

**Table S1. Representative peptides for mDAT identified by MS/MS**

Residue number	Peptide sequence	Mascot Score	Observed	Mr(expt)	Mr(calc)	Delta (ppm)	Expect
6-19	K.CSVGPMSSV VAPAK.E	72	695.3438	1388.6731	1388.6792	-4.40	1e-005
6-19	K.CSVGPMSSV VAPAK.E + Oxidation (M)	72	703.3430	1404.6714	1404.6741	-1.95	1.1e-005
6-27	K.CSVGPMSSV VAPAKEPNAVGP.R.E	61	737.3682	2209.0827	2209.0984	-7.07	0.00021
6-27	K.CSVGPMSSV VAPAKEPNAVGP.R.E + Deamidated (NQ)	53	737.7001	2210.0783	2210.0824	-1.83	0.0013
6-27	K.CSVGPMSSV VAPAKEPNAVGP.R.E + Oxidation (M)	59	742.6997	2225.0774	2225.0933	-7.13	0.00037
6-27	K.CSVGPMSSV VAPAKEPNAVGP.R.E + Deamidated (NQ); Oxidation (M)	94	743.0298	2226.0675	2226.0773	-4.39	1.2e-007
20-27	K.EPNAVGP.R.E	46	420.2232	838.4319	838.4297	2.65	0.0021
20-27	K.EPNAVGP.R.E + Deamidated (NQ)	40	420.7142	839.4137	839.4137	0.043	0.0095
36-60	K.EQNGVQLT NSTLINPPQTPVEVQER.E + Deamidated (NQ)	36	931.4699	2791.3878	2791.3988	-3.93	0.069
36-60	K.EQNGVQLT NSTLINPPQTPVEVQER.E + 2 Deamidated (NQ)	15	931.7993	2792.3760	2792.3828	-2.45	7.8
36-60	K.EQNGVQLT NSTLINPPQTPVEVQER.E + Deamidated (NQ); Phospho (ST)	55	958.1257	2871.3552	2871.3651	-3.45	0.0011
36-60	K.EQNGVQLT NSTLINPPQTPVEVQER.E + 2 Deamidated (NQ); Phospho (ST)	40	958.4501	2872.3284	2872.3491	-7.23	0.036
66-85	K.KIDFLLSVIGFAVDLANVWR.F	58	759.4261	2275.2564	2275.2729	-7.22	8.7e-005
86-92	R.FPYLCYK.N	16	495.7411	989.4676	989.4681	-0.46	2.8
126-133	R.EGAAGVWK.I	40	409.2164	816.4182	816.4130	6.36	0.011
134-139	K.ICPVLK.G	25	365.2225	728.4305	728.4255	6.85	0.23
219-227	R.GVLHLHQSR.G	51	523.7967	1045.5789	1045.5781	0.76	0.00081
219-227	R.GVLHLHQSR.G + Deamidated (NQ)	24	524.2909	1046.5672	1046.5621	4.88	0.49
228-236	R.GIDDLGPPR.W	50	470.2498	938.4851	938.4821	3.16	0.0012
260-282	K.TSGKVVWITATMPYVVLTALLR.G	45	844.8183	2531.4329	2531.4549	-8.70	0.00059
260-282	K.TSGKVVWITATMPYVVLTALLR.G + Oxidation (M)	15	850.1583	2547.4530	2547.4499	1.24	0.47
264-282	K.VVWITATMPYVVLTALLR.G	44	720.4235	2158.2486	2158.2588	-4.72	0.00061
264-282	K.VVWITATMPYVVLTALLR.G + Oxidation (M)	69	1088.1315	2174.2485	2174.2537	-2.39	2.3e-006
283-294	R.GVTLPGAMDGIR.A	32	593.8146	1185.6146	1185.6176	-2.56	0.062
283-294	R.GVTLPGAMDGIR.A + Oxidation (M)	60	601.8132	1201.6119	1201.6125	-0.51	0.00013
295-303	R.AYLSVDFYR.L	40	567.2849	1132.5552	1132.5553	-0.044	0.015
337-343	K.FTNNCYR.D	28	487.7087	973.4029	973.4076	-4.81	0.11
374-379	K.HNVPIR.D	29	368.2186	734.4227	734.4187	5.34	0.049
509-520	K.QMTGQRPNLYWR.L	55	517.2611	1548.7615	1548.7619	-0.31	0.00059
509-520	K.QMTGQRPNLYWR.L + Oxidation (M)	45	522.5919	1564.7539	1564.7569	-1.91	0.0069
515-520	R.PNLYWR.L	22	424.7254	847.4363	847.4341	2.69	0.68
579-587	K.FCSLPGSFR.E	28	535.7603	1069.5061	1069.5015	4.26	0.21
588-598	R.EKLAYAITPEK.D	24	631.8477	1261.6808	1261.6918	-8.71	0.4
588-600	R.EKLAYAITPEKDR.Q	50	638.8460	1275.6775	1275.6823	-3.73	0.0012
590-598	K.LAYAITPEK.D	42	503.2844	1004.5542	1004.5542	-0.026	0.0049
590-600	K.LAYAITPEKDR.Q	60	638.8507	1275.6869	1275.6823	3.61	9.6e-005
601-609	R.QLVDRGEVR.Q	18	536.2980	1070.5815	1070.5833	-1.59	1.3
610-614	R.QFTLR.H	35	332.6956	663.3765	663.3704	9.26	0.014
615-619	R.HWLLV.-	20	667.3943	666.3870	666.3853	2.51	0.3

**Table S2.** MS identification of PP2B in the mDAT complex

Uniprot ID <sup>1</sup>	Protein name	Residue	Peptide sequence	Mascot Score <sup>2</sup> (for each peptide)	
				WT	DAT <sup>-/-</sup> KO
PP2BA_MOUSE	Serine/threonine-protein phosphatase 2B catalytic subunit alpha isoform (PP2B catalytic subunit $\alpha$ )	20-28	AVPFPPSHR	18	-
		33-42	EVFDNDGKPR	14, 13, 25, 15	-
		43-47	VDILK	13	-
		101-112	LFEVGGSPANTR	25, 38, 19, 22	14
		113-122	YLFLGDYVDR	39, 44, 22, 29, 29	10
		244-254	TQEHFTHNTVR	15, 16, 25	-
		400-405	IRAIGK	15	-
		406-414	M <sub>ox</sub> ARVFSVLR	-	13
		415-424	EESESVLTLK	12	-
		425-441	GLTPTGM <sub>ox</sub> LPSGVLSGGK	12	26, 41
442-459	QTLQSATVEAIEADEAIK	12, 13	-		
488-501	DAMPSDANLNSINK	37	-		

<sup>1</sup> <http://www.uniprot.org/>

<sup>2</sup> The ions score for an MS/MS match is based on the calculated probability, P, that the observed match between the experimental data and the database sequence is a random event. The reported score is  $-10\log(P)$  ([http://www.matrixscience.com/help/interpretation\\_help.html](http://www.matrixscience.com/help/interpretation_help.html)).

M<sub>ox</sub>: Oxidation at Methionine

**Table S3.** Number of identified phosphopeptides from hDAT WT, hDAT T48A mutant and rDAT transiently expressed in tsA201 cells upon OA treatment (0.5  $\mu$ M, 1 hr)

	Peptide sequence	# of phosphopeptide	# of non-phosphopeptide
hDAT WT + OA	<sup>20</sup> EPNAVGPKEVELILVKEQNGVQLTSSTLTNPR <sup>51</sup> <sup>28</sup> EVELILVKEQNGVQLTSSTLTNPR <sup>51</sup> <sup>36</sup> EQNGVQLTSSTLTNPR <sup>51</sup>	6	45
hDAT T48A + OA	<sup>20</sup> EPNAVGPKEVELILVKEQNGVQLTSSTLANPR <sup>51</sup> <sup>36</sup> EQNGVQLTSSTLANPR <sup>51</sup>	0	51
rDAT WT + OA	<sup>36</sup> EQNGVQLTNSTLINPPQTPVEAQR <sup>60</sup>	0	18

The numbers (#) of phospho- and non-phosphopeptide denote the sum of the different peptide sequences displayed in the second column

**Table S4.** Number of identified phosphopeptides from hDAT T48A mutant and rDAT stably expressed in LLC-PK<sub>1</sub> cells upon OA treatment (0.5 μM, 1 hr)

	Peptide sequence	# of phosphopeptide	# of non-phosphopeptide
hDAT T48A + OA (stable cells)	<sup>20</sup> EPNAVGPKEVELILVKEQNGVQLTSSTLTNPR <sup>51</sup>	0	47 (ion trap)
	<sup>28</sup> EVELILVKEQNGVQLTSSTLTNPR <sup>51</sup> <sup>36</sup> EQNGVQLTSSTLTNPR <sup>51</sup>	0	239 (QTOF)
rDAT WT + OA (stable cells)	<sup>36</sup> EQNGVQLTNSTLINPPQTPVEAQR <sup>60</sup>	0	121 (QTOF)

The numbers (#) of phospho- and non-phosphopeptide denote the sum of the different peptide sequences displayed in the second column