

Disease Modifying Treatment of Spinal Cord Injury with Directly Reprogrammed Neural Precursor Cells in Non-Human Primates

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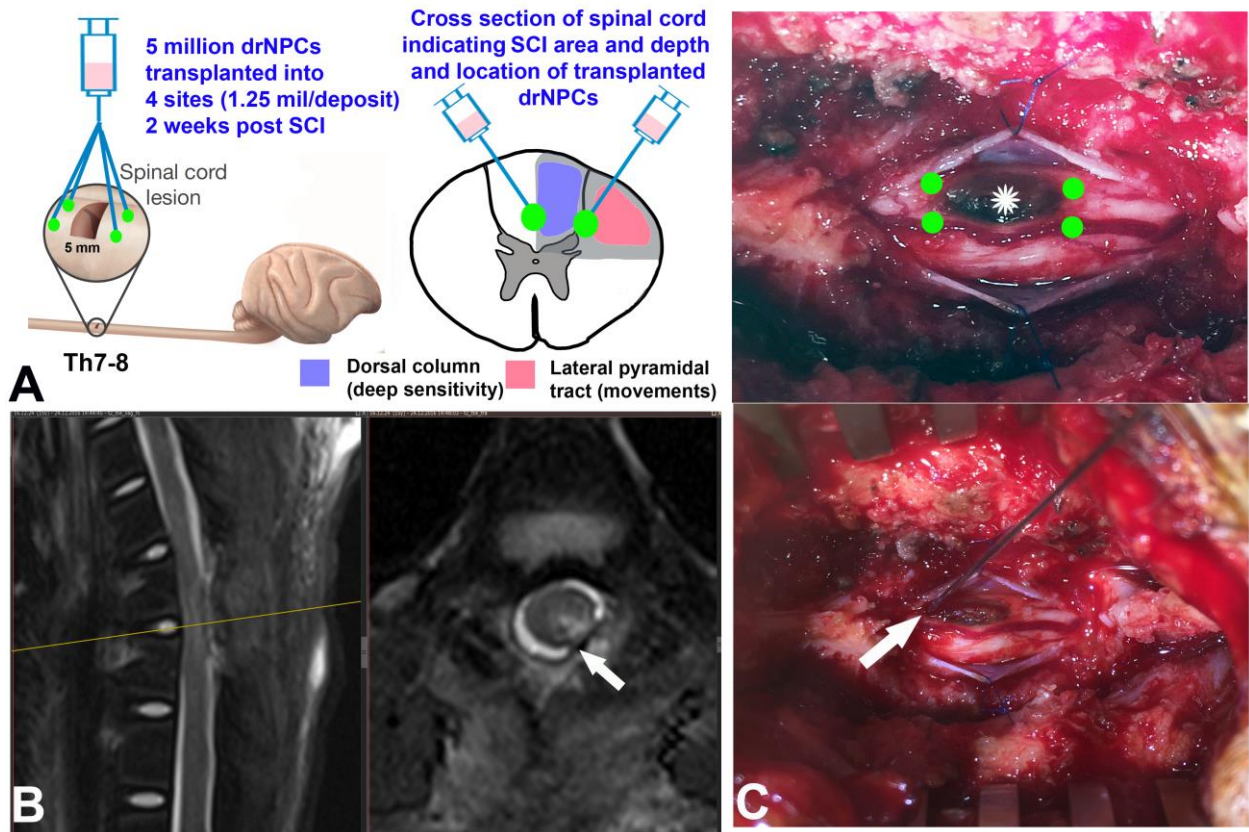
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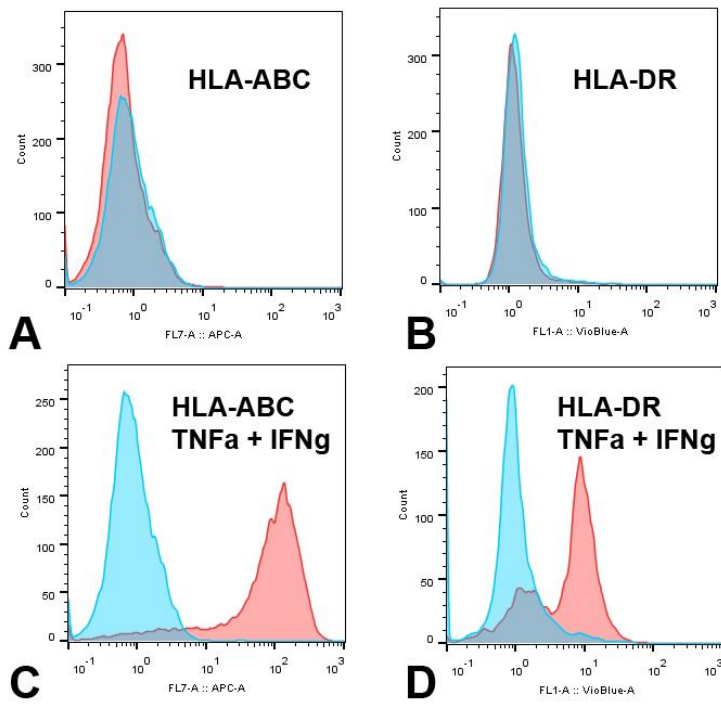
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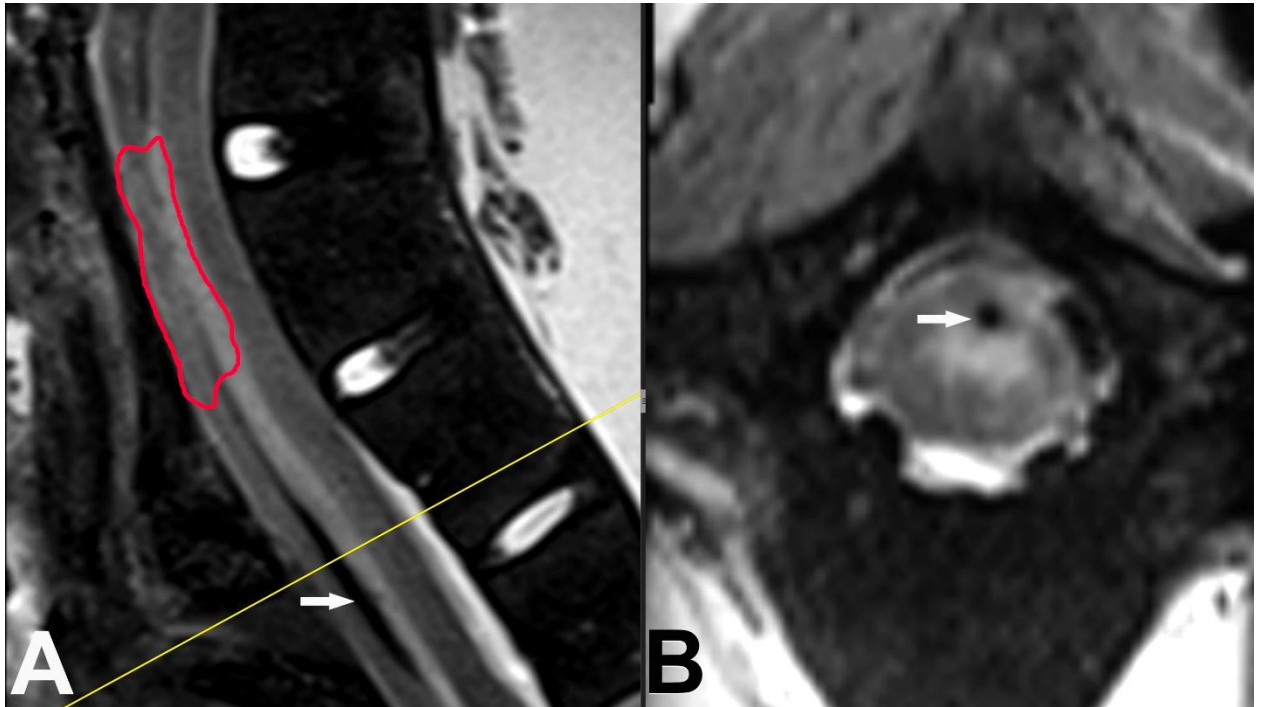
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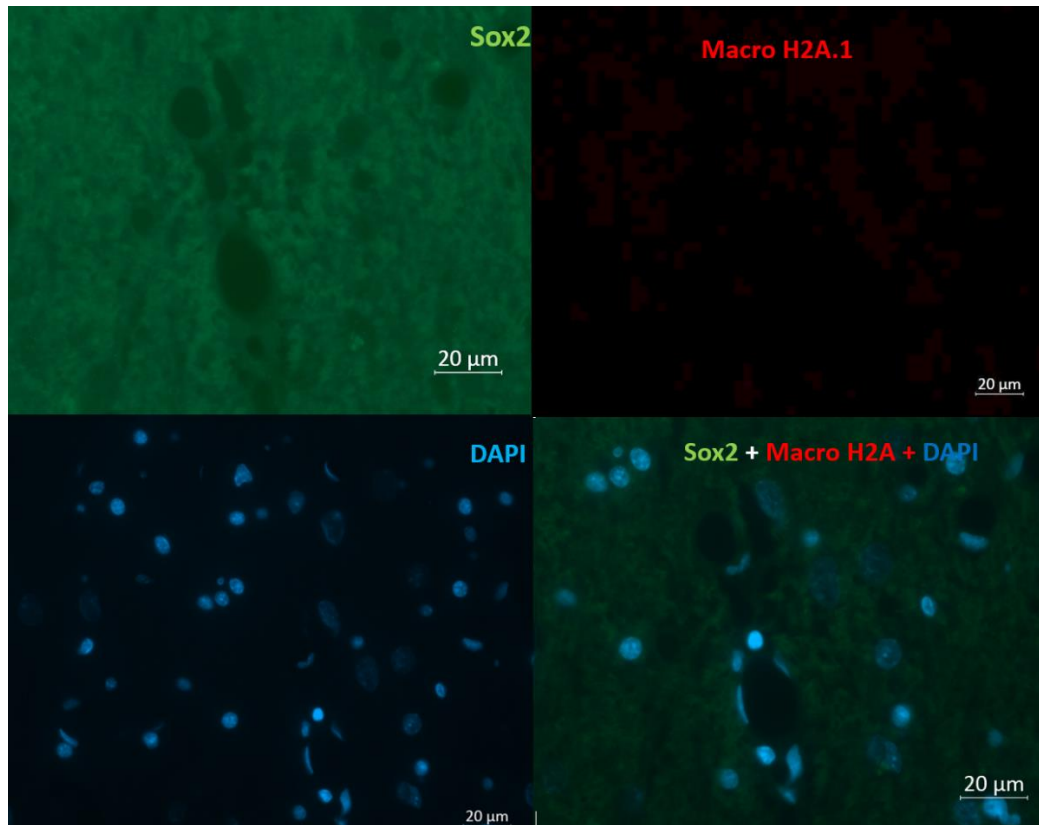
Supplementary Figure 1 Transplantation of drNPCs into perifocal area of SCI. A. The scheme indicating SCI area, and location/depth of drNPCs injection (shown by green dots). B. T2 weighted MRI of SCI site in sagittal (left) and axial (right) projection before the transplantation. The injury shown by arrow. C. Intraoperative photographs showing the injury (asterisk) and sites of injections (green dots). Lower photo: the moment of transplantation. The 28G needle in the lower medial position shown by white arrow.



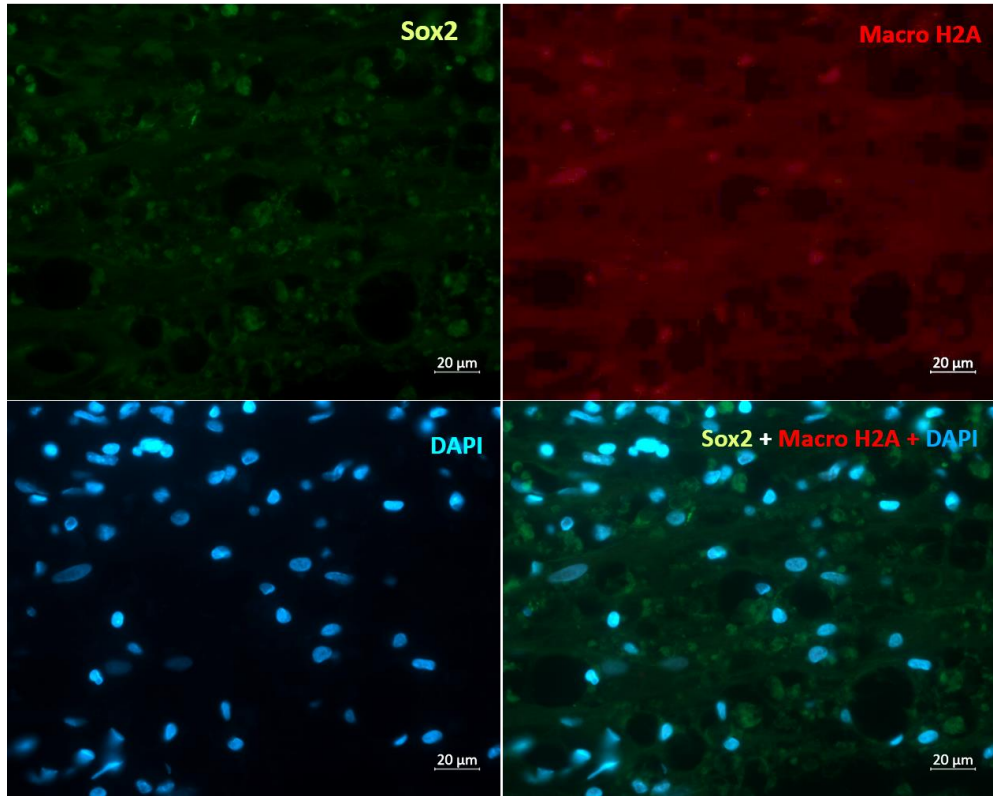
Supplementary Figure 2 Expression of HLA-ABC and HLA-DR by drNPCs in normal conditions (A, B, complete growth medium) and 2 days after administration of TNF α and Interferon- γ (C, D for HLA-ABC and HLA-DR respectively). Mouse Anti-Human HLA-ABC (Cat No.: 561346), BD Pharmingen™ APC Mouse Anti-Human HLA-DR (Cat No.:560744), BD Pharmingen™ APC Mouse IgG1, κ Isotype Control (Cat No.: 550854) and BD Pharmingen™ APC Mouse IgG1 κ Isotype Control (Cat No.: 554681).



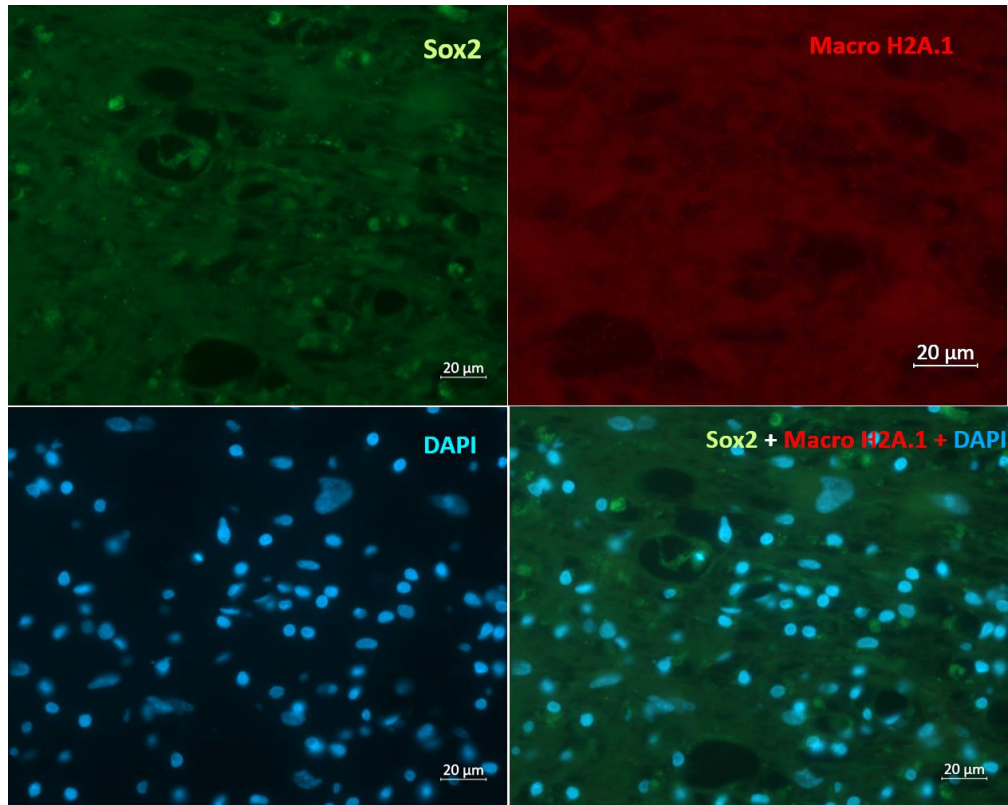
Supplementary Figure 3 MRI changes in one LC animal. A. T2 weighted image, sagittal projection showing dilation of the central spinal channel one segment below the injury (circled in red) B. T2 weighted image, transversal projection (projection plane marked by yellow line on A).



Supplementary Figure 4 Immunohistochemical analysis on spinal cord sagittal sections 12 weeks after vehicle injection (NHP LC1). There was no evidence of Sox2 (green channel) or MacroH2A.1 (red channel) expression.



Supplementary Figure 5 Immunohistochemical analysis on spinal cord sagittal sections 12 weeks after vehicle injection (NHP LC2). There was no evidence of Sox2 (green channel) or MacroH2A.1 (red channel) expression.



Supplementary Figure 6 Immunohistochemical analysis on spinal cord sagittal sections 12 weeks after vehicle injection (NHP LC3). There was no evidence of Sox2 (green channel) or MacroH2A.1 (red channel) expression.

Supplementary Table 1. The dynamics of Hindlimb score in experimental groups

Time point	Parameter\NHP	LC group			NPC group			
		LC1	LC2	LC3	NPC1	NPC2	NPC3	NPC4
Before txp	Mov. main joints (norm. = 3)	0	0	0	0	0	0	0
	Foothold (norm. = 2)	0	0	0	1	0	0	0
	Reflexes (norm. = 2)	1	1	1	0	1	0	1
	Muscle tone (norm. = 2)	1	1	1	1	1	1	1
	Digital function (norm. = 3)	0	0	0	0	0	0	0
	Activity and coordin.(norm. = 3)	1	1	1	1	1	1	1
	TOTAL	3	3	3	3	3	2	3
2 weeks after txp	Mov. main joints (norm. = 3)	0	1	0	1	0	1	1
	Foothold (norm. = 2)	0	0	0	1	0	0	0
	Reflexes (norm. = 2)	1	1	1	1	1		0
	Muscle tone (norm. = 2)	1	1	1	1	1	1	1
	Digital function (norm. = 3)	0	0	0	1	0	0	0
	Activity and coordin.(norm. = 3)	0	1	1	1	1	1	1
	TOTAL	2	4	3	6	3	3	3
4 weeks after txp	Mov. main joints (norm. = 3)	0	1	0	2	1	2	2
	Foothold (norm. = 2)	0	0	0	1	0	1	2
	Reflexes (norm. = 2)	1	1	1	1	1	1	1
	Muscle tone (norm. = 2)	1	1	1	1	1	1	1
	Digital function (norm. = 3)	0	0	0	1	0	1	0
	Activity and coordin.(norm. = 3)	0	1	1	2	1	1	1
	TOTAL	2	4	3	8	4	7	7
8 weeks after txp	Mov. main joints (norm. = 3)	0	1	0	2	2	2	3
	Foothold (norm. = 2)	0	0	0	2	1	2	2
	Reflexes (norm. = 2)	1	1	1	1	1	1	1
	Muscle tone (norm. = 2)	1	1	1	1	1	1	1
	Digital function (norm. = 3)	0	0	0	2	1	1	2
	Activity and coordin.(norm. = 3)	0	1	0	3	1	2	2
	TOTAL	2	4	2	11	7	9	11
12 weeks after txp	Mov. main joints (norm. = 3)	0	1	1	3	3	3	3
	Foothold (norm. = 2)	0	0	0	2	2	2	2
	Reflexes (norm. = 2)	1	1	1	2	1	1	1
	Muscle tone (norm. = 2)	1	1	1	2	1	2	2
	Digital function (norm. = 3)	0	0	0	2	2	1	3
	Activity and coordin.(norm. = 3)	0	1	0	3	3	3	3
	TOTAL	2	4	3	14	12	12	14

Supplementary Table 2. Annotation

Assessment of the hindlimb function: Movements in three main joints (hip, knee and ankle) 0 – absent; the extremity is unbent due to hypertension of extensors; 1 - low-amplitude movements in 1-2 joints; 2 - medium amplitude movements in two joints; 3 - active movements in all three joints, no paresis. **Foothold** 0 – absent; 1 - weak (unsure) support on the hindlimb; 2 - normal support for the hindlimb. **Reflexes (Knee-jerk and Achilles jerk)** 0 - expressed anisoreflexia, the presence of pathological flexural reflexes; 1 - moderate anisoreflexia; 2 - isoreflexia, pathological reflexes are absent. **Violations of muscle tone** 0 - ipsilateral spastic hypertonus or muscular atonia; 1 - moderately pronounced hypertonia; 2 - normal muscle tone, D = S. **Hindlimb digital function** 0 - absent; 1 - weakly expressed (involuntary) digital grasping; 2 - uncertain grip, the impossibility of carrying the weight of the body to the hindlimb when climbing; 3 - normal gripping of the hindlimbs by the fingers, which keeps the animal's body weight when climbing. **Activity and coordination of movements** 0 - absent, the animal predominantly sits / lies; 1 - poorly expressed motor activity and coordination of the upper and lower extremities on the side of the lesion; 2 - moderately expressed motor activity, the animal climbs the cage, the movement of the upper and lower limbs is coordinated; 3 - high activity and coordination of movements, the animal climbs, turns and jumps, using all the limbs.

Interpretation of results: 0 - 3: lower monoplegia; 3 - 6: severe monoparesis; 7 - 10: moderate monoparesis; 11 - 14: minimal dysfunction of corticospinal pathway; 15: no neurological signs.

Supplementary Table 3 Parameters of MEP from *m. abductor hallucis* (MEP-AH)

#	Timepoints	AH L					AH R				
		Lat ms	Amp μ V	Ln/L1	An/A1	EP score	Lat ms	Amp μ V	Ln/L1	An/A1	EP score
LC1	Background	19,0	77,7	100,0%	100,0%	5,0	18,5	103,3	100,0%	100,0%	5,0
	After lesion (D0)	0,0	0,0	0,0%	0,0%	0,0	17,6	340,7	95,1%	329,7%	5,0
	D14 (before transplant)	34,0	7,7	178,8%	9,9%	1,0	26,2	111,0	141,4%	107,4%	3,0
	8 weeks	0,0	0,0	0,0%	0,0%	0,0	27,7	95,9	149,5%	92,8%	5,0
	12 weeks	0,0	0,0	0,0%	0,0%	0,0	24,7	84,1	133,3%	81,4%	5,0
LC2	Background	25,3	425,3	100,0%	100,0%	5,0	25,1	316,7	100,0%	100,0%	5,0
	After lesion (D0)	0,0	0,0	0,0%	0,0%	0,0	30,1	87,0	119,8%	27,5%	3,0
	D14 (before transplant)	30,6	74,0	120,9%	17,4%	1,0	32,5	146,0	129,5%	46,1%	3,0
	8 weeks	0,0	0,0	0,0%	0,0%	0,0	31,2	139,7	124,4%	44,1%	3,0
	12 weeks	43,2	16,0	170,8%	3,8%	1,0	32,4	134,0	129,0%	42,3%	3,0
LC3	Background	22,8	285,3	100,0%	100,0%	5,0	20,5	382,7	100,0%	100,0%	5,0
	After lesion (D0)	0,0	0,0	0,0%	0,0%	0,0	17,7	45,7	86,2%	11,9%	1,0
	D14 (before transplant)	22,4	47,0	98,2%	16,5%	1,0	23,1	471,3	112,7%	123,2%	5,0
	8 weeks	0,0	0,0	0,0%	0,0%	0,0	27,6	179,7	134,4%	47,0%	3,0
	12 weeks	0,0	0,0	0,0%	0,0%	0,0	27,1	83,3	132,0%	21,8%	3,0
NPC1	Background	22,9	117,0	100,0%	100,0%	5,0	24,4	126,0	100,0%	100,0%	5,0
	After lesion (D0)	0,0	0,0	0,0%	0,0%	0,0	26,0	41,9	106,7%	33,3%	3,0
	D14 (before transplant)	0,0	9,3	0,0%	7,9%	1,0	23,0	49,3	94,3%	39,2%	3,0
	8 weeks	66,8	12,7	291,1%	10,8%	1,0	76,6	68,7	314,4%	54,5%	3,0
	12 weeks	57,3	65,3	249,9%	55,8%	3,0	34,5	45,3	141,5%	36,0%	3,0
NPC2	Background	17,8	144,7	100,0%	100,0%	5,0	17,8	299,0	100,0%	100,0%	5,0
	After lesion (D0)	0,0	0,0	0,0%	0,0%	0,0	16,7	305,7	93,6%	102,2%	3,0
	D14 (before transplant)	22,5	16,0	126,2%	11,1%	1,0	17,7	732,0	99,6%	244,8%	5,0
	8 weeks	27,1	26,7	152,1%	18,4%	1,0	17,4	697,7	97,6%	233,3%	5,0
	12 weeks	22,9	79,3	128,2%	54,8%	3,0	17,8	765,0	100,2%	255,9%	5,0
NPC3	Background	30,4	173,7	100,0%	100,0%	5,0	29,3	725,7	100,0%	100,0%	5,0
	After lesion (D0)	0,0	0,0	0,0%	0,0%	0,0	27,8	373,3	94,7%	51,4%	3,0
	D14 (before transplant)	0,0	0,0	0,0%	0,0%	0,0	26,5	109,0	90,3%	15,0%	1,0
	8 weeks	34,9	23,7	114,9%	13,6%	1,0	35,2	117,3	119,9%	16,2%	1,0
	12 weeks	35,8	23,9	117,8%	13,8%	1,0	29,5	290,3	100,7%	40,0%	3,0
NPC4	Background	22,5	529,0	100,0%	100,0%	5,0	22,7	361,3	100,0%	100,0%	5,0
	After lesion (D0)	0,0	0,0	0,0%	0,0%	0,0	21,3	827,0	93,7%	228,9%	5,0
	D14 (before transplant)	0,0	0,0	0,0%	0,0%	0,0	19,7	956,7	86,5%	264,8%	5,0
	8 weeks	23,5	33,7	104,4%	6,4%	1,0	21,0	242,7	92,4%	67,2%	5,0
	12 weeks	24,6	87,3	109,2%	16,5%	1,0	21,6	243,7	95,0%	67,4%	5,0

Abbreviations: L – left side (ipsilateral) R – right side; AH- *m. abductor hallucis*. Ln – initial latency at D0. L1 – the latency at a timepoint. An - initial amplitude at D0. A1 – the amplitude at a timepoint.

Supplementary Table 4 Parameters of MEP from *m. tibialis anterior* (MEP-TA)

#	Timepoints	TAL					TAR				
		Lat ms	Amp μ V	Ln/L1	An/A1	EP score	Lat ms	Amp μ V	Ln/L1	An/A1	EP score
LC1	Background	28,0	690,0	100,0%	100,0%	5,0	19,6	659,3	100,0%	100,0%	5,0
	After lesion (D0)	0,0	0,0	0,0%	0,0%	0,0	14,6	803,3	74,3%	121,8%	5,0
	D14 (before transplant)	0,0	0,0	0,0%	0,0%	0,0	26,3	400,7	134,2%	60,8%	5,0
	8 weeks	0,0	0,0	0,0%	0,0%	0,0	27,3	233,0	139,5%	35,3%	3,0
	12 weeks	31,7	41,0	113,1%	5,9%	1,0	27,0	434,8	137,8%	65,9%	3,0
LC2	Background	24,4	2113,3	100,0%	100,0%	5,0	23,7	2420,0	100,0%	100,0%	5,0
	After lesion (D0)	23,9	118,7	98,2%	5,6%	1,0	24,9	267,3	105,2%	11,0%	1,0
	D14 (before transplant)	32,7	47,0	134,3%	2,2%	1,0	30,5	90,7	128,7%	3,7%	1,0
	8 weeks	27,1	407,3	111,1%	19,3%	1,0	23,4	754,3	99,0%	31,2%	3,0
	12 weeks	23,7	395,0	97,1%	18,7%	1,0	22,7	792,7	96,1%	32,8%	3,0
LC3	Background	16,0	380,0	100,0%	100,0%	5,0	19,5	178,5	100,0%	100,0%	5,0
	After lesion (D0)	0,0	0,0	0,0%	0,0%	0,0	19,1	161,3	98,2%	90,4%	5,0
	D14 (before transplant)	14,8	21,7	92,7%	5,7%	1,0	17,8	192,0	91,3%	107,6%	5,0
	8 weeks	0,0	0,0	0,0%	0,0%	0,0	33,9	87,3	174,3%	48,9%	3,0
	12 weeks	0,0	0,0	0,0%	0,0%	0,0	43,0	69,3	221,1%	38,8%	3,0
NPC1	Background	22,7	2070,0	100,0%	100,0%	5,0	24,8	188,7	100,0%	100,0%	5,0
	After lesion (D0)	23,6	116,0	104,0%	5,6%	1,0	27,1	233,3	109,1%	123,7%	5,0
	D14 (before transplant)	24,1	131,0	106,2%	6,3%	1,0	20,9	753,7	84,2%	399,5%	5,0
	8 weeks	26,9	186,3	118,7%	9,0%	1,0	26,4	2236,7	106,2%	1185,5%	5,0
	12 weeks	27,3	481,7	120,4%	23,3%	3,0	23,8	1890,0	96,0%	1001,8%	5,0
NPC2	Background	18,9	1023,0	100,0%	100,0%	5,0	19,9	2696,7	100,0%	100,0%	5,0
	After lesion (D0)	20,4	61,0	107,9%	6,0%	1,0	18,3	3890,0	92,1%	144,3%	5,0
	D14 (before transplant)	20,0	73,7	105,5%	7,2%	1,0	17,9	831,0	89,8%	30,8%	3,0
	8 weeks	21,4	487,3	113,0%	47,6%	3,0	17,6	2130,0	88,6%	79,0%	3,0
	12 weeks	22,2	576,3	117,3%	56,3%	3,0	16,9	3566,7	84,9%	132,3%	5,0
NPC3	Background	26,3	3516,7	100,0%	100,0%	5,0	24,4	3113,3	100,0%	100,0%	5,0
	After lesion (D0)	41,9	174,0	159,6%	4,9%	1,0	24,6	977,7	100,7%	31,4%	3,0
	D14 (before transplant)	26,3	401,0	100,0%	11,4%	1,0	23,8	648,0	97,4%	20,8%	3,0
	8 weeks	24,8	487,7	94,3%	13,9%	1,0	23,7	667,7	96,9%	21,4%	3,0
	12 weeks	23,9	694,3	90,9%	19,7%	3,0	23,6	916,7	96,5%	29,4%	3,0
NPC4	Background	20,7	1913,3	100,0%	100,0%	5,0	20,4	1109,3	100,0%	100,0%	5,0
	After lesion (D0)	0,0	0,0	0,0%	0,0%	0,0	21,3	2086,7	104,6%	188,1%	5,0
	D14 (before transplant)	18,5	103,3	89,7%	5,4%	1,0	20,4	321,3	99,8%	29,0%	3,0
	8 weeks	21,9	1052,7	106,0%	55,0%	3,0	23,3	1356,7	114,1%	122,3%	3,0
	12 weeks	21,8	827,3	105,5%	43,2%	3,0	19,8	783,7	96,9%	70,6%	5,0

Abbreviations: L – left side (ipsilateral) R – right side; TA — *m. tibialis anterior*. Ln – initial latency at D0. L1 – the latency at a timepoint. An - initial amplitude at D0. A1 – the amplitude at a timepoint.

Supplementary Table 5 Parameters of MEP from *m. quadriceps femoris* (MEP-QF)

#	Timepoints	TAL					TAR				
		Lat ms	Amp μ V	Ln/L1	An/A1	EP score	Lat ms	Amp μ V	Ln/L1	An/A1	EP score
LC1	Background	10,8	280,3	100,0%	100,0%	5,0	11,0	470,0	100,0%	100,0%	5,0
	After lesion (D0)	12,1	51,3	112,1%	18,3%	1,0	10,6	555,0	96,7%	118,1%	5,0
	D14 (before transplant)	27,1	66,7	252,0%	23,8%	3,0	13,8	293,7	125,8%	62,5%	5,0
	8 weeks	17,4	125,7	161,3%	44,8%	3,0	14,2	133,0	128,6%	28,3%	3,0
	12 weeks	19,0	69,8	176,8%	24,9%	3,0	17,6	110,7	160,0%	23,5%	3,0
LC2	Background	18,2	659,7	100,0%	100,0%	5,0	17,5	956,3	100,0%	100,0%	5,0
	After lesion (D0)	17,9	118,0	98,4%	17,9%	1,0	18,0	634,0	103,1%	66,3%	5,0
	D14 (before transplant)	17,8	621,7	97,8%	94,2%	5,0	18,7	1661,3	107,3%	173,7%	5,0
	8 weeks	17,4	439,0	95,4%	66,5%	5,0	17,8	857,7	101,9%	89,7%	5,0
	12 weeks	17,2	861,7	94,7%	130,6%	5,0	17,1	838,0	97,7%	87,6%	5,0
LC3	Background	13,3	589,0	100,0%	100,0%	5,0	15,0	1056,0	100,0%	100,0%	5,0
	After lesion (D0)	16,9	28,3	127,0%	4,8%	1,0	14,5	69,7	96,7%	6,6%	5,0
	D14 (before transplant)	14,7	413,7	110,5%	70,2%	5,0	18,6	324,3	124,2%	30,7%	5,0
	8 weeks	23,8	146,3	178,5%	24,8%	3,0	19,4	1350,7	129,6%	127,9%	5,0
	12 weeks	18,8	157,6	140,8%	26,8%	3,0	20,5	977,3	136,4%	92,6%	5,0
NPC1	Background	18,2	693,0	100,0%	100,0%	5,0	15,0	1150,0	100,0%	100,0%	5,0
	After lesion (D0)	15,6	110,0	85,7%	15,9%	1,0	17,0	1780,0	113,1%	154,8%	5,0
	D14 (before transplant)	16,3	190,0	89,6%	27,4%	3,0	16,4	2283,3	109,6%	198,6%	5,0
	8 weeks	13,1	823,0	71,8%	118,8%	5,0	14,0	816,3	93,6%	71,0%	5,0
	12 weeks	20,6	969,3	112,8%	139,9%	5,0	18,5	2306,7	123,6%	200,6%	5,0
NPC2	Background	15,8	1042,3	100,0%	100,0%	5,0	16,7	1024,3	100,0%	100,0%	5,0
	After lesion (D0)	15,4	184,7	97,3%	17,7%	1,0	15,7	865,3	94,0%	84,5%	5,0
	D14 (before transplant)	17,7	483,3	111,6%	46,4%	3,0	13,6	4183,3	81,6%	408,4%	5,0
	8 weeks	15,4	949,7	97,5%	91,1%	5,0	16,2	1567,7	97,2%	153,0%	5,0
	12 weeks	18,4	956,7	116,2%	91,8%	5,0	15,4	2693,3	92,2%	262,9%	5,0
NPC3	Background	16,0	155,3	100,0%	100,0%	5,0	15,2	154,7	100,0%	100,0%	5,0
	After lesion (D0)	13,6	25,3	85,2%	16,3%	1,0	16,7	398,3	109,8%	257,5%	5,0
	D14 (before transplant)	16,8	74,0	105,0%	47,6%	3,0	16,7	1533,0	109,6%	991,2%	5,0
	8 weeks	14,1	110,0	88,3%	70,8%	5,0	15,6	1540,0	102,2%	995,7%	5,0
	12 weeks	15,2	149,7	95,2%	96,4%	5,0	14,5	208,3	95,4%	134,7%	5,0
NPC4	Background	14,6	115,3	100,0%	100,0%	5,0	13,8	297,3	100,0%	100,0%	5,0
	After lesion (D0)	15,2	18,7	104,6%	16,2%	1,0	13,1	117,7	95,2%	39,6%	3,0
	D14 (before transplant)	12,9	79,3	88,6%	68,8%	5,0	13,0	723,3	94,2%	243,3%	5,0
	8 weeks	12,9	84,7	88,6%	73,4%	5,0	13,1	544,3	95,2%	183,1%	5,0
	12 weeks	12,8	96,7	88,1%	83,8%	5,0	13,7	483,3	99,3%	162,6%	5,0

Abbreviations: L – left side (ipsilateral) R – right side; QF – *m. quadriceps femoris*. Ln – initial latency at D0. L1 – the latency at a timepoint. An - initial amplitude at D0. A1 – the amplitude at a timepoint.

