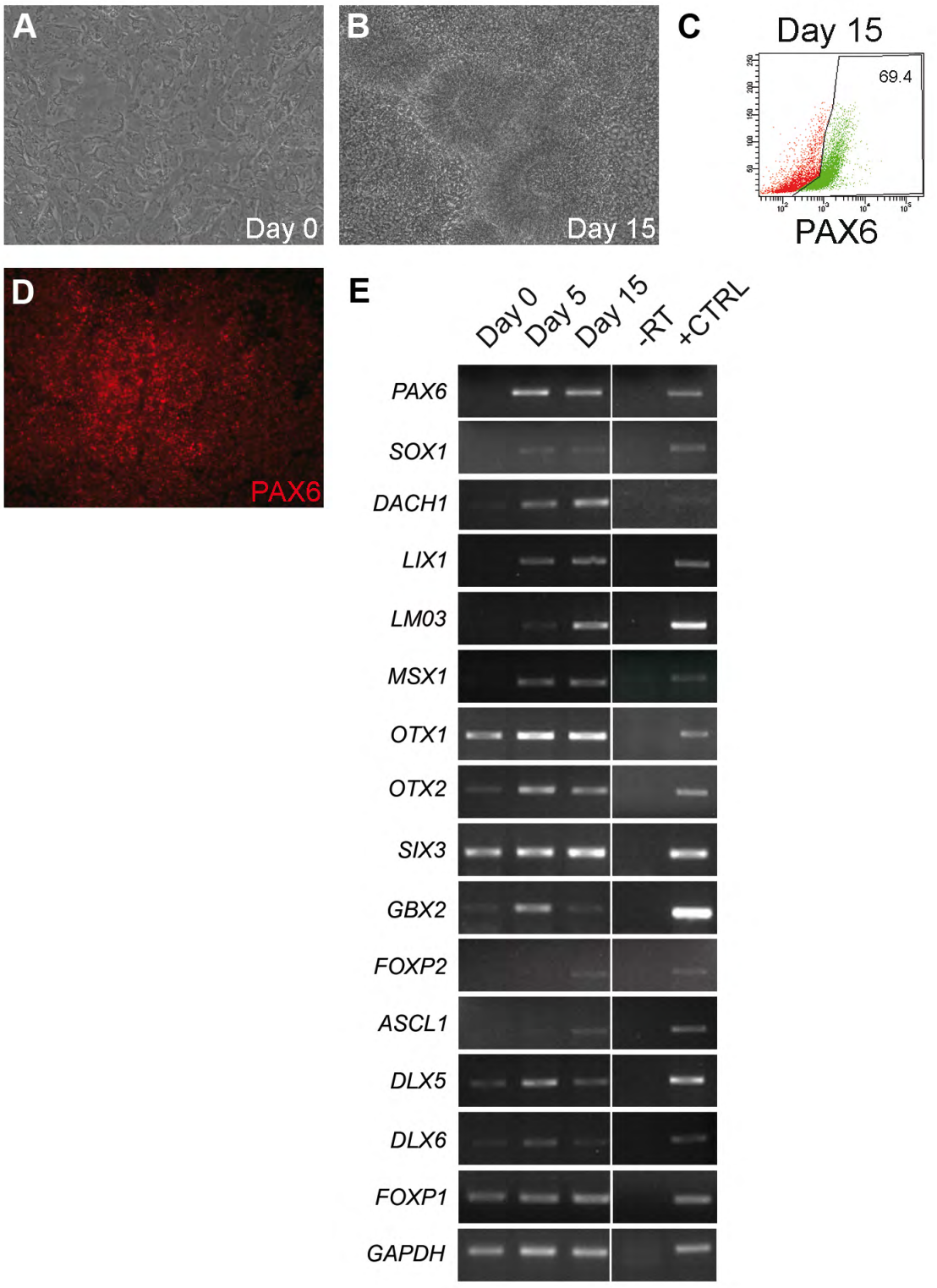

Fig. S1. Neural induction phase in the presence of noggin and SB431542. (**A**,**B**) Live images of cells during neural induction. Rosettes are evident at day 15 (B), as compared with day 0 (A) (original magnification 10×). (**C**) Flow cytometry at day 15 showing that 69.4% of the cells are PAX6^{+}. (**D**) In the presence of the noggin analog LDN-193189, the cells at day 15 express PAX6, similar to the effect of noggin and dorsomorphin exposure. (**E**) RT-PCR analysis for relevant neuroectodermal and regional markers. –RT, control without reverse transcriptase; +CTRL, cDNA from human fetal striatum. The experiments described were performed on hES H9 cell line.

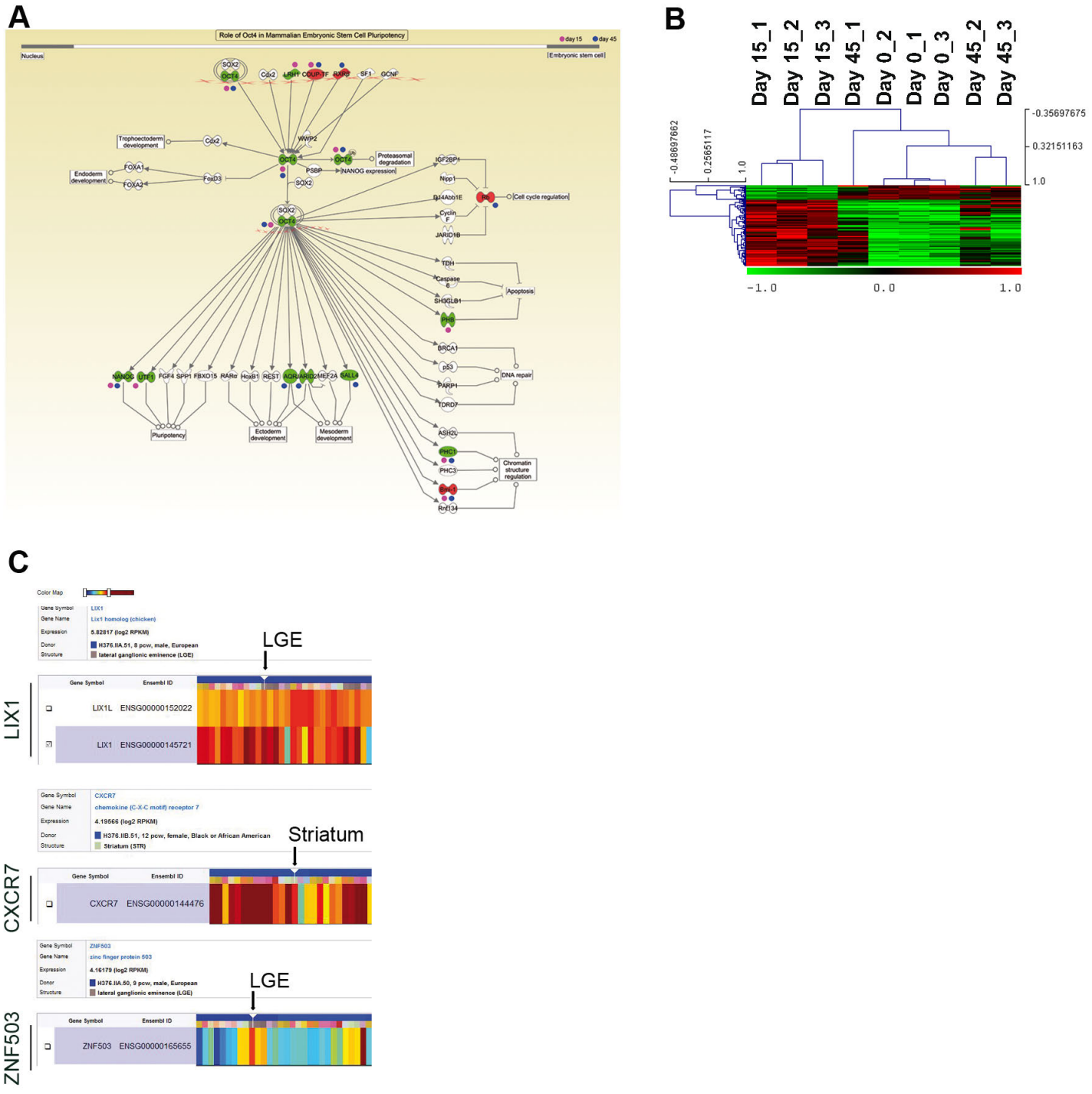

Fig. S2. Genome-wide gene expression analysis of the neural induction phase. (**A**) Ingenuity pathway analysis on day-15 and day-45 transcriptional signatures highlights significant enrichment in the OCT4-regulated pathway. (**B**) Stage-specific subcluster of the day-15 transcriptional signature (three biological replicates for each condition). (**C**) Candidate LGE-specific markers deriving from DEGs in the subcluster were online validated using the Allen human gene expression database (http://human.brain-map.org).

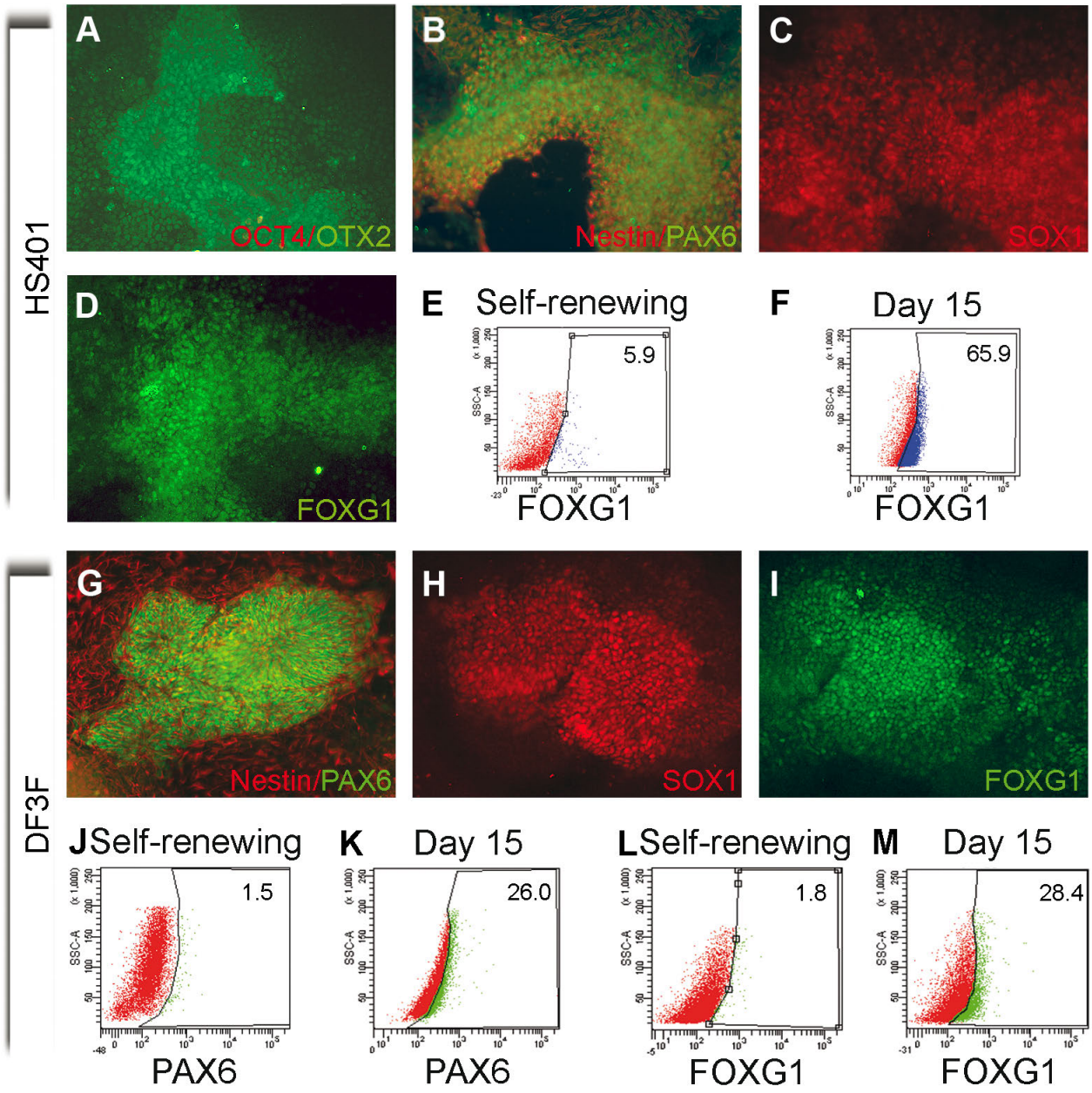

Fig. S3. Application of the neural conversion method to hES and hiPS cell lines leads to the efficient generation of ventral telencephalic progenitors. HS401 hES (A-F) and DF3F hiPS (G-M) cell lines. (**A**) OCT4 and OTX2 staining of HS401 cells at the end of neural induction, day 15. (**B**,**C**) PAX6, nestin and SOX1 are expressed in neuralized cells. (**D**) FOXG1$^+$ cells appear after the neural induction and patterning phase. (**E**,**F**) Flow cytometric analysis on generated neural progenitors. At day 15 of differentiation, 65.9% of the cells expressed FOXG1. (**G**,**H**) The neuroectodermal markers nestin, PAX6 and SOX1 are expressed on day 15 of DF3F differentiation. (**I**) FOXG1 expression revealed the emergence of telencephalic patterned progenitors (original magnification 20×). (**J-M**) Flow cytometric analysis during neural conversion for PAX6 and FOXG1. At day 15, 26% of cells are PAX6$^+$ and 28.4% are FOXG1$^+$.

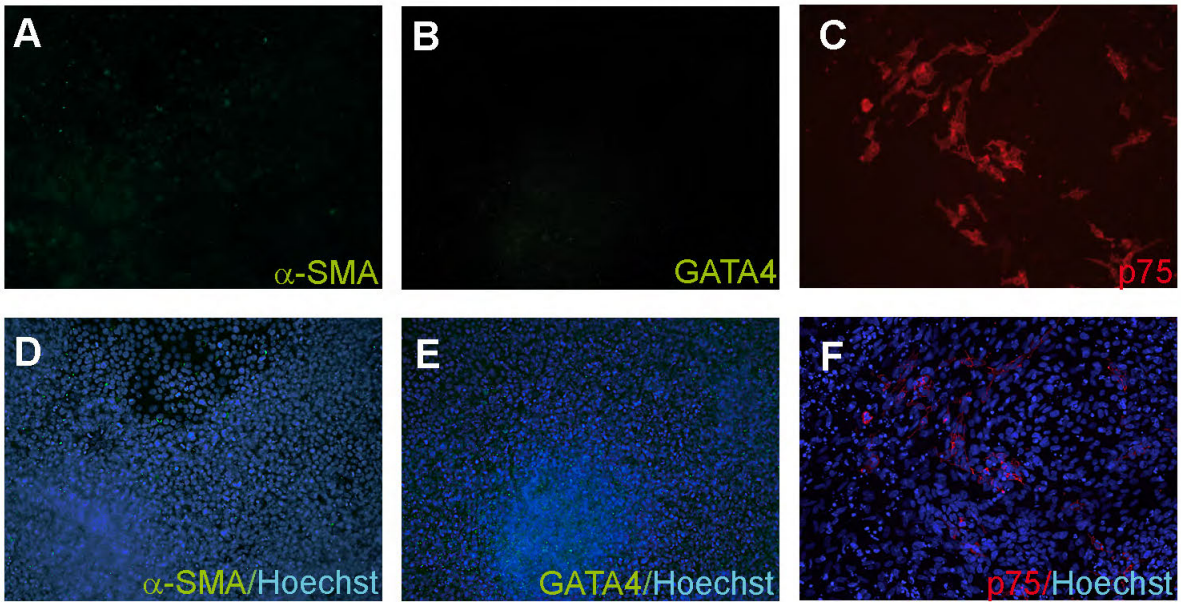

Fig. S4. Treatment with dorsomorphin and SB431542 selectively induces neuroectodermal fate. Lack of the mesodermal (α-SMA) (**A**,**D**) and endodermal (GATA4) (**B**,**E**) markers at day 15 of differentiation. Some cells are positive for the neural crest marker p75 (p75NTR; NGFR – Human Gene Nomenclature Committee) (**C**,**F**) after neural conversion at 15 days of differentiation. The experiments described were performed on hES H9 cell line.

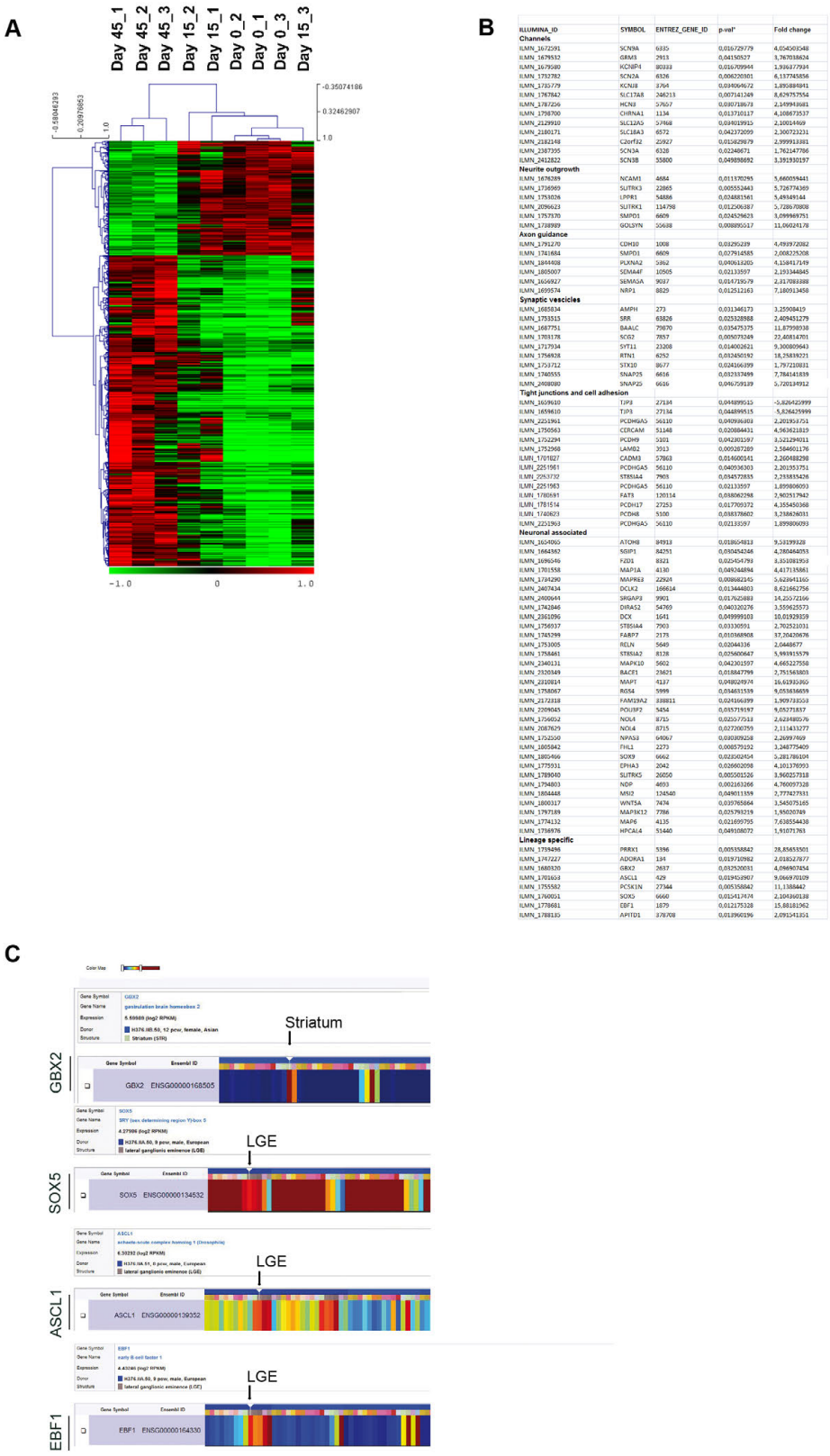

Fig. S5. Genome-wide gene expression analysis of the terminal differentiation phase. (**A**) Stage-specific subcluster of the day-45 transcriptional signature (three biological replicates for each condition). (**B**) Summary including a selected list of day-45-specific differentially expressed transcripts. (**C**) Candidate striatal-specific markers present in the day-45 transcriptional subcluster were online validated using the Allen human gene expression database (http://human.brain-map.org).

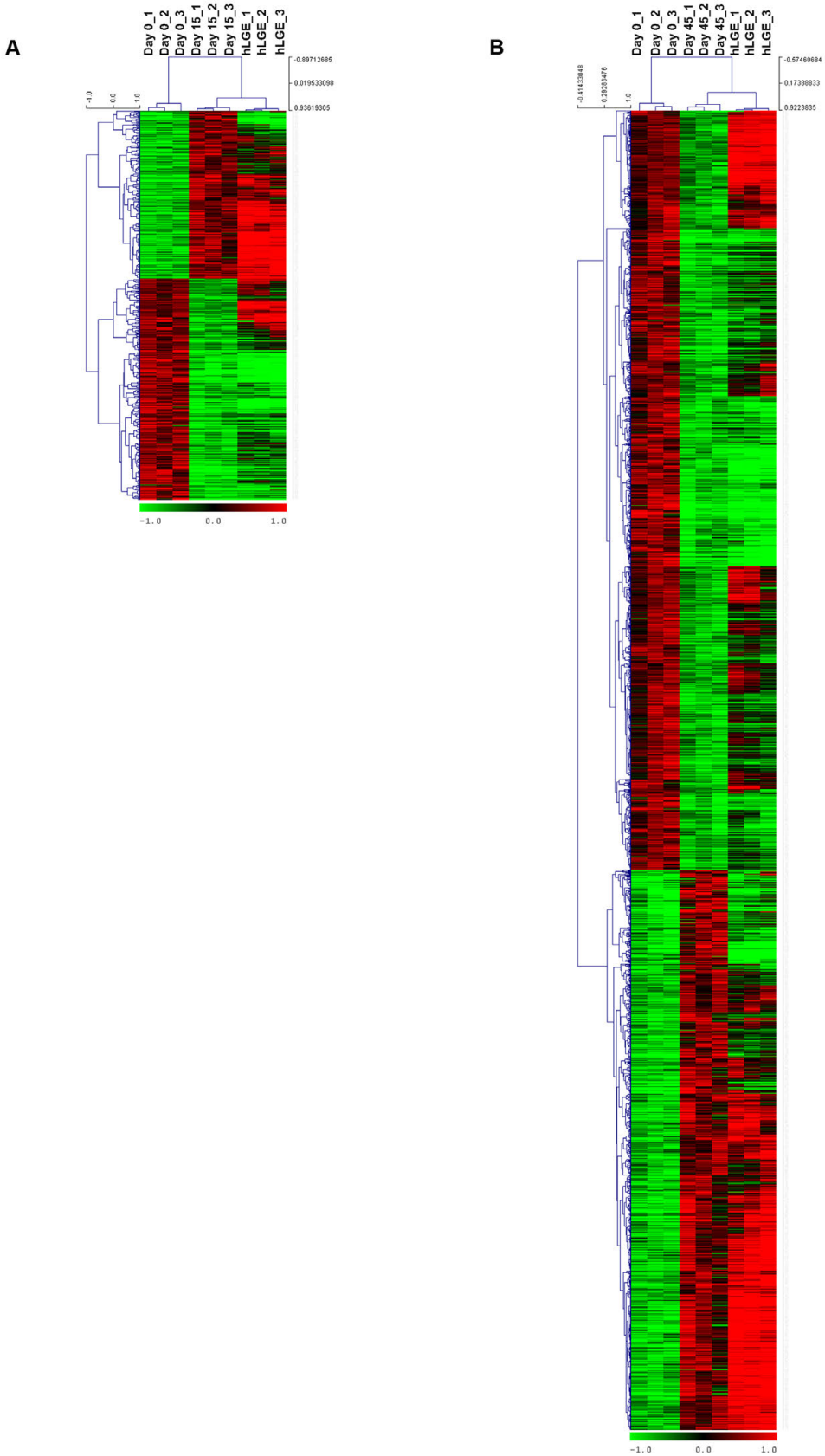

Fig. S7. Neurochemical investigation of neuronal subtypes differentiated from hES cells. (**A**,**B**) Immunofluorescence for MAP2ab, βIII-tubulin, nestin (A) and GFAP (B) at day 80 of differentiation. (**C**) The composition of the total cell population: 51±3% MAP2^{+} cells, *n*=1126 cells; 25±0.03% GFAP^{+} cells, *n*=712 cells; 7±4.9% nestin^{+} cells, *n*=220 cells; 17.3±5.03% nestin/βIII-tubulin double-positive cells, *n*=208 cells. (**D**,**E**) Co-expression of GABA/CTIP2 (D) and CALB1/CTIP2 (E). (**F**,**G**) Analysis of the interneuronal markers neuropeptide Y (NPY) and calretinin (CR) (F) and parvalbumin (PVALB) and somatostatin (SST) (G) at day 80. (**H**) Tyrosine hydroxylase (TH) and 5-hydroxytriptamine (5-HT) immunoreactive neurons at the end of terminal differentiation. (**I**) Immunodetection for vesicular glutamate transporter 1 (VGLUT1). The experiments described were performed on hES H9 cell line.

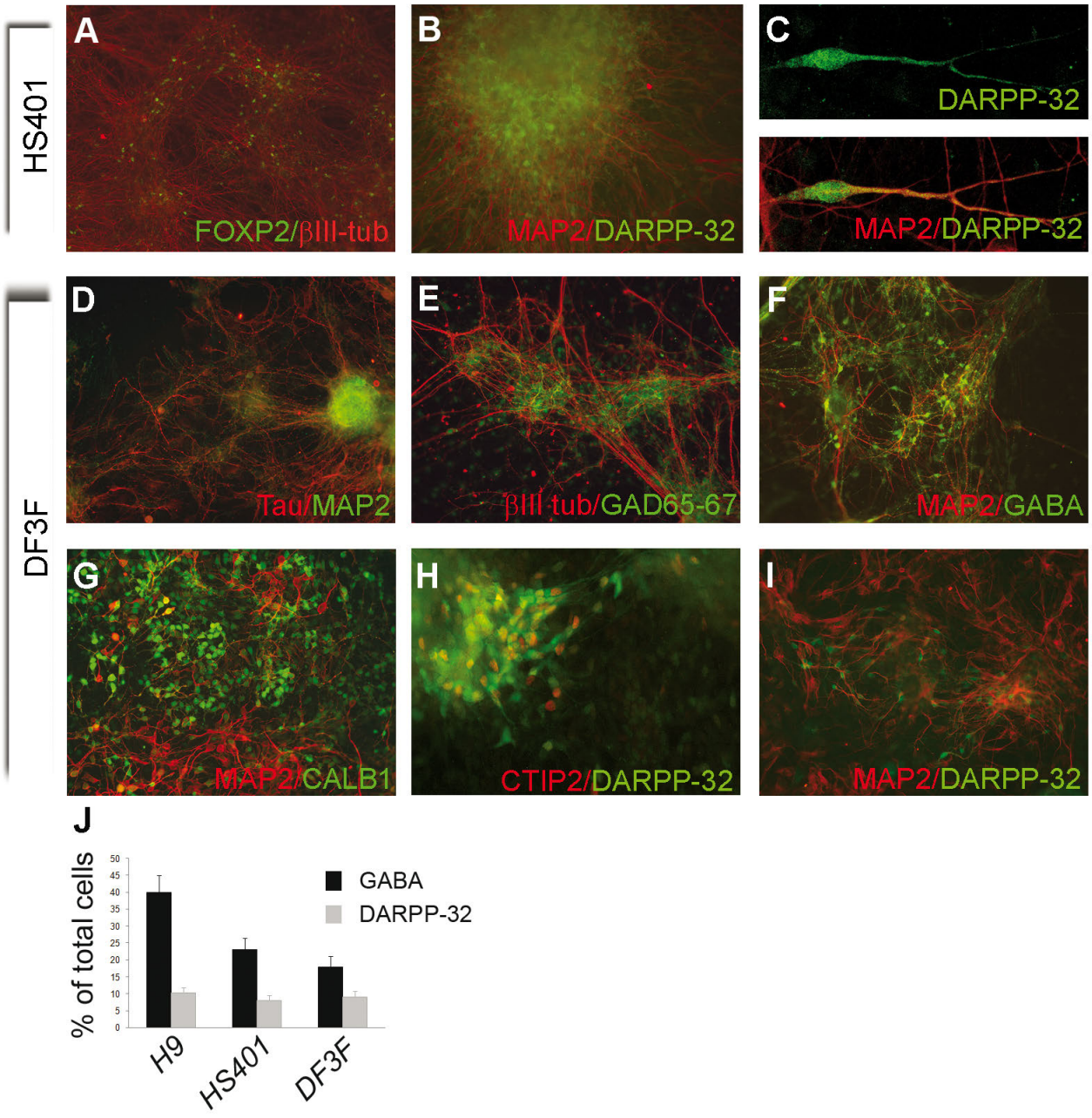

Fig. S8. HS401 and DF3F cells can also be patterned and differentiated to striatal neurons according to the established protocol. (A-C) HS401 cells; (D-I) DF3F cells. (**A**) Immunodetection of FOXP2 in differentiated HS401-derived neurons at day 45. (**B**) After terminal differentiation, 8.1±1% of the cells express DARPP-32 (*n*=1820 cells) (original magnification 20×). (**C**) Confocal analysis revealing the morphology of DARPP-32^{+} neurons (original magnification 40×). (**D-I**) Terminally differentiated DF3F-derived neurons express the mature pan-neuronal markers MAP2ab and TAU (D), the GABAergic markers GAD65/67 (E), GABA (F), calbindin (CALB1) (G), and the striatal markers CTIP2 (H) and DARPP-32 (I) (9.1±1.6% of the total cells, *n*=2145 cells; mean ± s.d.) (original magnification 20×). (**J**) A comparative assessment of the percentage of GABA^{+} and DARPP-32^{+} neurons among H9, HS401 hES cells and DF3F hiPS cell lines. HS401: GABA 23±3%, *n*=2226 cells; DARPP-32 8.1±1%, *n*=1820 cells. DF3F: GABA 17.8±3%, *n*=1153 cells; DARPP-32 9.1±1.6%, *n*=2145 cells (mean ± s.d.).

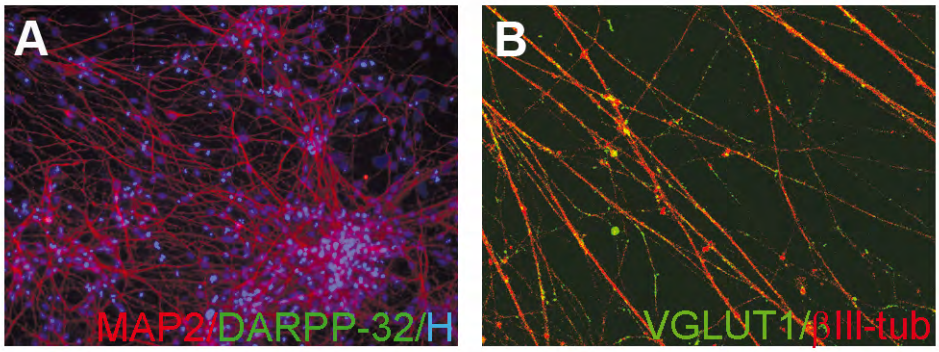

Fig. S9. Neuronal differentiation analysis in the absence of SHH/DKK1 morphogens. At the end of the differentiation the cells do not show positivity for DARPP-32 (**A**), whereas they widely express the glutamatergic marker VGLUT1 (**B**). The experiments described were performed on hES H9 cell line.

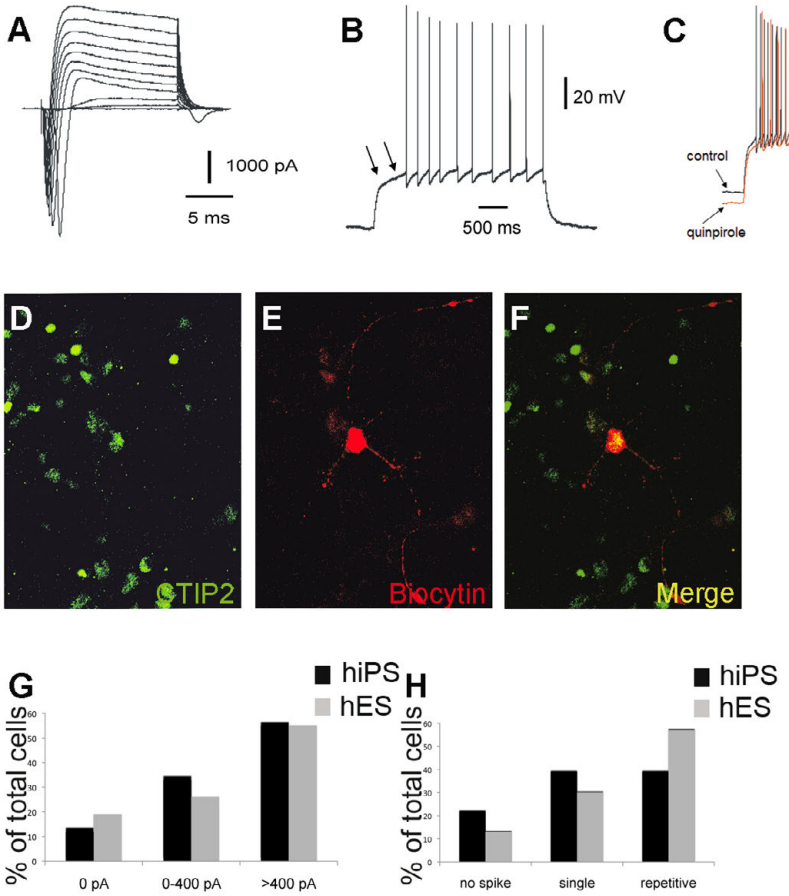

Fig. S10. Electrophysiological properties of fully differentiated hiPS-derived neurons. (**A**) Family of total inward and outward currents elicited at test potentials ranging from –70 to +40 mV from a holding voltage of –90 mV. (**B**) Sample trace obtained in current-clamp configuration showed repetitive firing in a 73-day differentiated neuron when excited with a suprathreshold depolarizing step of current. Note that the onset delay in the generation of the first spike (arrows) is a typical electrophysiological property of mature MSNs. (**C**) The application of quinpirole (5 μM), a D2 agonist, in two out of four cells induced hyperpolarization of the membrane potential. (**D-F**) Confocal image of double labeling of a recorded cell with biocytin and for CTIP2 (original magnification 40×). (**G**) The fraction of cells (hiPS and hES cells) subdivided into three groups according to the amplitude of the Na^+ current peak. hiPS cells: 0 pA, 12% (*n*=2 cells); <400 pA (315±25, mean ± s.e.m.), 33% (*n*=6 cells); >400 pA (–1746±481), 55% (*n*=10 cells). hES cells: 0 pA, 19% (*n*=11 cells); <400 pA (–218±25.00 pA), 26% (*n*=15 cells); >400 pA (–1750±250 pA), 55% (*n*=32 cells). (**H**) The fraction of cells (hiPS and hES cells) subdivided into three groups according to their firing properties. hiPS cell-derived neurons: no spike, 22% (*n*=4 cells); single spike, 39% (*n*=7 cells); repetitive firing, 39% (*n*=7 cells). hES cell-derived neurons: no spike, 13% (*n*=3 cells); single spike, 30% (*n*=7 cells); repetitive firing, 57% (*n*=13 cells). The experiments described were performed on the hiPS DF3F cell line compared with the hES H9 cell line.

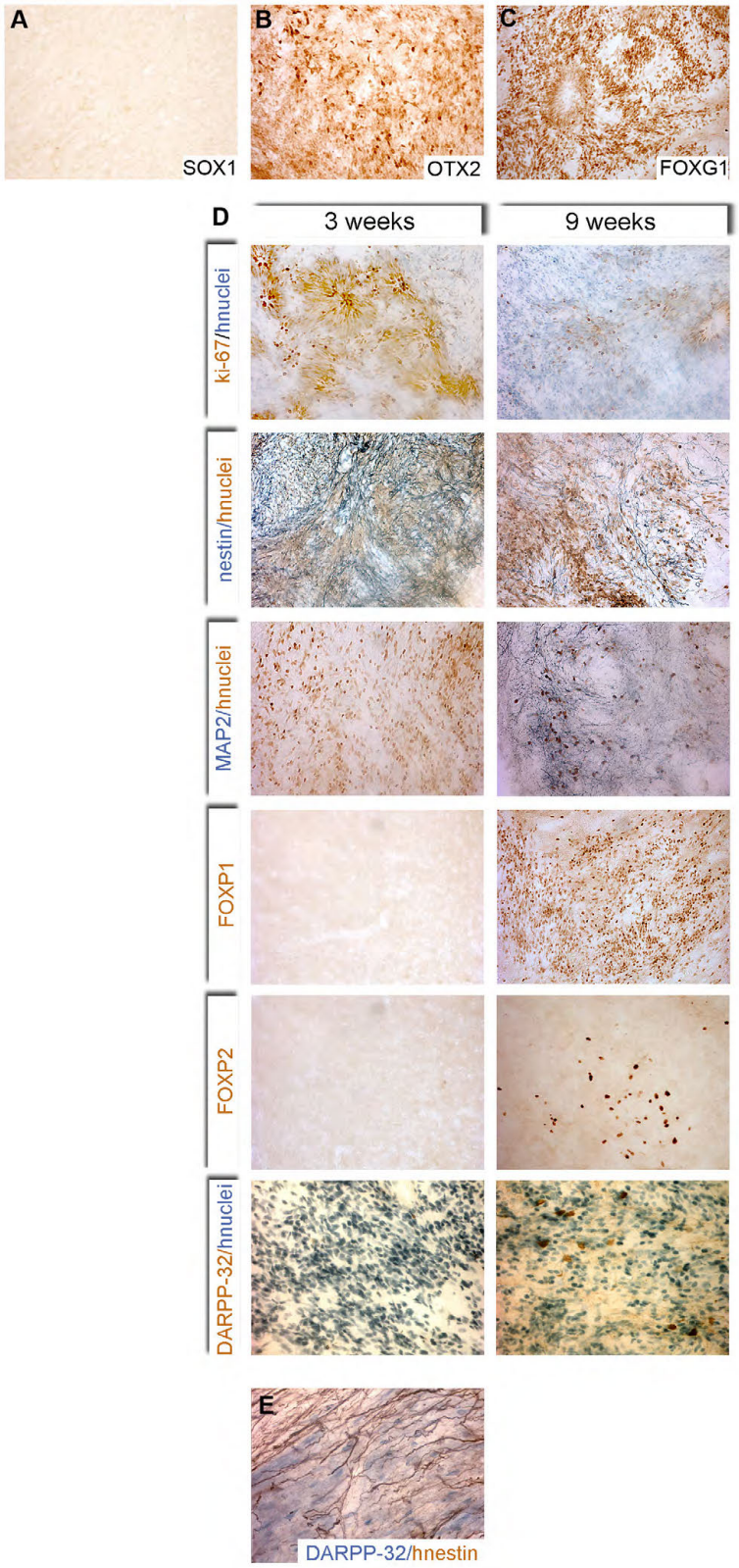

Fig. S11. Characterization of transplanted striatal precursors in striata from HD rats. (**A**) Immunohistochemical staining of a 9-week-old graft reveals no evidence of SOX1 expression. (**B**,**C**) OTX2 and FOXG1 immunostaining. (**D**) Timecourse analysis of proliferating (Ki-67, nestin), differentiated neurons (MAP2ab), and striatal neurons (FOXP1, FOXP2, DARPP-32) throughout the analyzed time points. Ki-67$^+$ cells: at 3 weeks, 56.38±7.72%; at 9 weeks, 22.55±3.02% of the hnuclei$^+$ cells; n=3 brains/time point. FOXP1: at 3 weeks, no cells; at 9 weeks, 18.49% of hnuclei$^+$ cells; n=1 brain/time point. FOXP2: at 3 weeks, no cells; at 9 weeks, 0.56% of hnuclei$^+$ cells; n=1 brain/time point. DARPP-32: at 3 weeks, no cells; at 9 weeks, 0.017±0.002% of hnuclei$^+$ cells; n=3 brains/time point. Percentage expressed as mean ± s.e.m. (**E**) Nestin-labeled projections (brown) were evident in the intact DARPP-32$^+$ striatum (blue). The experiments described were performed on hES H9 cell line.

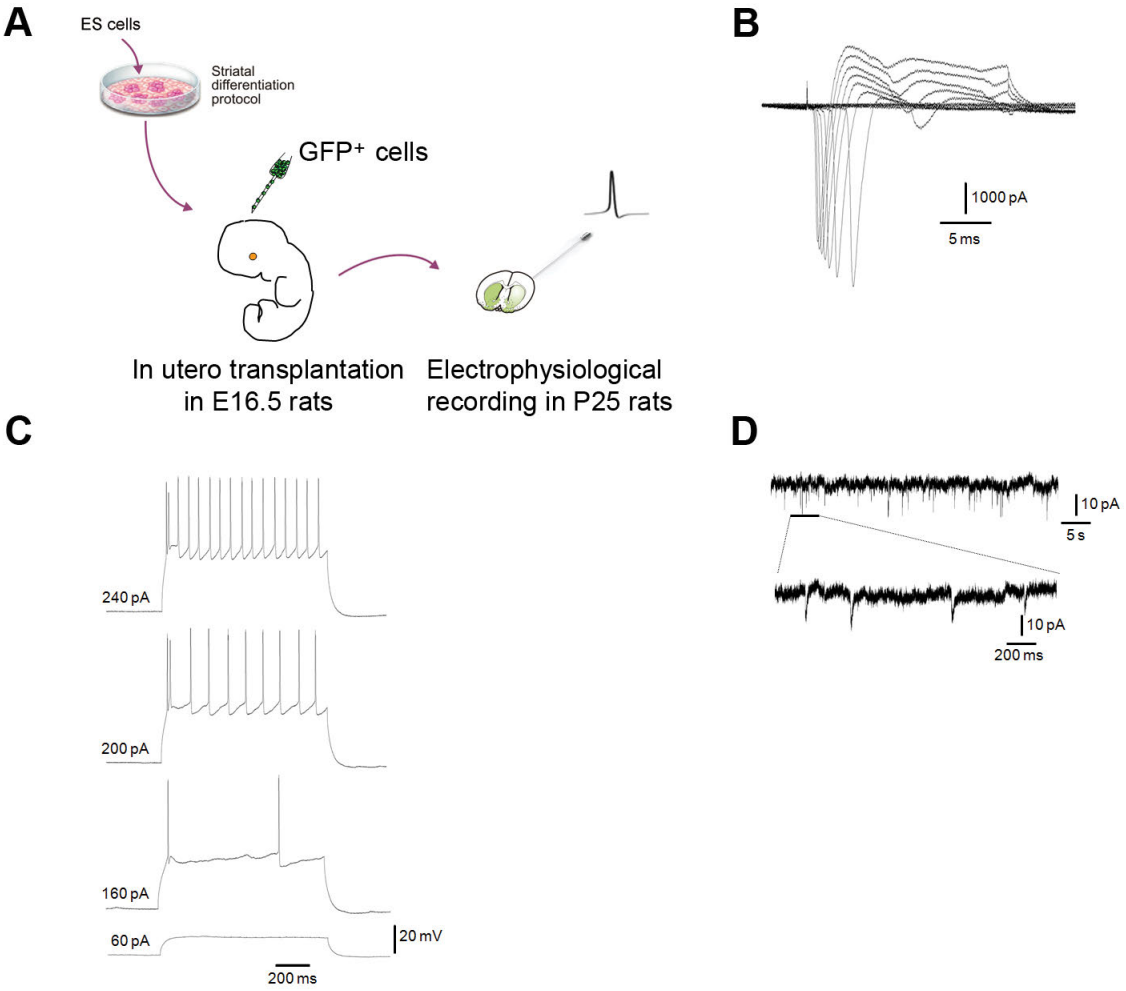

Fig. S12. ***In utero*** **transplanted precursors functionally mature and integrate into the rat brain.** (**A**) Schematic of the transplantation protocol in E16.5 rats. (**B**) I/V relationship. (**C**) Family of sub- and suprathreshold depolarizing steps from the resting potential of –73 mV evoked by injecting rectangular pulses of depolarizing current. Stimulus size is indicated on the left. The cell shows a high frequency of action potentials and a marked after-hyperpolarization. (**D**) Spontaneous postsynaptic currents recorded in voltage-clamp mode at the holding potential of –70 mV.

Table S1. Primer sequences, annealing temperatures and amplicons

Gene	Forward primer	Reverse primer	Ta (°C)	Amplicon (bp)
GAPDH	AGCTGAACGGGAAGCTCACT	AGGTCCACCACTGACACGTTG	60	67
PAX6	TCCATCAGTTCCAACGGAGAA	GTGGAATTGGTTGGTAGACAC	60	337
DACH1	GTGGAAAACACCCCTCAGAA	CTTGTTCCACATTGCACACC	57	208
LIX1	ATGAGTCACTGCCAGCTCCT	GTGGAGGCTACTGCTTCCTG	59	217
LMO3	GGGCTCCACCCTGTACACTA	TAGTCCGTCTGGCAAAGGAT	57	243
MSX1	CCTTCCCTTTAACCCTCACAC	CCGATTTCTCTGCGCTTTTCT	58	285
OTX1	CGTTCACAGCTGGACGTG	CTTTCGGAGCCCGAGCTC	60	230
OTX2	TCAACTTGCCCGAGTCGAGG	CAATGGTCGGGACTGAGGTG	61	204
SIX3	CCGGAAGAGTTGTCCATGTT	CGACTCGTGTTTGTTGATGG	57	171
GBX2	CTCGCTGCTCGCCTTCTC	GCCAGTCAGTCAGATTGTCATCCG	62	173
EMX2	GGGATCCGTCCACCTTCTAC	CTCAAAGGCGTGTTCCAGCC	61	374
FOXP2	GCTTTGACAGTGAGCTAGCTTC	CTGGTGTCACCACTTGATCTTC	58	280
ASCL1	GTCCTGTCGCCCACCATCTC	CCCTCCCAACGCCACTGAC	64	215
FOXG1	TGTTGACTCAGAACTCGCTGG	CTGCTCTGCGAAGTCATTGAC	60	262
DLX5	TTCAGAAGACTCAGTACCTCGC	GAGTTACACGCCATTGGGTC	60	184
DLX6	TACCTCCAGTCCTACCACAAC	AATAAATGGTCCGAGGCTTCCG	60	145
FOXP1	CTACCGCTTCCATGGGAAATC	CTGTTGTCACTAAGGACAGGG	59	207
ISL1	TACAGGCTAACCCAGTGGAAG	GACTGGCTACCATGCTGTTAG	60	207
DrD1	AGGGACATGTCTTTGGCTTCAG	GGGAACAGTGTTAGCACCTGTT	60	173
ARPP21	GTGCAAAGCGTGATGGTTTCC	CCTTGACCTGCCTGGTTAGG	58	128
DARPP-32	CTGAGGACCAAGTGGAAGAC	GATGTCCCCTCCACTTCCTC	58	125
NKX2-1	ACCGGGTTCAGACTCAGTTC	ATCGACATGATTCGGCGTCGG	60	221
EAR	GAGGCTGAGGCAGGAGAATCG	GTCGCCCAGGCTGGAGTG	60	88

Table S2. Antibodies used for immunofluorescence, immunohistochemistry and flow cytometry

Application	Antibody	Dilution	Supplier
Immunofluorescence	OCT4	1:100	Santa Cruz
	OTX2	1:500	Chemicon
	α smooth muscle actin	1:800	Sigma
	GATA4	1:200	Santa Cruz
	p75	1:200	Santa Cruz
	PAX6	1:200	Covance
	nestin	1:200	R&D
	SOX1	1:200	Santa Cruz
	FOXG1	1:1000	StemCulture
	GSX2	1:2000	Gift from Prof. K. Campbell, Cincinnati Children's Hospital Medical Center
	βIII-tubulin	1:1000	Sigma
	MAP2ab	1:500	BD Bioscience
	calbindin	1:200	SWANT
	GABA	1:500	Sigma
	GAD65/67	1:200	Millipore
	FOXP1	1:1000	Abcam
	FOXP2	1:2000	Abcam
	CTIP2	1:500	Abcam
	DARPP-32	1:200	Epitomics
	DRD2	1:200	Millipore
	A2A	1:1000	Upstate
	Synaptophysin	1:200	Sigma
	5-HT	1:500	Sigma
	TH	1:200	Immunological Science
	VGLUT1	1:300	Millipore
	GFAP	1:1000	DAKO
	calretinin	1:200	BD Bioscience
	NPY	1:5000	Immunostar
	parvalbumin	1:500	Chemicon
	somatostatin	1:100	Millipore
	ChAT	1:200	Chemicon
Immunohistochemistry	Human nuclei	1:1000	Millipore
	FOXG1	1:1000	StemCulture
	Human nestin	1:500	Neuromics
	MAP2ab	1:500	Sigma
	Ki67	1:500	DAKO
	N-cadherin	1:100	BD Bioscience
	ZO-1	1:100	Invitrogen
	OTX2	1:1000	Chemicon
	FOXP1	1:500	Abcam
	DARPP-32	1:200	Epitomics
	DARPP-32	1:30,000	Gift from Prof. Hemmings, Cornell University
	α smooth muscle actin	1:500	Sigma
	SOX1	1:150	Santa Cruz
	OCT4	1:100	Santa Cruz
	FOXP2	1:2000	Abcam
	GFAP	1:4000	DAKO
Flow cytometry	OCT4	1:100	Santa Cruz
	OTX2	1:1000	Chemicon
	PAX6	1:5000	Hybridoma Bank
	FOXG1	1:1000	StemCulture

Table S3. Composition of solutions used for electrophysiological recording

Solution	Application	Composition (mM)
1	EXTRACELLULAR: Total currents and voltage signal (neurons in culture)	140 NaCl, 3 KCl, 10 glucose, 10 HEPES, 2 $CaCl_2$, 1 $MgCl_2$ (pH 7.35 with NaOH)
2	INTRACELLULAR: Total currents and voltage signal (neurons in culture)	130 K-gluconate, 4 NaCl, 2 $MgCl_2$, 1 EGTA, 10 HEPES, 5 CP (phosphocreatine), 2 Na_2-ATP, 0.3 Na_3-GTP (pH 7.25 with KOH)
3	INTRACELLULAR: Post-synaptic currents (neurons in culture)	135 CsCl, 3 NaCl, 10 EGTA, 10 HEPES, 0.5 $CaCl_2$, 1 $MgCl_2$, 4 Na_2-ATP, 0.3 Na_3-GTP (pH 7.25 with CsOH)
4	EXTRACELLULAR: Slice preparation (bubbled with 95% O_2/5% CO_2)	70 sucrose, 80 NaCl, 2.5 KCl, 26 $NaHCO_3$, 15 glucose, 1 $CaCl_2$, 7 $MgCl_2$, 1.25 NaH_2PO_4
5	EXTRACELLULAR: aCSF slices (bubbled with 95% O_2/5% CO_2)	125 NaCl, 2.5 KCl, 26 $NaHCO_3$, 15 glucose, 1.3 $MgCl_2$, 2.3 $CaCl_2$, 1.25 $NaHPO_4$