

**Supplementary Table 1. Accuracy of targeted addition for *XIST* transgene on Chr21 in Down syndrome iPSCs**

Ratio of <i>XIST</i> to Puro	<i>XIST</i> + clones (Puro+)	Random integration	Targeted integration	Single target	Double target	Triple target
3:1	65	1 (1.5%)	64 (98.5%)	57 (87.7%)	7 (10.8%)	0 (0.0%)
5:1	16	1 (6.3%)	15 (93.8%)	8 (50.0%)	5 (31.3%)	2 (12.5%)

**Supplementary Table 1.** Changes in the ratio of selectable marker to *XIST* transgene produce conditions where multiple targeted integrations of *XIST* are seen more frequently. Random integrations may be over-estimated, as much searching was required to find them.

**Supplementary Table 2. Percentage of down regulated genes ( $p < 0.01$ ) by microarray**

	Chr 21	whole genome
Disomic/Trisomic	93.5	42.2
Parental line (DOX/no DOX)	41.0	36.6
Clone 1 (DOX/no DOX)	96.3	51.2
Clone 2 (DOX/no DOX)	94.5	49.4
Clone 3 (DOX/no DOX)	98.1	49.5

**Supplementary Table 2.** Of genes with significant change in expression ( $p < 0.01$ ), ~95% were repressed on Chr21, with balanced (~50%) changes (up and down) on other chromosomes in three transgenic clones. Repression in dox-treated clones reflects repression seen if the third Chr21 was lost (Disomic/Trisomic).

**Supplementary Table 3. Primers for Chr21 gene amplification (allele-specific SNP silencing analysis)**

genes	Forward primer	reverse primer
<i>ADAMTS1</i>	5' - TCTCTGAAACCATAGCAGCCA -3'	5' - CTTGTGCAGACCATCCCTGC -3'
<i>ETS2</i>	5' - GCCTTTTGCAACCAGGAACAGC -3'	5' - ATCACACAGAAGAACGTGGAGC -3'
<i>SPA13</i>	5' - AACTCTGCTCCAAATGCCGA -3'	5' - CCTGTACATCATTCTCTGCTTGG -3'
<i>TIAM1</i>	5' - TGGGGTGATTTGCTTTCCAGTGC -3'	5' - GTGCAGTGTCTGCCCAAGC -3'

**Supplementary Table 4 Primers for qRT-PCR**

genes	Forward primer	reverse primer
<i>CXADR</i>	5' - TCGGTCTAAACGTTGTCCCT - 3'	5' - AGTGGACGTACGGCTCTTTG - 3'
<i>COL6A1</i>	5' - ATCAGCCAGACCATCGACAC - 3'	5' - GCCCTTCTCTCCCTTGTAGC - 3'
<i>PTTG1IP</i>	5' - GTTGGGTGAACTTTGAGGCG - 3'	5' - GTGCTGGAGCGCTTTAGTTG - 3'
<i>ADAMTS1</i>	5' - CCCTCACTCTGCGGAACTTTT - 3'	5' - ATTAAGGCTGGCACACTGCTT - 3'
<i>BTG3</i>	5' - CCCATGTGAGGTGTGCTGT - 3'	5' - AGGGCCCTGGTAACTTTCCT - 3'
<i>TIAM1</i>	5' - TCAAAACCGAGAGCCTTCCC - 3'	5' - CGGAGACGGCATCAGAATCA - 3'
<i>USP16</i>	5' - AGCCTTCAGTTTGGCTGTGT - 3'	5' - GGCTTTGGAGTTGTAATGCTGG - 3'
<i>APP</i>	5' - GGAGCGCTCTCGACTTTTCT - 3'	5' - TGTGCATGTTTCAGTCTGCCA - 3'
<i>β-ACTIN</i>	5' - TTGCCGACAGGATGCAGAAGGA - 3'	5' - AGGTGGACAGCGAGGCCAGGAT - 3'