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In vitro profiling of application ready human surrogate primary progenitor stromal cell fractions

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Running title:

Characterization of cultured and configured stromal progenitor cell based platforms derived from human biological discard - for in vitro applications

Table 1 : Summary table comparing the gene expression status of CNCs with other cells

SNo	Marker name	Neuronal lineage	Compared with Stromal cells	Compared with Sorted cells	Compared with SH5SY
1	Pax6	Neural stem cell	↑ (0.0009)	↑ ns	↑ (0.0009)
2	CD133	Neural stem cell	↑ (0.015)	↑ ns	↑ (0.006)
3	SSEA1	Neural stem cell	↑ (0.001)	↑ (0.001)	↑ (0.00009)
4	Musashi1	Neural stem cell	↑ (0.008)	↑ (0.002)	↓ (0.003)
5	Tra1-81	Pluripotent markers	↑ (0.004)	↑ ns	↓ (0.0005)
6	Nestin	Neuroepithelial	↑ (0.01)	↑ (0.01)	↓ (0.006)
7	TH	DA neurons	-	-	-
8	Nurr1	DA neurons	↓ (0.000508)	↓ ns	↓ (0.006)
9	DAT	DA neurons	↑ ns	↑ ns	↑ (0.047)
10	EN	DA neurons	↑ ns	-	↓ (0.004)
11	FoxA2	DA neurons	↑ (0.023)	↑ ns	↑ (0.013)
12	Otx2	DA neurons	↑ (0.072)	↑ (0.04)	↑ (0.06)
13	EAAT	Radial Glial	↑ (0.008)	↑ ns	↑ (0.008)
14	Tenascin	Radial Glial	-	-	-
15	Vimentin	Radial Glial	↑ (0.003)	↑ ns	↑ (0.002)
16	GFAP	Radial Glial	-	-	↓ (0.02)
17	CNPase	Oligodendrocyte	↑ (0.022)	↑ ns	↓ ns
18	TUBB	Immature neurons	↑ (0.015)	↑ (0.021)	↑ (0.023)
19	NeuroD1	Immature neurons	↑ (0.0006)	↑ ns	↑ (0.00004)
20	MAP2	Mature neurons	-	-	-
21	NEUN	Mature neurons	-	-	↓ (0.001)
22	NFL	Mature neurons	↑ (0.0003)	↑ (0.0006)	↓ (0.02)
23	NSE	Mature neurons	-	-	-

Note: Upward arrow depicts increased and downward arrow depicts decreased expression in CNC compared to the representative cells. *p* values were calculated by student *t* test and the value has been mentioned in the parenthesis, ns stands for not significant, arrow signifies increased or decreased values.

Table 2: Primer sequences

SNo	Gene	Forward primer	Reverse Primer	amplicon size
1	TH	GCTGGACAAGTGTCATCACCTG	CCTGTAAGTGAAGGCGATCTCA	
2	NURR1	AAACTGCCCAGTGGACAAGCGT	GCTCTTCGGTTTCGAGGGCAAA	145
3	DAT	CCTCAACGACACTTTTGGGACC	AGTAGAGCAGCACGATGACCAG	149
4	NESTIN	TCAAGATGTCCCTCAGCCTGGA	AAGCTGAGGGAAGTCTTGAGC	106
5	PAX6	CTGAGGAATCAGAGAAGACAGGC	ATGGAGCCAGATGTGAAGGAGG	131
6	CD133	CACTACCAAGGACAAGGCGTTC	CAACGCCTCTTTGGTCTCCTTG	151
7	EN-1	GTGGTCAAACTGACTCGCAGC	CCGCTTGTCTCCTTCTCGTTC	135
8	PITX3	AGGAGATCGCCGTGTGGACCA	CCGCGAAGCTGCCTTTGCATAG	126
9	FOXA2	GGAACACCACTACGCCTTCAAC	AGTGCATCACCTGTTCGTAGGC	134
10	OTX2	GGAAGCACTGTTTGCCAAGACC	CTGTTGTTGGCGGCACTTAGCT	133
11	GFAP	CTGGAGAGGAAGATTGAGTCGC	ACGTCAAGCTCCACATGGACCT	122
12	CNP	CGGTCAGAAGAAGGGCGACAAG	GGTGCTCTTGCAAGAAGGCGAG	134
13	TUBB3	TCAGCGTCTACTACAACGAGGC	GCCTGAAGAGATGTCCAAAGGC	120
14	TRA1-81	CCTGAACCTCACAGGAAACACC	TGGAACAGATGCCAGCCGTATG	136
15	SSEA-1	GGGTTTGGATGAACTTCGAGTCG CAAGTAATGTCGATAAAGAGGAG	GGTAGCCATAAGGCACAAAGACG	123
16	PCNA	G	GTGTCACCGTTGAAGAGAGTGG	126
17	KI-67	GAAAGAGTGGCAACCTGCCTTC	GCACCAAGTTTACTACATCTGCC	151
18	MUSASHI	GCTCAGCCAAAGGAGGTGATGT	GCGTAGGTTGTGGCTTGAAAC	134
19	EAAT1	GGTTGCTGCAAGCACTCATCAC	CACGCCATTGTTCTCTTCCAGG	95
20	TENASCIN C	ATGTCCTCCTGACAGCCGAGAA	AGTCACGGTGAGGTTTTCCAGC	101
21	VIMENTIN NEUROGE	AGGCAAAGCAGGAGTCCACTGA	ATCTGGCGTTCCAGGGACTCAT	100
22	NIN-2	CAAGCTCACCAAGATCGAGACC	AGCAAACTGCCTCGGAGAAGA	138
23	NEUROD1	GGTGCCTTGCTATTCTAAGACGC	GCAAAGCGTCTGAACGAAGGAG	139
24	MAP2	AGGCTGTAGCAGTCCTGAAAGG	CTTCTCCACTGTGACAGTCTG	153
25	NEUN	TACGCAGCCTACAGATACGCTC	TGGTCCAATGCTGTAGGTCGC	133
26	NF-L	CCAAGACCTCCTCAACGTGAAG	ATGCTTCCCACGCTGGTGAAC	108
27	NSE	CTGTATCGCCACATTGCTCAGC	AGCTTGTTGCCAGCATGAGAGC	101
28	FOXA2	GGAACACCACTACGCCTTCAAC	AGTGCATCACCTGTTCGTAGGC	134
29	Oct	ACTGGAGAAGGAGAAGCTGG	GTCGTTTGGCTGAACACCTT	180
30	Sox2	GCCGAGTGGAACTTTTGTGCG	GCAGCGTGTACTTATCCTTCTT	154
31	Nanog	GTCTTCTGCTGAGATGCCTCACA	CTTCTGCGTCACACCATTGCTAT	384
32	AFP	TTTGGGACCCGAACCTTCCA	CATCCTGCAGACAATCCAGC	154
33	Desmin	CTGTGAGCCTTGGCTGTTG	GTATCCCAACACCCTGCTCT	151
34	GAPDH	ATCACCATCTTCCAGGAGCGA	TTCTCCATGGTGGTGAAGACG	101
35	Rex1/ZFP42	AACGGGCAAAGACAAGACAC	AACTCACCCTTATGACGCA	223
36	SSEA4	TCCGACTGGTTTGACAGCCACT	CTTCTCCAGCACCTCATTGGTG	147
37	cMyc	CCTGGTGCTCCATGAGGAGAC	CAGACTCTGACCTTTTGCCAGG	128