Supplementary information

Methods

Construction of networks from microarray gene expression data

In this work, we create networks using different values of gene expression fold change (calculated as a logarithm of the base 2), from PSMD10$^{\text{Gankyrin}}$ overexpression data. The selection of the right value of fold change for the network needs careful optimization. Choosing too high a value of fold change, f, might obviously exclude many biological meaningful interactions. Again, setting too low a value of f would lead to the inclusion of too many interactions in the network. Therefore, we constructed networks at various values of f like 2, 2.5, 3, 3.5. Networks constructed with f of value two or more already contain a lot of nodes and edges. Networks constucted with genes possessing values of f of 4 or more do not contain too many nodes and would lead to exclusion of important information. In our networks, two genes are connected by interactions selected from text-mined interactomes (1). Only interactions above a score of over 0.9 (90% or more) have been chosen to create networks of very high quality. We then analyse these networks using various topological metrics to find important genes and important interactions as detailed below. We find that the list of important interactions attained from the network of genes possesing f of 3 or more is not remarkably different from the network of genes having f of 3.5 or more. Therefore, in this work we have restricted ourselves to create networks from upregulated genes induced by PSMD10$^{\text{Gankyrin}}$ overexpression exceeding a fold change ≥ 3 and choose a rather stringent cutoff value of 90% in our text-mined interactomes (1) to mitigate the possibility of selecting spurious or noisy interactions.

Topological analysis of networks: important genes and important interactions

We have conducted an extensive topological analysis of the networks constructed by methods outlined above. Both weighted and unweighted networks have been investigated here. However, we find the important conclusions to be essentially similar for both weighted and unweighted networks. Our network is constructed out of genes exhibiting a value of f of 3 or more and contains 249 genes and 292 edges.

Degree or the number of connections that a node has to other nodes in the network is arguably the most commonly studied metric in network science. However, it is well-known from the theory of complex networks, that other network metrics should also be examined in parallel. In this work, we have also measured node betweenness (2, 3), edge betweenness (4), edge closeness and edge proximity (2), in addition to degree.

Betweenness (2, 4), B_k of edge (or node), k, in a network, G, measures the number of shortest paths passing through k in the network, G. Thus, if σ_{ij} is the number of shortest paths in G from edge (or node), i, to edge (or node), j, and $\sigma_{ij}(k)$ is the number of shortest paths in G from i to j which pass through k, then, $B_k = \sum_{i \neq k \neq j} [\sigma_{ij}(k)/\sigma_{ij}]$ Betweenness is thought to capture the flow of information in a given network. Nodes with low degree but high betweenness usually capture the bottlenecks in a network.

Node closeness, C_i, of node i, is the reciprocal of average distance of i to every other node, j, in a network, G, of N nodes. Thus, $C_i = (N-1)/\sum_j d_{ij}$

Edge proximity (2, 3), P_k, of an edge, k, in the network G, with M edges, is the inverse of the sum of its shortest distance d_{kl} to all other *edges*, l. Therefore, $P_k = (M-1)/\sum_l d_{kl}$ Calculation of edge proximity would require the construction of the line graph in the manner detailed in the earlier report (2, 3).

Virus production and hNPCs transduction

The HEK293-FT cells were transfected with pTRIPZ-empty vector or pTRIPZ-3XFlag- PSMD10$^{\text{Gankyrin}}$ construct with the packaging vectors (PAX2 and pMD2G) at 4 : 3 : 1 ratio. Media was collected at 48 h and 72 h post-transfection, filtered with 0.22 μM syringe-filter (*Millipore*) and ultracentriguged at 30,000 rpm, 4℃ for 90 min. The virus pellet was resuspended in fresh Neural Stem Cells maintenance media and stored at -80℃ for transduction purpose. The virus titer was calculated by tranducing HEK293 cells with pTRIPZ empty vector virus and determined the % of RFP +ve cells by FACS analysis. By using the formula [Virus Titer=(No. of cell seeded $\mathbf{X}$% of cell RFP+ve) $\mathbf{X}$ dilution factor] virus titer was calculated. hNPCs were transduced with the virus particle in presence of polybrene (10 μg/mL: *Sigma*). For RFP or Flag- PSMD10$^{\text{Gankyrin}}$ expression, cells were induced with 1 μg/mL of doxycycline (*Sigma*).

Materials

Antibodies

Primary antibodies: **anti-PSMD10**^{Gankyrin} 1 : 1000 (mouse monoclonal, *Sigma* and rabbit polyclonal, *Sigma*), **anti-FLAG** 1 : 8000 (mouse monoclonal, *Sigma*), **anti-β-actin** in 1 : 2000 (mouse monoclonal, *Sigma*), **anti-α-tubulin** 1 : 2000 (mouse monoclonal, *Sigma*), **anti-β7** 1 : 1000 (mouse monoclonal, *Biomol*), **anti-α4** in 1 : 1000 (mouse monoclonal, *Biomol*), **anti-α5** in 1 : 1000 (mouse monoclonal, *Biomol*), **anti-Rpt6** 1 : 1000 (mouse monoclonal, *Biomol*), **anti-β-III tubulin** 1 : 1000/ 1 : 100 (WB/IF) (*Millipore* polyclonal mouse, polyclonal chicken), **anti-GFAP** 1 : 1000/1 : 100 (WB/IF) (*Millipore* polyclonal rabbit), **anti-nestin** 1 : 1000/1 : 100 (WB/IF) (*Millipore* monoclonal mouse), **anti-nestin** 1 : 1000/1 : 300 (WB/IF) (*Sigma* polyclonal rabbit), **anti-SOX2** 1 : 1000/1 : 100 (WB/IF) (*Millipore* polyclonal rabbit), **anti-Musashi** 1 : 1000/1 : 100 (WB/IF) (*Millipore* polyclonal rabbit), **anti-Oligodendrocyte marker O1** 1 : 1000 (*Millipore* monoclonal mouse), **anti-β-catenin** 1 : 1000 (mouse monoclonal, *Cell signalling*),

Secondary antibodies; anti-mouse-HRP (*GE-healthcare*), anti-rabbit-*HRP* (*GE healthcare*), **anti-mouse-FITC** (*Santa Cruz*), **anti-mouse-Alexaflour568** (*Invitrogen*), **anti-mouse-TR** (*Santa Cruz*), **anti-rabbit-Alexaflour488** (*Invitrogen*), **anti-rabbit-Alexaflour568** (*Invitrogen*), **anti-chicken-Alexa633** (*Invitrogen*) were used.

List of cloning primer:

PSMD10^{Gankyrin} in pCMV10-3XFlag	Fw: AAGCTTATGGACTACAAAGACCATG
	Rv: GAATTCTTAACCTTCCACCATTCTCTTG
FLAG-PSMD10^{Gankyrin} in pTRIPZ	Fw: ACCGGTCGCCACCATGGACTACAAAGACCATG
	Rv: GAATTCTTAACCTTCCACCATTCTCTTG

List of real time primer:

Primer Name	Primer Sequence
GAPDH-Fw	5′-ATCGTGGAAGGACTCATGACC-3′
GAPDH-Rv	5′-AGGGATGATGTTCTGGAGAGC-3′
α4-Fw	5′- CGCTACATCGCCAGTCTGAAG -3′
α4-Rv	5′- GAGCCTAGGAGTGCCATCAAAG -3′
β1- Fw	5′- AATCGAGTGACTGACAAGCTGAC -3′
β1-Rv	5′- CAGTGGAGGCTCATTCAGTTC -3′
β2-Fw	5′- TGAAGGGATGGTTGTTGCTGAC -3′
β2-Rv	5′- GGAAGAAATGAGCTGGGTTGTC -3′
β5-Fw	5′- CGGCAATGTCGAATCTATGAGC -3′
β5-Rv	5′- GCCTCTCTTATCCCAGCCACAG -3′
β7-Fw	5′- CTCAGTCCTCGGCCGTTAAGTTC -3′
β7-Rv	5′- GCATGGTACTGTTGTTGACTCG -3′
PSMD4 Fw	5′- CTACATACTGTCCAACCCAA -3′
PSMD4-Rv	5′- AGTTTCACCAGATCCTTCTC -3′
PSMD9-Fw	5′-AAGGCCAACTATGACGTGCTG-3′
PSMD9-Rv	5′-ATATGATGTTGTGCCTGGCG-3′
PSMD10-Fw	5′- GCAGCTTCGAAAAACAGGCA -3′
PSMD10-Rv	5′- GGATGTTTGTGGATGCTTTG -3′
NGN1-Fw	5′- GACCTATCCGGCTTCCTCAC -3′
NGN1-Rv	5′- TCCTGCTCGTCGTCCTGTG -3′
NRG1-Fw	5′-AGGTGAGAACGCCCAAGTC -3′
NRG1-Rv	5′-TCTCCTTCTCCGCACATTTTAC -3′
β-catenin-Fw	5′-CTGGTGCCACTACCACAGCTC -3′
β-catenin-Rv	5′-GCTCGTACCCTCTGAGCTCG -3′

1. Iossifov I, Rodriguez-Esteban R, Mayzus I, Millen KJ, Rzhetsky A. Looking at cerebellar malformations through text-mined interactomes of mice and humans. PLoS Comput Biol. 2009;5(11):e1000559. doi: 10.1371/journal.pcbi.1000559. PubMed PMID: 19893633; PubMed Central PMCID: PMCPMC2767227.
2. Kaur Grewal R, Mitra D, Roy S. Mapping networks of light-dark transition in LOV photoreceptors. Bioinformatics. 2015;31(22):3608-16. doi: 10.1093/bioinformatics/btv429. PubMed PMID: 26209799.
3. Banerjee SJ, Sinha S, Roy S. Slow poisoning and destruction of networks: edge proximity and its implications for biological and infrastructure networks. Phys Rev E Stat Nonlin Soft Matter Phys. 2015;91(2):022807. doi: 10.1103/PhysRevE.91.022807. PubMed PMID: 25768552.
4. Girvan M, Newman ME. Community structure in social and biological networks. Proc Natl Acad Sci U S A. 2002;99(12):7821-6. doi: 10.1073/pnas.122653799. PubMed PMID: 12060727; PubMed Central PMCID: PMCPMC122977.

Table S1. KEGG pathway_GSEA over representation (Bold letter corresponds to the pathways related to neurogenesis):

Gene Set Name	# Genes in Gene Set (K)	Description	# Genes in Overlap (k)	k/K	p-value	FDR q-value
KEGG_NEUROACTIVE_LIGAND_RECEPTOR_ INTERACTION	**272**	**Neuroactive ligand-receptor interaction**	**20**	**0.0735**	**3.25E-07**	**5.92E-05**
KEGG_AMYOTROPHIC_LATERAL_SCLEROSIS_ALS	53	Amyotrophic lateral sclerosis (ALS)	9	0.1698	6.36E-07	5.92E-05
KEGG_CALCIUM_SIGNALING_PATHWAY	178	Calcium signaling pathway	14	0.0787	8.64E-06	5.36E-04
KEGG_PATHWAYS_IN_CANCER	328	Pathways in cancer	19	0.0579	2.01E-05	9.36E-04
KEGG_WNT_SIGNALING_PATHWAY	**151**	**Wnt signaling pathway**	**12**	**0.0795**	**3.38E-05**	**1.26E-03**
KEGG_FOCAL_ADHESION	201	Focal adhesion	13	0.0647	1.35E-04	4.18E-03
KEGG_COLORECTAL_CANCER	62	Colorectal cancer	7	0.1129	1.71E-04	4.53E-03
KEGG_LONG_TERM_POTENTIATION	70	Long-term potentiation	7	0.1	3.65E-04	8.48E-03
KEGG_SMALL_CELL_LUNG_CANCER	84	Small cell lung cancer	7	0.0833	1.10E-03	2.27E-02
KEGG_MAPK_SIGNALING_PATHWAY	267	MAPK signaling pathway	13	0.0487	1.93E-03	3.59E-02

Table S2. Reactome signaling_GSEA over representation (Bold letter corresponds to the signaling pathways related to neurogenesis):

Gene Set Name	# Genes in Gene Set (K)	Description	# Genes in Overlap (k)	k/K	p-value	FDR q-value
REACTOME_SIGNALING_BY_GPCR	920	Genes involved in Signaling by GPCR	52	0.0565	4.50E-12	3.03E-09
REACTOME_GPCR_DOWNSTREAM_ SIGNALING	805	Genes involved in GPCR downstream signaling	44	0.0547	5.78E-10	1.95E-07
REACTOME_GPCR_LIGAND_BINDING	408	Genes involved in GPCR ligand binding	29	0.0711	1.70E-09	3.81E-07
REACTOME_NEURONAL_SYSTEM	**279**	**Genes involved in Neuronal System**	**23**	**0.0824**	**5.16E-09**	**8.69E-07**
REACTOME_CLASS_A1_RHODOPSIN_ LIKE_RECEPTORS	305	Genes involved in Class A/1 (Rhodopsin-like receptors)	23	0.0754	2.74E-08	3.69E-06
REACTOME_NEUROTRANSMITTER_ RECEPTOR_BINDING_AND_ DOWNSTREAM_TRANSMISSION_ IN_THE_POSTSYNAPTIC_CELL	**137**	**Genes involved in Neurotransmitter Receptor Binding And Downstream Transmission In The Postsynaptic Cell**	**14**	**0.1022**	**3.83E-07**	**4.31E-05**
REACTOME_TRANSMISSION_ ACROSS_CHEMICAL_SYNAPSES	**186**	**Genes involved in Transmission across Chemical Synapses**	**16**	**0.086**	**6.13E-07**	**5.91E-05**
REACTOME_PEPTIDE_LIGAND_ BINDING_RECEPTORS	188	Genes involved in Peptide ligand-binding receptors	16	0.0851	7.08E-07	5.96E-05
REACTOME_G_ALPHA_I_ SIGNALLING_EVENTS	195	Genes involved in G alpha (i) signalling events	16	0.0821	1.15E-06	8.62E-05
REACTOME_ACTIVATION_OF_NMDA_ RECEPTOR_UPON_GLUTAMATE_ BINDING_AND_POSTSYNAPTIC_EVENTS	37	Genes involved in Activation of NMDA receptor upon glutamate binding and postsynaptic events	7	0.1892	5.38E-06	3.48E-04

Table S3. David functional analysis: (Bold letter corresponds to the cellular part related to neurogenesis):

Category	Term	Count	%	p-Value	List Total	Pop Hits	Pop Total	Fold Enrichment	Bonferroni	Benjamini	FDR
GOTERM_ CC_2	**GO:0097458~ neuron part**	**50**	**12.43781**	**3.50E-05**	**363**	**1333**	**17877**	**1.847259**	**0.003248**	**0.003248**	**0.038286**
GOTERM_ CC_2	GO:0044459~plasma membrane part	79	19.65174	2.83E-04	363	2635	17877	1.476504	0.026024	0.013098	0.309918
GOTERM_ CC_2	GO:0005886~ plasma membrane	135	33.58209	3.69E-04	363	5141	17877	1.293225	0.033769	0.011386	0.403565
GOTERM_ CC_2	GO:0071944~ cell periphery	136	33.83085	6.69E-04	363	5256	17877	1.2743	0.060357	0.015443	0.730154
GOTERM_ CC_2	GO:0098590~plasma membrane region	34	8.457711	0.001434	363	936	17877	1.788921	0.12496	0.026344	1.559043
GOTERM_ CC_2	**GO:0098793~ presynapse**	**15**	**3.731343**	**0.001933**	**363**	**282**	**17877**	**2.619571**	**0.164647**	**0.029538**	**2.095433**
GOTERM_ CC_2	GO:0042995~ cell projection	56	13.93035	0.002235	363	1846	17877	1.493978	0.18784	0.029285	2.419404
GOTERM_ CC_2	**GO:0008021~ synaptic vesicle**	**8**	**1.99005**	**0.014866**	**363**	**127**	**17877**	**3.102232**	**0.751654**	**0.159801**	**15.12314**

Table S4. David functional tool: (Bold letter corresponds to the functions related to neurogenesis):

Category	Term	Count	%	p-Value	List Total	Pop Hits	Pop Total	Fold Enrichment	Bonferroni	Benjamini	FDR
UP_KEYWORDS	Glycoprotein	123	30.59701	5.83E-05	402	4551	20581	1.38369	0.018439	0.018439	0.078661
UP_KEYWORDS	Calcium	34	8.457711	2.53E-04	402	877	20581	1.984814	0.07764	0.039604	0.34114
UP_KEYWORDS	Cell membrane	88	21.89055	4.88E-04	402	3175	20581	1.41899	0.144292	0.050616	0.656705
UP_KEYWORDS	Cleavage on pair of basic residues	15	3.731343	0.001993	402	294	20581	2.612067	0.470848	0.147106	2.6553
UP_KEYWORDS	Cell adhesion	20	4.975124	0.002886	402	479	20581	2.137642	0.602263	0.168391	3.823287
UP_KEYWORDS	Cell junction	25	6.218905	0.00353	402	675	20581	1.896167	0.676387	0.171414	4.658345
UP_KEYWORDS	Transducer	30	7.462687	0.005727	402	899	20581	1.708449	0.839909	0.230271	7.453784
UP_KEYWORDS	Secreted	55	13.68159	0.006232	402	1965	20581	1.432981	0.863871	0.22063	8.086078
UP_KEYWORDS	Ion channel	15	3.731343	0.011216	402	359	20581	2.13913	0.972629	0.329551	14.11353
UP_KEYWORDS	Palmitate	14	3.482587	0.011381	402	324	20581	2.212195	0.97404	0.305887	14.30553
UP_KEYWORDS	Membrane	169	42.0398	0.012687	402	7494	20581	1.154552	0.982973	0.309452	15.82018
UP_KEYWORDS	G-protein coupled receptor	27	6.716418	0.01437	402	842	20581	1.641694	0.990119	0.31939	17.73513
UP_KEYWORDS	Heme	8	1.99005	0.014946	402	132	20581	3.102819	0.991801	0.308935	18.38163
UP_KEYWORDS	Lipoprotein	27	6.716418	0.016562	402	852	20581	1.622425	0.995145	0.316512	20.16962
UP_KEYWORDS	Defensin	5	1.243781	0.018432	402	52	20581	4.922742	0.997354	0.326753	22.19245
UP_KEYWORDS	Signal	99	24.62687	0.021174	402	4160	20581	1.218379	0.998916	0.347334	25.07359
UP_KEYWORDS	Monooxygenase	6	1.492537	0.028792	402	88	20581	3.490672	0.99991	0.422009	32.56765
UP_KEYWORDS	Receptor	44	10.94527	0.030492	402	1648	20581	1.366897	0.999949	0.42236	34.14311
UP_KEYWORDS	Disease mutation	63	15.67164	0.038119	402	2550	20581	1.264855	0.999996	0.479265	40.79761
UP_KEYWORDS	Transmembrane helix	127	31.59204	0.038985	402	5634	20581	1.154057	0.999997	0.469673	41.5128
UP_KEYWORDS	Transmembrane	127	31.59204	0.042302	402	5651	20581	1.150585	0.999999	0.481377	44.17769
UP_KEYWORDS	Growth arrest	3	0.746269	0.047172	402	18	20581	8.532753	1	0.503741	47.88728
UP_KEYWORDS	**Synapse**	**13**	**3.233831**	**0.048113**	**402**	**357**	**20581**	**1.864299**	**1**	**0.495354**	**48.57715**
UP_KEYWORDS	**Neurogenesis**	**10**	**2.487562**	**0.057013**	**402**	**250**	**20581**	**2.047861**	**1**	**0.541715**	**54.6969**
UP_KEYWORDS	Serine protease	7	1.741294	0.05811	402	141	20581	2.541671	1	0.534157	55.40262
UP_KEYWORDS	Extracellular matrix	10	2.487562	0.066699	402	258	20581	1.984361	1	0.571266	60.58619
UP_KEYWORDS	Protease	17	4.228856	0.067406	402	544	20581	1.599891	1	0.561545	60.98677
UP_KEYWORDS	Ligand-gated ion channel	5	1.243781	0.070963	402	80	20581	3.199782	1	0.567681	62.94677
UP_KEYWORDS	Voltage-gated channel	7	1.741294	0.073636	402	150	20581	2.389171	1	0.568877	64.35893
UP_KEYWORDS	Potassium transport	6	1.492537	0.076655	402	116	20581	2.648096	1	0.571743	65.89426
UP_KEYWORDS	Polymorphism	250	62.18905	0.077621	402	12043	20581	1.062786	1	0.56458	66.37227
UP_KEYWORDS	DNA-binding	50	12.43781	0.077683	402	2050	20581	1.248696	1	0.553421	66.40308
UP_KEYWORDS	Methylation	27	6.716418	0.083821	402	1001	20581	1.380925	1	0.570982	69.29664
UP_KEYWORDS	Alternative splicing	221	54.97512	0.090965	402	10587	20581	1.06871	1	0.591317	72.37335
UP_KEYWORDS	EGF-like domain	9	2.238806	0.095161	402	238	20581	1.936003	1	0.598045	74.04459
UP_KEYWORDS	Activator	19	4.726368	0.097436	402	661	20581	1.471609	1	0.596831	74.91089

A *Neural progenitor cells (Undifferentiated)*

14-days

B *Neural progenitor cells (Differentiated 14-days)*

Low density   *High density*

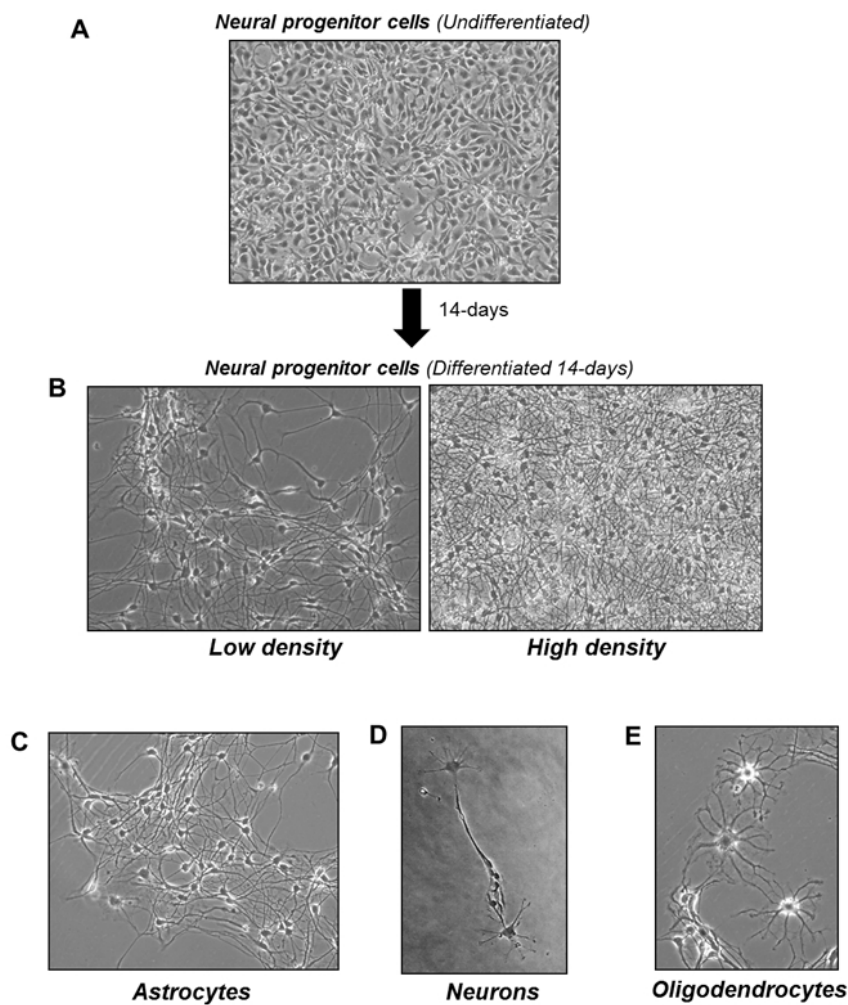**C** *Astrocytes*   **D** *Neurons*   **E** *Oligodendrocytes*

Fig. S1. Human neural progenitor cells were differentiated into Astrocytes, Neurons and oligodendrocytes: (A) Phase Contrast image of human Neural progenitor cells. Cells were grown on Laminin coated plate in Neural stem cell maintenance media with supplements EGF (20 ng/mL) and FGF (20 ng/mL) for a period of 72 hr. Image shows >90% confluent mono layer cells. (B) hNPCs were grown on Laminin coated plates till they reached ~80% confluence. Then media was replaced with fresh DF media with changes on alternate days until 12th day of differentiation. Images were taken using a phase-contrast microscope. Differentiated cells (C) astrocytes showing the network like structure, (D) neurons showing the axon and cell body with dendritic processes and (E) oligodendrocytes dendritic net like phenotype.
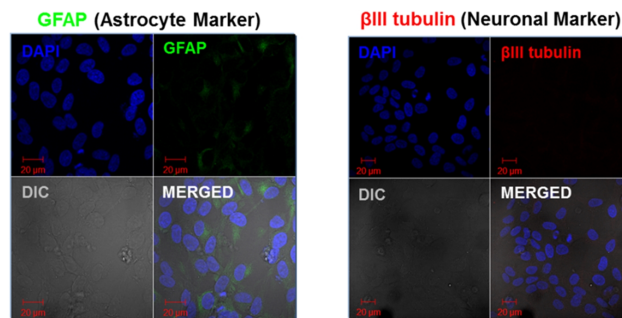
Fig. S2. Human neural progenitor cells do not express differentiation markers: hNPCs were grown on laminin coated glass coverslips to ~80% confluency. Immunofluorescence was performed following the protocol described in materials and methods. Cells show negative/minimal expression of GFAP (in green) and β-III tubulin (in red), DAPI was (in blue) used for nuclear staining. Images were acquired in Laser confocal microscope (Zeiss LSM meta-510).
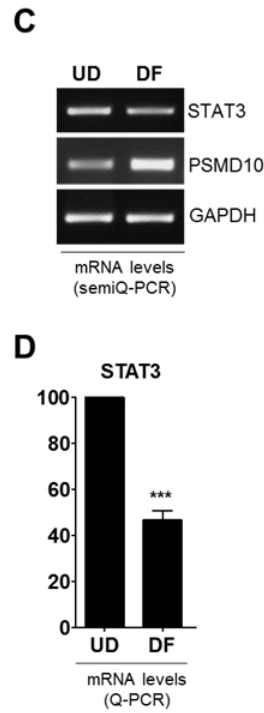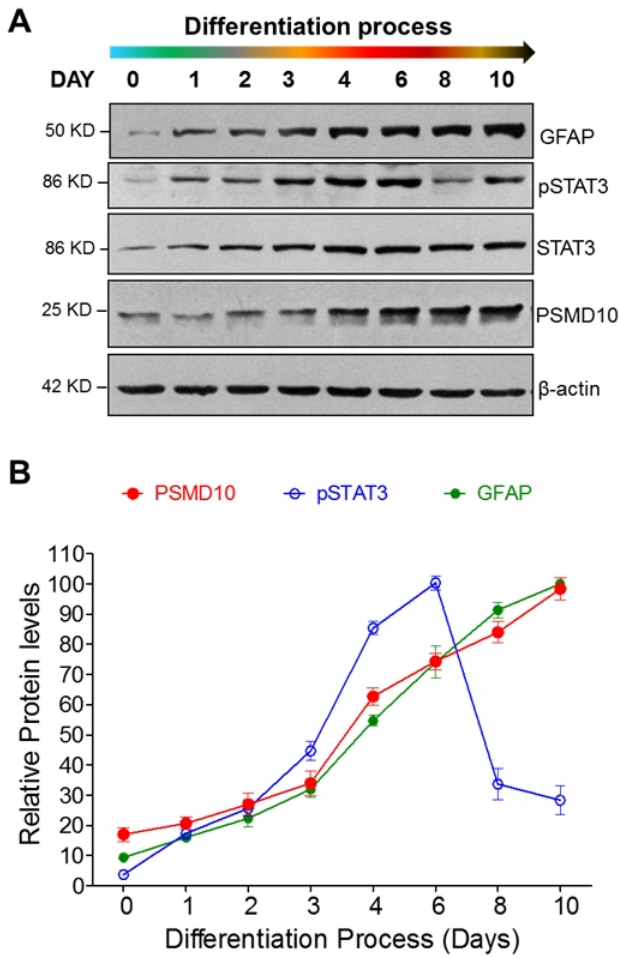
Fig. S3. STAT3 expression and phosphorylation during the hNPCs differentiation process. (A) Progenitor cells grown on laminin coated plates to ~80% confluency, were differentiated as described in Fig. 2 with a minor change; differentiation was stopped at day 10. Cells at each day of differentiation were collected, lysed and analyzed by western blot. (B) Line graphs show the expression levels of depicted proteins from Fig. 3J at each day during the differentiation process. Data represents the average protein levels of two independent (biological repeat) experiments. (C) Semi-Q PCR gel image shows the mRNA levels of PSMD-10^Gankyrin and STAT3 in the progenitor cells (UD) and differentiated cells. (D) Graph represents mRNA levels of STAT3 in the progenitor cells (UD) and differentiated cells (DF) measured by the Real-time PCR. Data represents the average fold increase of mRNA levels (normalized with GAPDH) of three independent experiments with ±SEM.
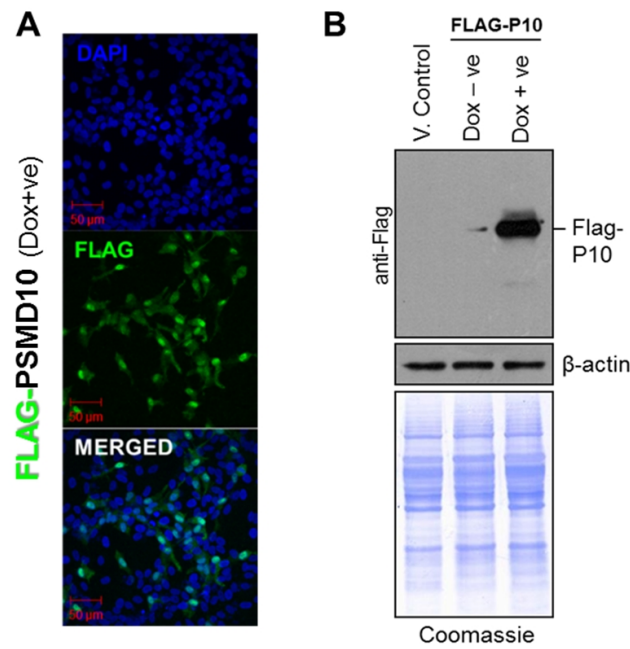
Fig. S4. Trans-expression of Flag-PSMD10[Gankyrin] in hNPCs by lentiviral transduction. hNPCs were grown on Laminin coated glass coverslips to ~30% confluency and transduced with virus particles carrying of pTRIPZ-3XFlag- PSMD10[Gankyrin]. To induce the expression of PSMD10[Gankyrin], cells were treated either with doxycycline (1 μg/mL) for 48 hr or left untreated. (A) Immunofluorescence was performed using anti-Flag antibody after 48 h of transduction following the protocol described in materials and methods. Cells show expression of Flag-PSMD10[Gankyrin] (in green). DAPI (in blue) was used for nuclear staining. Images were acquired in Laser confocal microscope (Zeiss LSM meta-510). (B) Cell lysate were prepared after 48 h of transduction and analyzed by WB.