

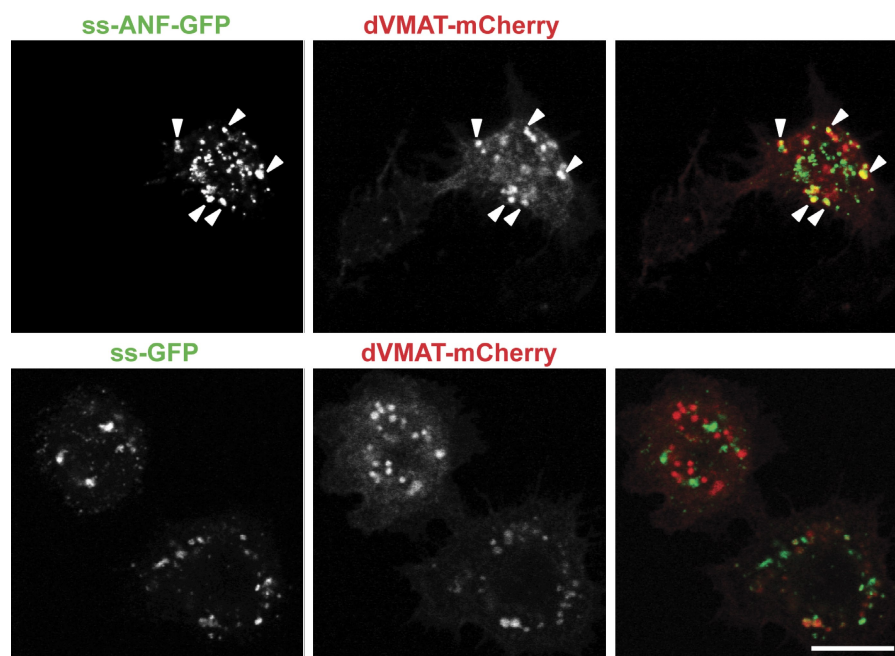
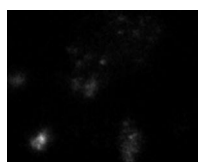
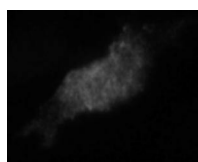
Asensio et al., <http://www.jcb.org/cgi/content/full/jcb.201006131/DC1>

Figure S1. **Colocalization of dVMAT with ANF-GFP but not ss-GFP in S2 cells.** S2 cells transiently transfected with mCherry-dVMAT and either ANF-GFP (top) or ss-GFP (bottom) were plated on conA, washed, fixed, and visualized by confocal microscopy (LSM510; Carl Zeiss, Inc.) using a 100× 1.3 NA Plan Neofluar oil immersion objective. dVMAT colocalizes partially with ANF-GFP (arrowheads), but very little if at all with ss-GFP. Bar, 10 μ m.



Video 1. **Live imaging of VMAT2-pHluorin basal and regulated exocytosis in PC12 cells treated with control siRNA.** PC12 cells were transfected with 50 nM control siRNA, cotransfected 2 d later with the same siRNA and VMAT2-pHluorin, then imaged live by TIRF microscopy after an additional 2 d. Basal exocytosis of VMAT2-pHluorin was measured in Tyrode's solution containing 2.5 mM K^+ , and release stimulated after 30 s with Tyrode's containing 90 mM K^+ . Two images were collected per second for a total of 1 min. Video corresponds to Fig. 5.



Video 2. **Live imaging of VMAT2-pHluorin basal and regulated exocytosis in PC12 treated with AP-38 siRNA.** PC12 cells were transfected with 50 nM AP-38 siRNA, cotransfected 2 d later with the same siRNA and VMAT2-pHluorin, then imaged live by TIRF microscopy after an additional 2 d as described for Video 1.

Table S1. Genes identified in the S2 cell screen that were excluded from further analysis

Cg	Cell no.	GFP (z score)	Protein
	% mean		
17603	0.1	6.5	Transcription initiation factor TFIID
8117	0.4	4.1	Transcription elongation factor TFIIIS
8151	0.4	7.0	TFIIH basal transcription factor complex subunit 1
1420	0.6	-1.3	Pre-mRNA-splicing factor SLU7
8151	2.0	0.2	TFIIH basal transcription factor complex subunit 1
8987	2.3	8.8	DNA polymerase gamma subunit 1
12225	4.2	-1.7	Transcription elongation factor SPT6
7238	5.5	0.3	Essential protein present in native splicing complexes
11979	5.5	-3.0	DNA-directed RNA polymerase II
3350	5.7	-0.1	Transcription factor-like protein 4
5193	6.2	-0.9	Transcription initiation factor IIB
4651	7.1	1.4	60S ribosomal protein L13
1433	9.2	-0.3	Transcription unit protein
12324	9.5	-2.2	40S ribosomal protein S15Ab
2875	9.6	0.9	Nucleolar complex protein 4 homologue
12261	9.7	-0.9	Mitochondrial ribosomal protein S22
2503	9.9	-0.8	Paf1, RNA polymerase II-associated factor
4647	10.3	0.0	Mitochondrial 39S ribosomal protein L49
12275	11.3	-1.4	40S ribosomal protein S10a
6249	11.4	-1.7	3'-5' exoribonuclease CSL4 homologue
5317	11.9	1.6	Ribosomal protein L7
3455	12.2	1.7	Proteasome subunit p42
8142	12.3	0.0	Replication factor C subunit 4
5931	12.5	1.0	U5 small nuclear ribonucleoprotein 200-kD helicase
7993	15.5	1.5	Processing of pre-rRNA and the assembly of the 60S ribosomal subunit
12373	15.7	-0.2	Mitochondrial ribosomal protein L18
4760	16.0	-0.6	RNP
5818	16.3	-0.8	Mitochondrial ribosomal protein L4
3312	16.5	-0.4	RNA-binding protein 4F
9348	19.0	5.0	TAF6 RNA polymerase II
7728	19.3	0.0	Ribosome biogenesis protein BMS1 homologue
8064	19.4	0.3	18S rRNA biogenesis
2063	19.6	2.0	Transcriptional regulator protein
3294	19.7	-0.6	U2 small nuclear ribonucleoprotein auxiliary factor 35
1307	20.1	-0.1	Peptidyl-tRNA hydrolase 2
33123	21.0	0.4	Leucyl-tRNA synthetase
2670	23.5	0.3	Transcription initiation factor TFIID subunit 7
4651	23.7	0.7	60S ribosomal protein L13
3180	28.1	0.2	DNA-directed RNA polymerase II
8939	28.2	0.1	Ribosomal RNA methyltransferase
9091	29.0	1.9	60S ribosomal protein L37
3308	29.6	0.4	Deoxyribonuclease
18174	30.5	-0.9	26S proteasome non-ATPase regulatory subunit 14
7246	33.5	-0.5	U3 small nucleolar RNA-associated protein 6
3817	37.8	-0.7	Nucleolar protein, constituent of pre-60S ribosomal
7847	38.7	1.2	Transcription factor
7028	38.7	-1.1	PRP4 pre-mRNA-processing factor 4
7006	40.0	-0.7	60S ribosome subunit biogenesis protein NIP7
11879	42.7	1.7	Yemanuclein
3193	42.9	2.5	Spliceosome assembly factor
6539	43.3	-0.1	DEAD/DEAH RNA helicase 1
1420	43.5	1.2	Step II splicing factor SLU7
2972	44.0	-3.3	RNA-binding protein NOB1
1957	45.0	-0.6	Cleavage and polyadenylation specificity factor
2194	45.2	1.5	Dihydropyrimidine dehydrogenase
4528	46.2	4.4	U1 small nuclear ribonucleoprotein A
3695	47.1	-1.5	Cofactor required for Sp1 transcriptional activation, subunit 3

Table S1. Genes identified in the S2 cell screen that were excluded from further analysis (Continued)

Cg	Cell no.	GFP (z score)	Protein
2939	47.6	0.1	Transcription factor
4573	48.0	-0.3	Mitochondrial glutamyl-tRNA synthetase
8950	49.7	-0.3	Transcription factor IIIC- γ subunit
12288	49.9	0.9	Required for pre-25S rRNA processing
3808	50.0	0.1	tRNA methyltransferase
9054	58.0	-0.4	ATP-dependent RNA helicase
9193	58.0	0.3	Cyclin
7776	63.9	0.8	Histone H4/H2A acetyltransferase
1873	67.2	-0.3	Elongation factor 1- α
9151	72.5	0.1	Transcription factor
8915	74.6	-0.5	RNA helicase
3663	76.2	0.6	Isochorismatase domain containing 1
13867	78.7	3.1	Mediator of RNA polymerase II transcription
1245	83.2	0.3	Transcription factor
12752	85.1	0.5	NTF2-related export protein 2
6348	96.1	-0.3	Transcription factor
3668	98.5	0.9	Fork head transcription factor
11107	108.4	-0.1	Pre-mRNA splicing factor ATP-dependent RNA helicase
6493	109.7	9.6	Dicer-2
18783	124.1	0.6	Transcription factor
8922	126.6	0.2	40S ribosomal protein S5a
6684	128.8	-0.1	40S ribosomal protein S25
7439	139.7	9.4	Argonaute-2
3497	144.4	-0.3	Transcription factor RBP-L
5874	145.1	2.9	Negative elongation factor A
17841	146.5	-0.3	Purine-binding transcription factor
8900	158.2	0.3	40S ribosomal protein S18
12288	173.9	-0.6	RNA-binding protein 34
1671	247.8	3.0	Transducing

These genes, with their known or putative function indicated, were excluded from further analysis because of the low number of cells available for analysis or effects on expression of the GFP-dVMAT reporter relative to other wells from the same plate. The remainders were excluded because, like those reducing cell number or altering GFP expression, they function in transcription or translation.