 (725-840)	This study
NusA-Neuroligin-1 CT AA[#3]	NusA-Tev-hNeuroligin-1 (725-840), T816A/F817A	This study
GST-Neuroligin-4X CT	GST-Tev-hNeuroligin-4X (721-817)	This study (cDNA from Nils Brose)
GST-Cav1.3 CT	GST-Tev-rCav1.3 (1914-2155)	This study (Zhang et al., 2005)
GST-BAI3 CT	GST-Tev-hBAI3 (1314-1502)	This study (Bolliger et al., 2011)
GST-mGluR5 CT	GST-Tev-rmGluR5 (912-1171)	This study (Ronesi et al., 2012)
GST-GluR6 CT	GST-Tev-rGluR6 (841-908)	This study (Nasu-Nishimura et al., 2010)
GST-P2RX7 CT	GST-Tev-hP2RX76 (359-519)	Openbiosystems (BC011913)
CD16-7-hPCDH10 CT[#7]	hCD16 (1-185)-hCD7 (146-203)-hPCDH10 (739-1040)-mCherry	This study (Blasutig et al., 2008)
CD16-7-hPCDH10 CT AA[#3, 7]	hCD16 (1-185)-hCD7 (146-203)-hPCDH10 (739-1040, T1002A/F1003A)-mCherry	This study (Blasutig et al., 2008)
CD16-7-PCDH17 CT[#7]	hCD16 (1-185)-hCD7 (146-203)-hPCDH17 (729-1159)-mCherry	This study (Blasutig et al., 2008)
CD16-7-PCDH17 CT AA[#3, 7]	hCD16 (1-185)-hCD7 (146-203)-hPCDH17 (729-1159, T1000A/F1001A)-mCherry	This study (Blasutig et al., 2008)
CD16-7-neuroligin1 CT[#7]	hCD16 (1-185)-hCD7 (146-203)-hNeuroligin1 (725-840)-mCherry	This study (Blasutig et al., 2008)
CD16-7-neuroligin1 CT AA[#3, 7]	hCD16 (1-185)-hCD7 (146-203)-hNeuroligin1 (725-840, T816A/F817A)-mCherry	This study (Blasutig et al., 2008)
Sra1-YPet[#8]	hSra1 (1-2353 full length)-(GGS)4-YPet	This study

Notes:

* Protein species: h for human, m for mouse, r for rat and d for drosophila. All sequences were confirmed by DNA sequencing.

#1. To facilitate purification of full-length hWAVE1 from bacterial expression, a His_6 tag was inserted into the unstructured poly-proline region for the MBP-tagged full-length hWAVE1. This allowed double-affinity purification first by amylose beads and then by Ni NTA beads to remove degraded materials.

#2. Inserting or replacing an internal sequence was done using overlapping PCR.

#3. Point mutations were made using QuikChange (Stratagene).

#4. To facilitate cloning, an XhoI and a HindIII restriction site were introduced flanking the WIRS site using QuikChange, producing a variant containing G995L/A996E/A1008L. These mutations did not seem to affect the binding of the cytoplasmic tail to the WRC. To introduce desired mutations to the WIRS, paired DNA oligos were designed to contain the mutations and produce compatible ends for XhoI and HindIII when annealed to form double-stranded oligos. The annealed oligos were directly ligated into the XhoI/HindIII double-digested GST-PCDH10 CT variant.

#5. The commercial cDNA clone lacked the c-terminus of the cytoplasmic tail (4402-4555) and had an internal insertion of 33 amino acids (NASIVTVIQLVNNVVDSIENEVSVMDQGQNYNR) between D4347 and A4349. The insertion does not contain a WIRS, and is conserved in the human FAT3 homologue, but not in rat. This construct contains both predicted WIRS motifs (Table S2).

#6. Two EST clones were assembled to generate a hybrid FAT3 full-length cytoplasmic tail containing 4185-4386 of mFAT3 and 4421-4589 of hFAT3 by overlapping PCR. The human FAT3 (4421-4589) sequence is 88% identical to the mouse FAT3.

#7. These vectors were modified from the pEGFP-N1 vector (Clontech). First the EGFP coding sequence was replaced by mCherry or YPet sequence between the BamHI and NotI sites. The CD16-CD7 coding sequence (Blasutig et al., 2008) was then inserted between NheI and XhoI of the new mCherry N1 vector. The coding sequences of different cytoplasmic tails were then subcloned in frame between XhoI and BamHI.

#8. First the pEGFP-N1 vector (Clontech) was modified by replacing the EGFP coding sequence with the YPet sequence between BamHI and NotI, with a $(GGS)_4$ linker inserted in frame following the BamHI site. The hSra1 coding sequence was then subcloned between XhoI and SacII. The resulting fusion coding sequence for hSra1-$(GGS)_4$-YPet was subcloned into the pGC-IRES vector (Costa et al., 2000) by replacing the sequences between two BamHI sites using the SLIC method (Li and Elledge, 2007).