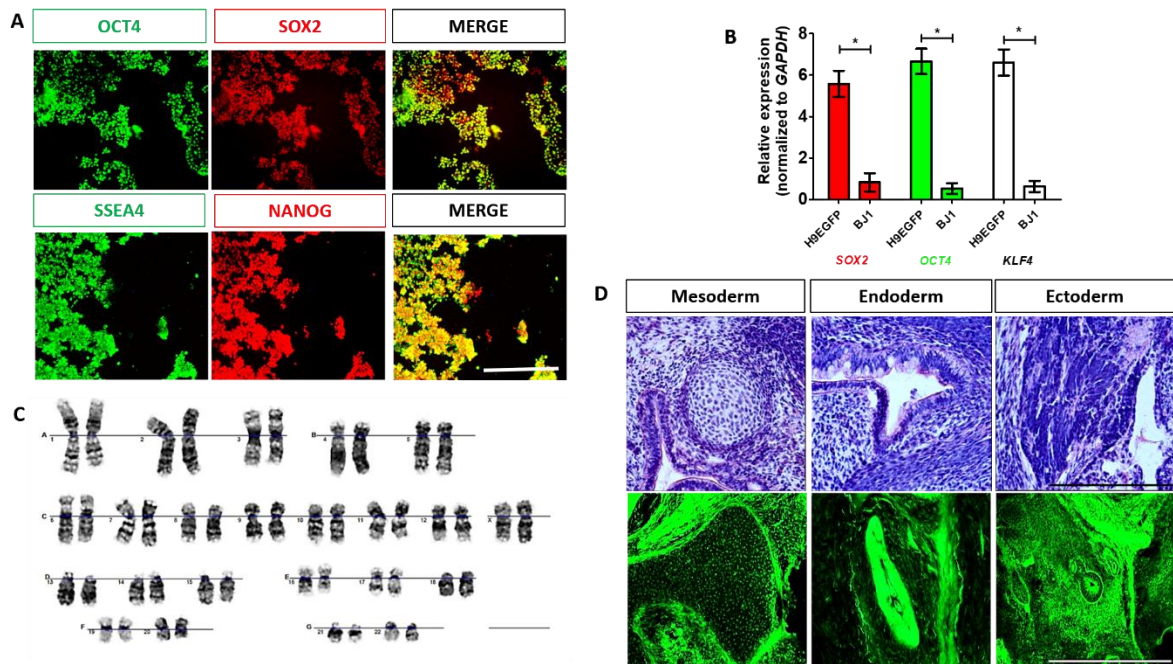


Supplementary Figures



Supplementary Figure 1.- Pluripotency of cell lines H9 and H9-EGFP. A)

Immunocytochemical analysis on H9 cell line. Scale bar, 200 μ m. B) Gene expression, by RT-

qPCR, for *SOX2*, *OCT4*, and *KLF4* on H9-EGFP cell line. Negative controls are BJ1 human

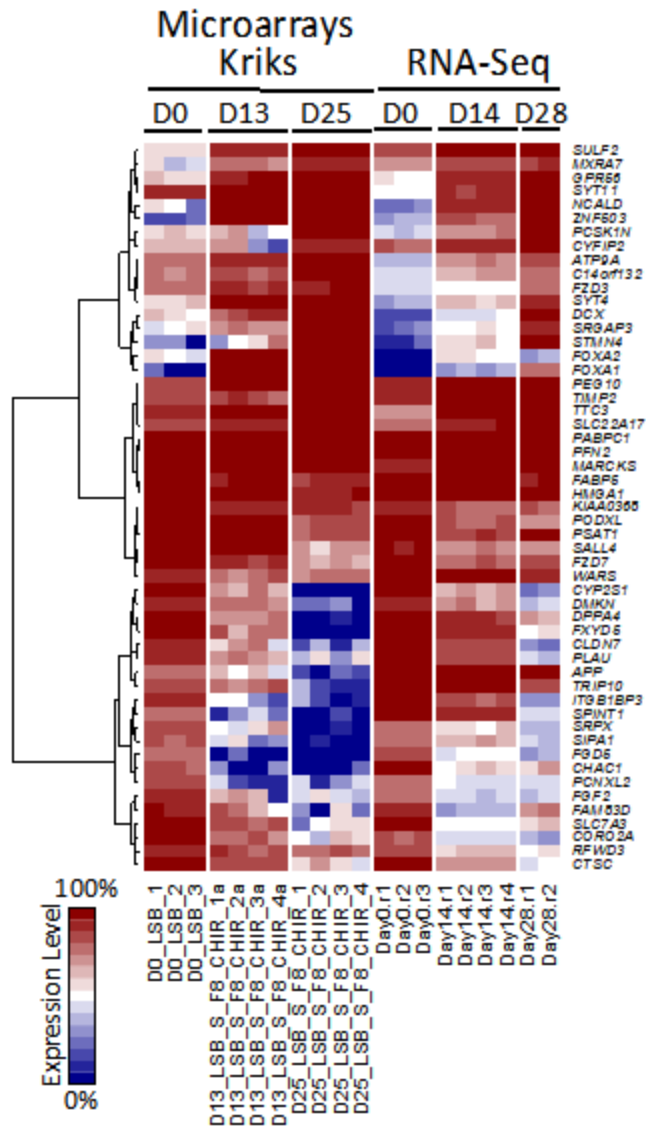
fibroblasts. C) H9-EGFP cell line presents a normal female karyotype (46XX). D) Teratoma

formation after H9-EGFP subcutaneous inoculation where cells representative of the three

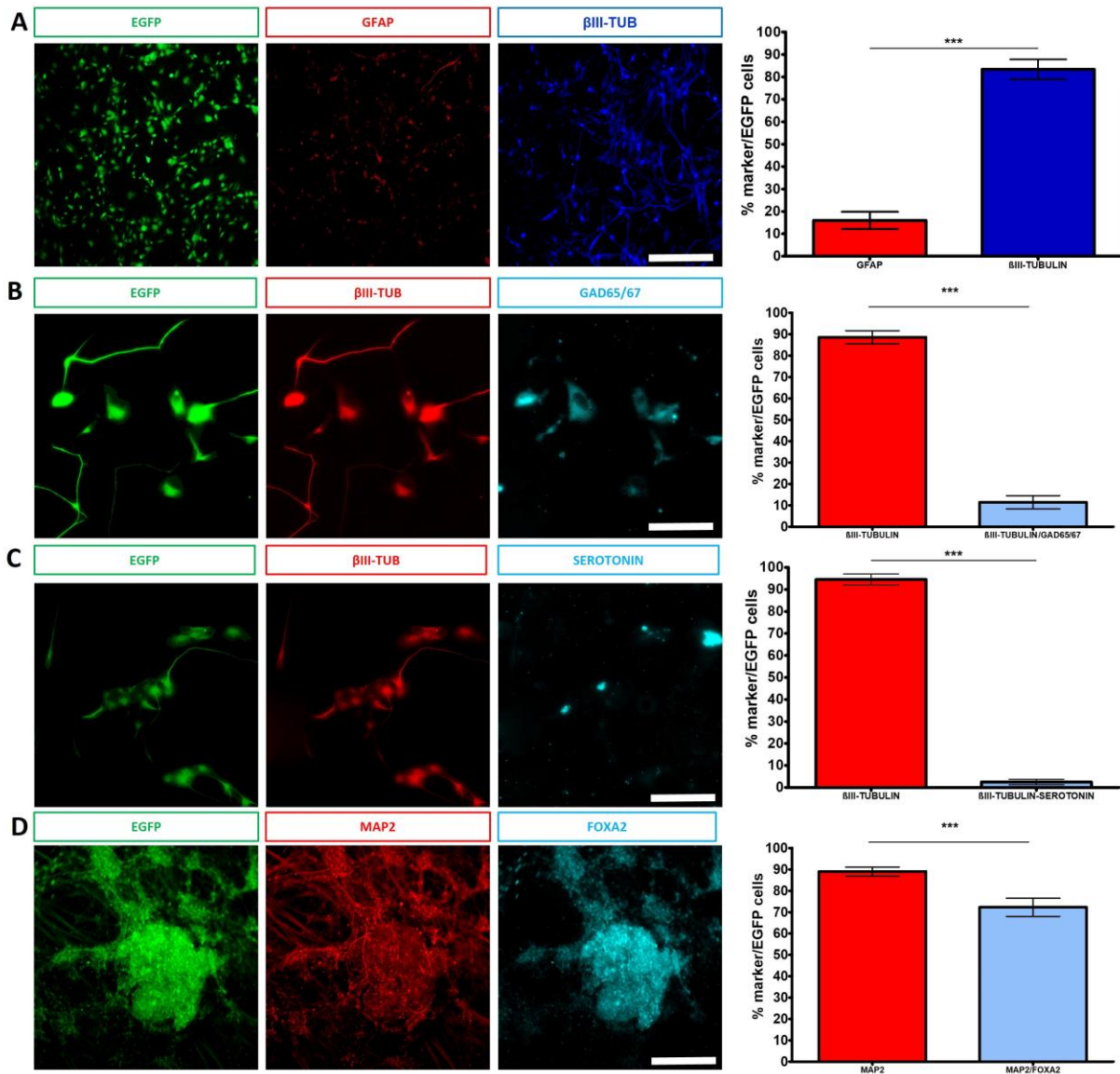
embryonic germ layers can be identified by hematoxylin and eosin staining (upper panel). The

lower panel shows adjacent slices to observe EGFP. Mean \pm SEM; * P < 0.05, n=3 independent

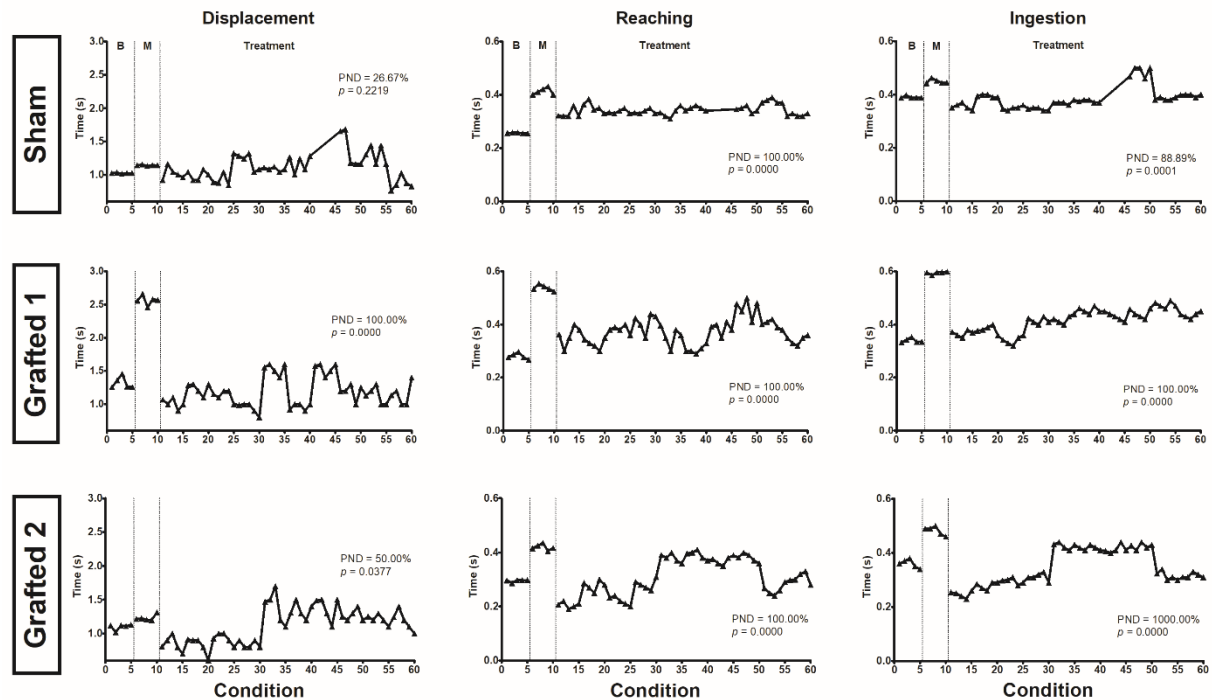
experiments. Scale bar, 300 μ m.



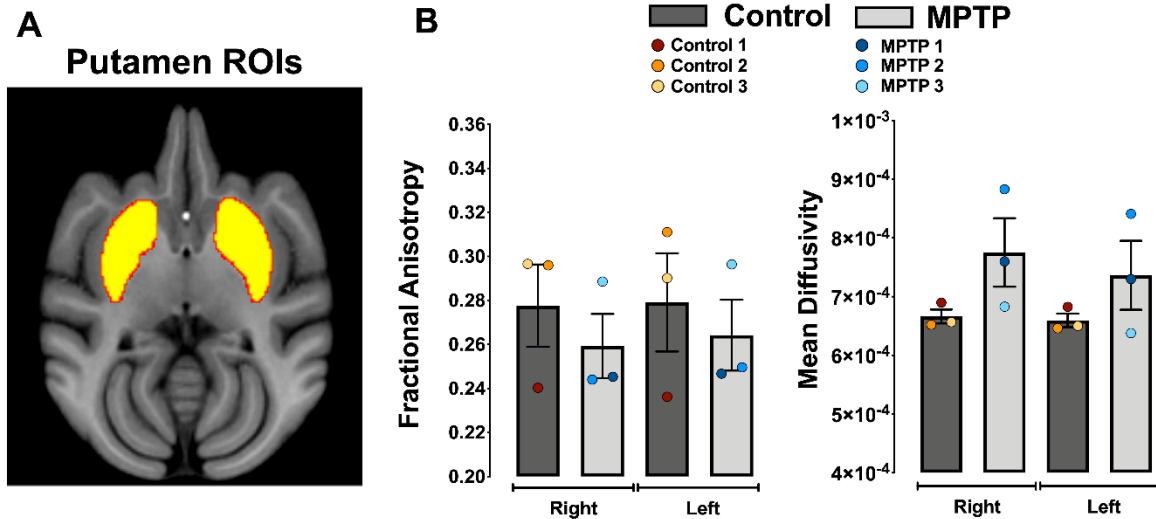
Supplementary Figure 2.- Comparison of expression data at similar time points from previous work with our RNA-Seq results. The microarray results from Kriks et al. [12] were aligned with the reads of the same genes found in our sequencing experiments [40]. Although the times are not the same (day 13 vs day 14 and day 25 vs day 28), the expression patterns are similar, indicating that our differentiation proceeded as reported for the floor-plate induction of dopamine neurons.



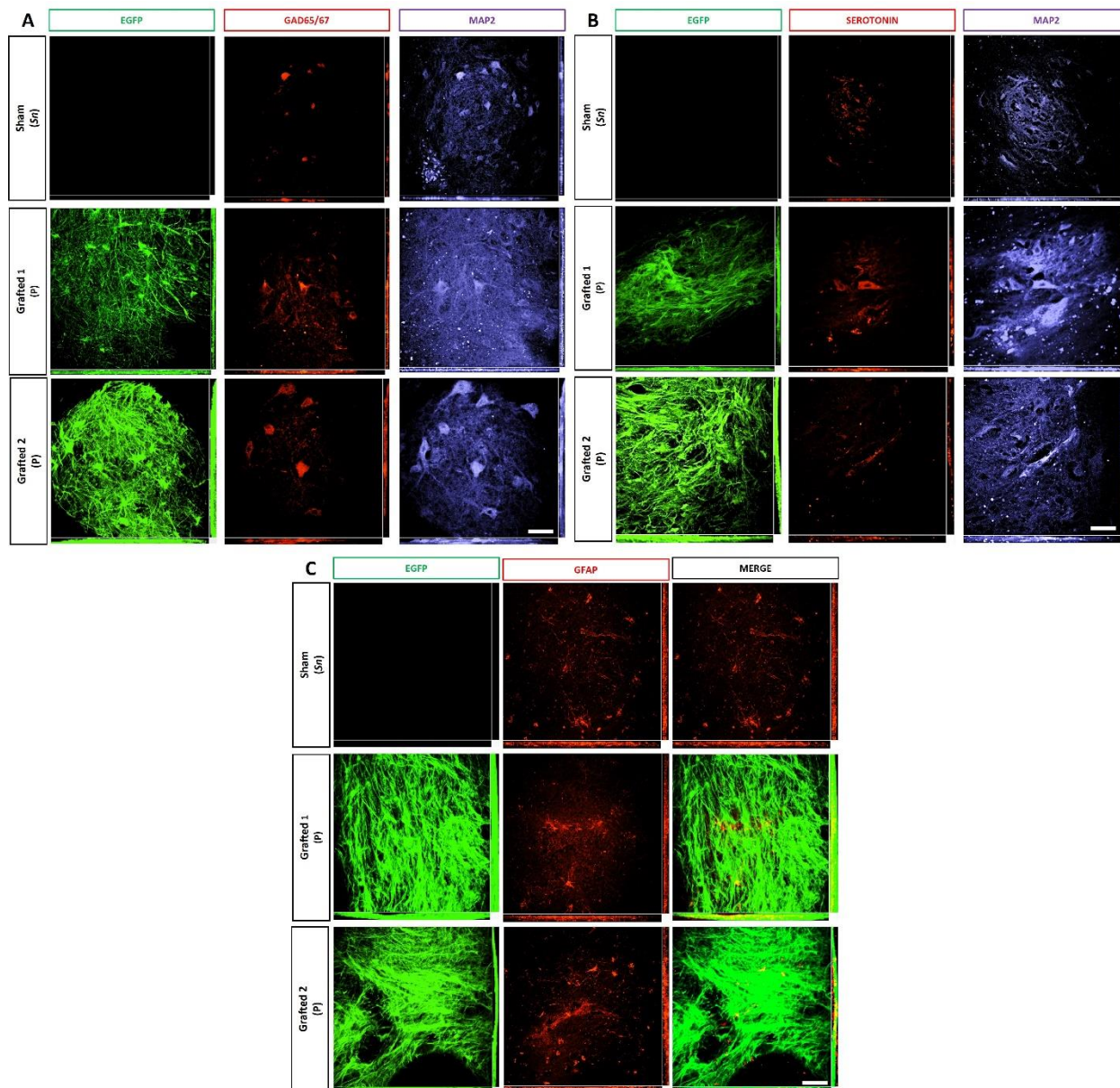
Supplementary Figure 3.- Cell phenotypes obtained through the differentiation protocol. A) Immunocytochemistry at day 21 for expression of EGFP, GFAP, and βIII-TUBULIN (βIII-TUB) Scale bar, 300 μm. **B and C)** Expression of βIII-TUB, GAD65/67, and Serotonin at day 28. Scale bar, 100 μm. **D)** Immunodetection of FOXA2 and MAP2 at day 35. Scale bar, 200 μm. Mean ± SEM; *** $P < 0.001$, $n=3$ independent experiments.



Supplementary Figure 4.- Single-case design for each task and every NHP. Every graph has the values for basal (B), MPTP (M), and the post-sham or -grafting surgery times up, to 10 months (Treatment) in the 3 motor tasks. The values for the 5 consecutive days of the tests are represented for B, M, and each individual month. The 3 top graphs correspond to Sham, the middle panels to Grafted 1, and the bottom are Grafted 2. The subtraction between MPTP and the average of all 10-month times was employed to compare the means (Fig 2D, bottom part). PND, Percentage of Nonoverlapping Data (Treatment vs M).



Supplementary Figure 5.- MPTP intoxication of three NHPs diminishes Fractional Anisotropy (FA) and increases Mean Diffusivity (MD) in the putamen compared to healthy subjects. A) MRI template and region of interest (ROI, in yellow) measured in each putamen onto anatomical brain image. FA and MD were determined bilaterally in the putamen. Three healthy NHPs were used for Control (non-parkinsonian) and three for the MPTP group. **B)** Graphs show FA and MD's comparison between the Control and MPTP-treated groups in the right and left putamina. Mean \pm standard error. Two-tailed Mann-Whitney test. No significant differences were found.



Supplementary Figure 6.- Cell phenotypes found in the grafts.- A) Surviving cells in the substantia nigra (Sn) and co-expression of GAD65/67 (red), MAP2 (cyan), and EGFP (green) by grafted neurons in the putamen (P) after ten months. B) Co-expression for Serotonin (red), MAP2 (cyan), and EGFP (green). C) GFAP (red) and EGFP expression. Scale bar, 50 μ m.

55 **Supplementary Table 1.- Surgical coordinates**

56

		Sham						Grafted 1						Grafted 2					
Graft		Left (cm)			Right (cm)			Left (cm)			Right (cm)			Left (cm)			Right (cm)		
A	L	0.9			-0.95			0.9			-1			1.2			-1		
	AP	-1.85			-1.85			-2.2			-2.2			-1.3			-1.3		
	V	-	-	-	-	-	-	-	-	-	-	-	-	-	-2	-	-	-2	-
		1.64	1.44	1.25	1.84	1.64	1.45	2.2	1.9	1.6	2.2	1.9	1.6	2.3		1.8	2.3		1.8
M	L	1.05			-1.1			1			-1.1			1.3			-1.1		
	AP	-1.65			-1.65			-2			-2			-1			-1		
	V	-	-	-	-	-	-	-	-2	-	-	-2	-	-	-2	-	-	-2	-
		1.64	1.44	1.25	1.84	1.64	1.45	2.2		1.8	2.2		1.8	2.3		1.8	2.3		1.8
P	L	1.2			-1.25			1.1			-1.2			1.4			-1.3		
	AP	-1.36			-1.36			-1.8			-1.8			-0.8			-0.8		
	V	-	-	-	-	-	-	-	-2	-	-	-2	-	-	-	-	-	-	-
		1.64	1.54	1.44	1.84	1.74	1.45	2.2		1.8	2.2		1.8	2.2	1.9	1.7	2.2	1.9	1.7

57 Coordinates are normalized to stereotaxic zero coordinates.

58 A, Anterior; M, Medial; P, Posterior; L, Lateral; AP; Anteroposterior; V, Ventral.

59

Supplementary Table 2.- Actual *p*-values of Fig. 1A, Fig. 1F, Fig. 1H, Fig. 2D, Fig. 3D, and Fig. 6C

Fig. 1A

<i>LMX1A</i>	<i>p-value</i>	<i>FOXA2</i>	<i>p-value</i>	<i>TH</i>	<i>p-value</i>
0 vs 7	0.0150	0 vs 7	0.0001	0 vs 7	1.0000
0 vs 14	0.0001	0 vs 14	0.0001	0 vs 14	1.0000
0 vs 21	0.0001	0 vs 21	0.0001	0 vs 21	0.0001
0 vs 31	0.0001	0 vs 31	0.0001	0 vs 31	0.0001
0 vs 38	0.0001	0 vs 38	0.0001	0 vs 38	0.0001
7 vs 14	0.0150	7 vs 14	0.0330	7 vs 14	1.0000
7 vs 21	0.0001	7 vs 21	0.0001	7 vs 21	0.0001
7 vs 31	0.0001	7 vs 31	0.0001	7 vs 31	0.0001
7 vs 38	0.0001	7 vs 38	0.0001	7 vs 38	0.0001
14 vs 21	0.0080	14 vs 21	0.0180	14 vs 21	0.0001
14 vs 31	0.0001	14 vs 31	0.0001	14 vs 31	0.0001
14 vs 38	0.0001	14 vs 38	0.0001	14 vs 38	0.0001
21 vs 31	0.0830	21 vs 31	0.1460	21 vs 31	0.0001
21 vs 38	0.0420	21 vs 38	0.0180	21 vs 38	0.0001
31 vs 38	0.9980	31 vs 38	0.8000	31 vs 38	0.3620

Comparison of the expression on different days of the differentiation protocol.

Fig. 1F

% <i>FOXA2</i> ⁺ / <i>βIII-TUB</i> ⁺	<i>p-value</i>	% <i>TH</i> ⁺ / <i>βIII-TUB</i> ⁺	<i>p-value</i>
24 vs 28	0.0485	24 vs 28	0.0049
24 vs 35	0.0466	24 vs 35	0.0042
24 vs 42	0.0016	24 vs 42	0.0021
28 vs 35	0.1880	28 vs 35	0.0880
28 vs 42	0.2500	28 vs 42	0.2922
35 vs 42	0.1620	35 vs 42	0.4600

Comparison of cells⁺ for different markers on different days of the differentiation protocol.

Fig. 1H

DA release	<i>p</i> -value
B vs HK	0.0045

Comparison in DA release *in vitro* in DAN at day 70 of differentiation in basal (B) solution vs isosmotic high-potassium (HK) medium.

Fig. 2D

SHAM	<i>p</i>-value		
Conditions compared	Displacement	Reaching	Ingestion
B vs. MPTP	0.6268	0.0001	0.2485
B vs. 1	0.7942	0.3471	0.8299
B vs. 2	0.9999	0.0017	0.9999
B vs. 3	0.4067	0.1114	0.7061
B vs. 4	0.0021	0.1309	0.6801
B vs. 5	0.9674	0.1290	0.9990
B vs. 6	0.0060	0.0579	0.9999
B vs. 8	0.0001	0.0344	0.0058
B vs. 9	0.0002	0.0008	0.9999
B vs. 10	0.0015	0.3881	0.9999
MPTP vs. 1	0.0367	0.2688	0.0091
MPTP vs. 2	0.9067	0.9804	0.6796
MPTP vs. 3	0.0061	0.5147	0.0047
MPTP vs. 4	0.6428	0.4933	0.0041
MPTP vs. 5	0.9999	0.5143	0.0955
MPTP vs. 6	0.8234	0.7005	0.1582
MPTP vs. 8	0.0001	0.7848	0.9940
MPTP vs. 9	0.4162	0.9967	0.2746
MPTP vs. 10	0.0001	0.2462	0.7767
1 vs. 2	0.7342	0.9387	0.8111
1 vs. 3	0.9999	0.9999	0.9999
1 vs. 4	0.0001	0.9999	0.9999
1 vs. 5	0.2296	0.9999	0.9995
1 vs. 6	0.0001	0.9999	0.9974
1 vs. 8	0.0001	0.9994	0.0001
1 vs. 9	0.0001	0.8587	0.9812
1 vs. 10	0.5110	0.9999	0.6870
2 vs. 3	0.3819	0.9946	0.7018
2 vs. 4	0.0222	0.9929	0.6800
2 vs. 5	0.9979	0.9942	0.9958
2 vs. 6	0.0515	0.9996	0.9992
2 vs. 8	0.0001	0.9999	0.0917
2 vs. 9	0.0052	0.9999	0.9999
2 vs. 10	0.0025	0.9249	0.9999
3 vs. 4	0.0001	0.9999	0.9999
3 vs. 5	0.0689	0.9999	0.9971
3 vs. 6	0.0001	0.9999	0.9893
3 vs. 8	0.0001	0.9999	0.0001

3 vs. 9	0.0001	0.9759	0.9500
3 vs. 10	0.7922	0.9999	0.5600
4 vs. 5	0.3579	0.9999	0.9962
4 vs. 6	0.9999	0.9999	0.9868
4 vs. 8	0.0001	0.9999	0.0001
4 vs. 9	0.9999	0.9708	0.9422
4 vs. 10	0.0001	0.9999	0.5361
5 vs. 6	0.5431	0.9999	0.9999
5 vs. 8	0.0001	0.9999	0.0023
5 vs. 9	0.1875	0.9748	0.9999
5 vs. 10	0.0002	0.9999	0.9836
6 vs. 8	0.0001	0.9999	0.0051
6 vs. 9	0.9999	0.9960	0.9999
6 vs. 10	0.0001	0.9998	0.9956
8 vs. 9	0.0001	0.9989	0.0128
8 vs. 10	0.0001	0.9990	0.1317
9 vs. 10	0.0001	0.8362	0.9997

GRAFTED 1	<i>p</i>-value		
Conditions compared	Displacement	Reaching	Ingestion
B vs. MPTP	0.0001	0.0001	0.0001
B vs. 1	0.4514	0.4684	0.9564
B vs. 2	0.9999	0.8345	0.9249
B vs. 3	0.9709	0.1359	0.9999
B vs. 4	0.0486	0.0038	0.0001
B vs. 5	0.0255	0.0701	0.0654
B vs. 6	0.0017	0.999	0.0001
B vs. 7	0.006	0.073	0.0011
B vs. 8	0.9998	0.0001	0.0002
B vs. 9	0.9097	0.0349	0.0001
B vs. 10	0.9419	0.7219	0.0058
MPTP vs. 1	0.0001	0.0986	0.0001
MPTP vs. 2	0.0001	0.0378	0.0001
MPTP vs. 3	0.0001	0.1882	0.0001
MPTP vs. 4	0.0001	0.7056	0.3424
MPTP vs. 5	0.0001	0.3777	0.0008
MPTP vs. 6	0.0001	0.004	0.0343
MPTP vs. 7	0.0001	0.3071	0.0143
MPTP vs. 8	0.0001	0.9976	0.0262
MPTP vs. 9	0.0001	0.4234	0.1569
MPTP vs. 10	0.0001	0.0448	0.004
1 vs. 2	0.4557	0.9999	0.9999
1 vs. 3	0.9994	0.9999	0.9979
1 vs. 4	0.9996	0.9595	0.0033
1 vs. 5	0.0001	0.9998	0.924
1 vs. 6	0.9159	0.9865	0.1437
1 vs. 7	0.0001	0.9999	0.3344
1 vs. 8	0.9788	0.2208	0.1856
1 vs. 9	0.9999	0.9991	0.0185
1 vs. 10	0.9999	0.9999	0.5979
2 vs. 3	0.9409	0.9993	0.9941
2 vs. 4	0.0744	0.7852	0.0082
2 vs. 5	0.2429	0.9895	0.9672
2 vs. 6	0.0055	0.9998	0.2356
2 vs. 7	0.1176	0.9938	0.4699
2 vs. 8	0.9974	0.075	0.2919
2 vs. 9	0.8669	0.9738	0.0394
2 vs. 10	0.9029	0.9999	0.7333
3 vs. 4	0.8686	0.9966	0.0001

3 vs. 5	0.0024	0.9999	0.2987
3 vs. 6	0.3609	0.868	0.0045
3 vs. 7	0.0005	0.9999	0.0201
3 vs. 8	0.9999	0.4172	0.0068
3 vs. 9	0.9999	0.9999	0.0002
3 vs. 10	0.9999	0.9998	0.0659
4 vs. 5	0.0001	0.9999	0.3987
4 vs. 6	0.9999	0.2376	0.9924
4 vs. 7	0.0001	0.9998	0.9431
4 vs. 8	0.6105	0.9781	0.9844
4 vs. 9	0.9474	0.9999	0.9999
4 vs. 10	0.9308	0.8333	0.7543
5 vs. 6	0.0001	0.7042	0.9809
5 vs. 7	0.9999	0.9999	0.9989
5 vs. 8	0.0163	0.7457	0.9903
5 vs. 9	0.0011	0.9999	0.7227
5 vs. 10	0.0017	0.9949	0.9999
6 vs. 7	0.0001	0.7392	0.9999
6 vs. 8	0.1515	0.0041	0.9999
6 vs. 9	0.5234	0.5866	0.9999
6 vs. 10	0.4836	0.9991	0.9998
7 vs. 8	0.0048	0.6425	0.9999
7 vs. 9	0.0002	0.9999	0.997
7 vs. 10	0.0004	0.9973	0.9999
8 vs. 9	0.9999	0.7992	0.9998
8 vs. 10	0.9999	0.0887	0.9999
9 vs. 10	0.9999	0.9854	0.9544

GRAFTED 2	<i>p</i>-value		
Conditions compared	Displacement	Reaching	Ingestion
B vs. MPTP	0.1919	0.1592	0.0003
B vs. 1	0.0001	0.5592	0.0087
B vs. 2	0.0001	0.9999	0.1594
B vs. 3	0.0001	0.925	0.5386
B vs. 4	0.0001	0.9999	0.8186
B vs. 5	0.0001	0.4959	0.291
B vs. 6	0.0001	0.3883	0.6234
B vs. 7	0.0001	0.732	0.8762
B vs. 8	0.0072	0.6679	0.3985
B vs. 9	0.0105	0.9999	0.9794
B vs. 10	0.0204	0.9999	0.8379
MPTP vs. 1	0.0001	0.0008	0.0001
MPTP vs. 2	0.0001	0.1733	0.0001
MPTP vs. 3	0.0001	0.0075	0.0001
MPTP vs. 4	0.0001	0.258	0.0001
MPTP vs. 5	0.0001	0.9999	0.8279
MPTP vs. 6	0.4834	0.9999	0.5679
MPTP vs. 7	0.0001	0.9995	0.3186
MPTP vs. 8	0.9981	0.9999	0.756
MPTP vs. 9	0.9987	0.0834	0.0001
MPTP vs. 10	0.9999	0.3599	0.0001
1 vs. 2	0.1731	0.841	0.9978
1 vs. 3	0.0911	0.9999	0.9704
1 vs. 4	0.3968	0.8146	0.8535
1 vs. 5	0.0001	0.0062	0.0001
1 vs. 6	0.0001	0.0036	0.0001
1 vs. 7	0.0001	0.017	0.0002
1 vs. 8	0.0001	0.0137	0.0001
1 vs. 9	0.0001	0.981	0.5822
1 vs. 10	0.0001	0.7562	0.8426
2 vs. 3	0.9999	0.9904	0.9999
2 vs. 4	0.9999	0.9999	0.9997
2 vs. 5	0.0001	0.4841	0.0001
2 vs. 6	0.0001	0.3878	0.001
2 vs. 7	0.0001	0.7015	0.0046
2 vs. 8	0.0001	0.6399	0.0003
2 vs. 9	0.0001	0.9999	0.9838
2 vs. 10	0.0001	0.9999	0.9996
3 vs. 4	0.9999	0.9851	0.9999

94

3 vs. 5	0.0001	0.0424	0.002
3 vs. 6	0.0001	0.0273	0.0105
3 vs. 7	0.0001	0.0965	0.0364
3 vs. 8	0.0001	0.0792	0.0037
3 vs. 9	0.0001	0.9999	0.9997
3 vs. 10	0.0001	0.9716	0.9999
4 vs. 5	0.0001	0.6044	0.0082
4 vs. 6	0.0001	0.5075	0.0359
4 vs. 7	0.0001	0.8012	0.1046
4 vs. 8	0.0001	0.7474	0.0143
4 vs. 9	0.0001	0.9999	0.9999
4 vs. 10	0.0001	0.9999	0.9999
5 vs. 6	0.0037	0.9999	0.9999
5 vs. 7	0.9999	0.9999	0.9998
5 vs. 8	0.0001	0.9999	0.9999
5 vs. 9	0.0001	0.2835	0.038
5 vs. 10	0.0001	0.7194	0.0094
6 vs. 7	0.0005	0.9999	0.9999
6 vs. 8	0.977	0.9999	0.9999
6 vs. 9	0.9771	0.2138	0.1274
6 vs. 10	0.9344	0.6294	0.0401
7 vs. 8	0.0001	0.9999	0.9999
7 vs. 9	0.0001	0.4714	0.2913
7 vs. 10	0.0001	0.8818	0.1144
8 vs. 9	0.9999	0.4142	0.0601
8 vs. 10	0.9999	0.8396	0.0163
9 vs. 10	0.9999	0.9999	0.9999

95

96 Comparison of the different conditions and the evaluated behaviors. B, basal; MPTP, lesioned; 1-
97 10, month evaluated.

98 **Fig. 3D**

Pearson's correlation	Reaching	Ingestion	FA-R	FA-L	MD-R	MD-L
Displacement	$r = 0.8980$ $p = 0.0150$	$r = 0.8603$ $p = 0.0278$	$r = -0.5380$ $p = 0.2707$	$r = -0.5597$ $p = 0.2480$	$r = 0.1563$ $p = 0.7673$	$r = -0.0951$ $p = 0.8577$
Reaching		$r = 0.9909$ $p = 1.2e-4$	$r = -0.6726$ $p = 0.1432$	$r = -0.3766$ $p = 0.4617$	$r = 0.2952$ $p = 0.5699$	$r = 0.0475$ $p = 0.9287$
Ingestion			$r = -0.7151$ $p = 0.1101$	$r = -0.4147$ $p = 0.4135$	$r = 0.3878$ $p = 0.4474$	$r = 0.1595$ $p = 0.7627$
FA-R				$r = 0.5860$ $p = 0.2215$	$r = -0.5602$ $p = 0.2475$	$r = -0.5183$ $p = 0.2921$
FA-L					$r = -0.6582$ $p = 0.1551$	$r = -0.5918$ $p = 0.2158$
MD-R						$r = 0.9415$ $p = 0.0050$

99

100 FA-R, Fractional anisotropy right; FA-L, Fractional anisotropy left; MD-R, Mean diffusivity right; MD-L,
101 Mean diffusivity left.

102

103 **Fig. 6C**

TH+ in <i>Sn</i>	<i>p</i> -value	TH+ / EGFP+ in P	<i>p</i> -value
S vs G1	0.0891	G1 vs G2	0.0958
S vs G2	0.0767		

107 TH+ in the *substantia nigra* (*Sn*) and TH+/EGFP+ in the putamen (P) after grafting.

108

109 **Supplementary video legends.**

110

111 **Supplementary video S1.-** Motor behavioral assessment representative video before MPTP
112 lesion (Sham Basal), after one month of the MPTP neurotoxic lesion (Sham MPTP administration),
113 4 months after surgery, and 10 months after surgery in sham NHP.

114

115 **Supplementary video S2.-** Motor behavioral assessment representative video before MPTP lesion
116 (Grafted 1 Basal), after one month of the MPTP neurotoxic lesion (Grafted 1 MPTP
117 administration), 4 months after surgery, and 10 months after surgery in Grafted 1 NHP.

118

119 **Supplementary video S3.-** Motor behavioral assessment representative video before MPTP lesion
120 (Grafted 2 Basal), after one month of the MPTP neurotoxic lesion (Grafted 2 MPTP
121 administration), 4 months after surgery, and 10 months after surgery in Grafted 2 NHP.

122