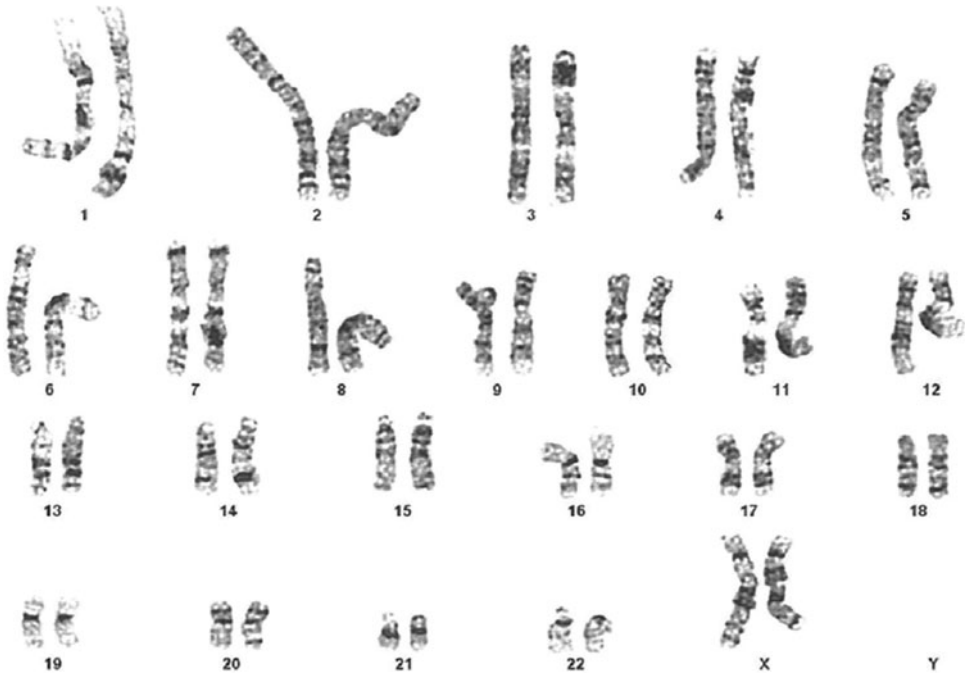
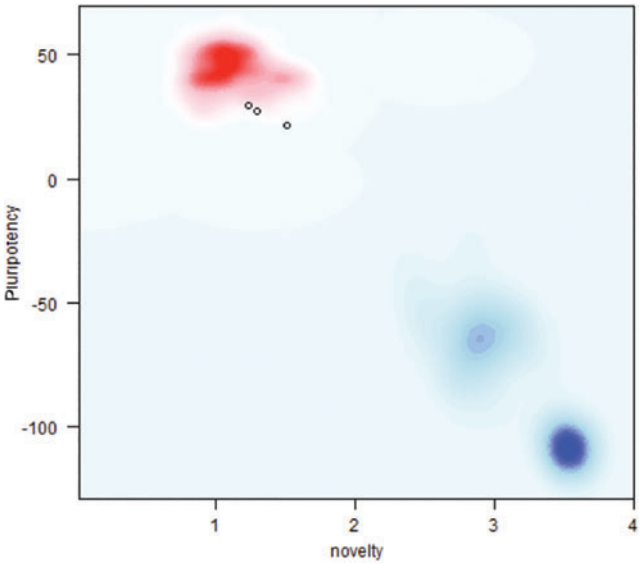
Supplementary Data



SUPPLEMENTARY FIG. S1. Karyotype analysis by G-banding at a 550 banding resolution for all three 23i-SD *TRPV4* clones. Analysis showed a normal 46, XX karyotype for all three 23i clones (n1, n12, and n14) with no evidence of chromosomal rearrangement. The normal karyotype for clone 23i-n14 iPSC is shown. iPSC, induced pluripotent stem cell; SD, skeletal dysplasia.



SUPPLEMENTARY FIG. S2. Pluripotency assessment of the 23i-SD *TRPV4* iPSC clones. A bioinformatics-based PluriTest assay [3] to determine a pluripotent signature of the 23i clones (n1, n12, and n14) is shown by a graphical depiction of the Novelty score plotted against the Pluripotency Score. The *red* and *blue background* indicate the empirical distribution of human pluripotent iPSCs (*red*) and differentiated cells (*blue*) in the stem cell matrix. The graph compares well-characterized pluripotent lines (*red spot*) and the three 23i-SD clones (*circles*) in a stem cell model matrix. A low Novelty score (see also Table 1) indicates that the 23i-SD iPSC clones are a good fit with karyotypically normal, reproducible signatures from well-characterized pluripotent standard cell lines. The Pluripotency score indicates whether a sample contains an expression signature consistent with pluripotency. However, it does not indicate whether the sample is consistent with a normal genetically stable human iPSC line [3]. Based on these scores, the iPSCs generated here fit the standards associated with pluripotency (*Red* cloud area, *top left*).

SUPPLEMENTARY TABLE S1. PRIMERS USED FOR qPCR (TOP) AND RT-PCR (BOTTOM) ASSAYS

Target genes for qPCR	*Sequence*	*Product size (bp)*	*Exons*	*RefSeq#*
COL2A1-IIA	F: 5′-CCCCGCGGTGAGCCATGATT-3′ R: 5′-CTGCCAGCCTCCTGGACATCCT-3′	112	1 1–2	NM_001844.4
COL2A1-IIB	F: 5′-AGGATGTCCGGCAACCAG-3′ R: 5′-TTTGTCACCACGATCCCCTC-3′	137	1–3 6	NM_033150.2
SOX9	F: 5′-TCTGGAGACTTCTGAACGAGAGCG-3′ R: 5′-CTTCACCGACTTCCTCCGCCG-3′	125	1–2 2	NM_000346.6
AGGRECAN	F: 5′-GACAACCTCGCCTGTGATTGA-3′ R: 5′-CCCCTCAGACCCAACTACCA-3′	88	14–15 15	NM_001135.3
COL10A1	F: 5′-ACCTTCTGCACTGCTCATCTGGG-3′ R: 5′-ATGGATTCTGCGTGCTGGGAGT-3′	87	1 1–2	NM_000493.3
RUNX2	F: 5′-ACCACAAGTGCGGTGCAAACT-3′ R: 5′-TGCTTGCAGCCTTAAATGACTCTGT-3′	123	1 1–2	NM_001015051.3
COL1A1	F: 5′-AAGGGTAACAGCGGTGAAC-3′ R: 5′-AACACCAACAGGGCCAG-3′	81	19–20 21	NM_000088.3
GAPDH	F: 5′-GTATCGTGGAAGGACTCATGACCA-3′ R: 5′-TAGAGGCAGGGATGATGTTCTGGA-3′	126	6–7 7	NM_001256799.1
MSX1	F: 5′-CGAGAGGACCCCGTGGATGCAGAG-3′ R: 5′-GGCGGCCATCTTCAGCTTCTCCAG-3′	307	1 2	NM_002448.3
PAX6	F: 5′-TCCTTCTACGGACGGAACTG-3′ R: 5′-AGAAATGCCTGAGGAAAGCA-3′	140	6 **7**	NM_000280.4
HAND1	F: 5′-CATCGCCTACCTGATGGACG-3′ R: 5′-TCCCTTTTCCGCTTGCTCTC-3′	114	1 2	NM_004821.2
BMP4	F: 5′-GCGAGAGAGACGCAGACGCA-3′ R: 5′-CGGAATGGCTCCATGTTCCCC-3′	86	1 1–2	NM_130851.2
AFP	F: 5′-GAATGCTGCAAACTGACCACGCTGGAAC-3′ R: 5′-TGGCATTCAAGAGGGTTTTCAGTCTGGA-3′	281	8 9	NM_001134.2
GATA4	F: 5′-GGCCGCTCATCAAGCCTCAG-3′ R: 5′-AAGGAGCTGCTGGTGTCTTA-3′	249	3 5–6	NM_002052.3
SOX17	F: 5′-GTACGCTGTAGACCAGACCG-3′ R: 5′-CTCGCCCTCACCTTCATGT-3′	314	1 1	NM_0022454.2
TDGF	F: 5′-TCCTTCTACGGACGGAACTG-3′ R: 5′-AGAAATGCCTGAGGAAAGCA-3′	140	4 5	NM_001174136.1
EBNA1	F: 5′-GGTCCCGAGAATCCCCATCC-3′ R: 5′-TTCATGGTCGCTGTCAGACAGA-3′	197	73n 279n	GenBank M13180.1
GAPDH	F: 5′-GTGGACCTGACCTGCCGTCT-3′ R: 5′-GGAGGAGTGGGTGTCGCTGT-3′	153	6–7 7	NM_001256799.1

The primers were designed with Primer 3 software [1,2] available at www.ncbi.nlm.nih.gov/tools/primer-blast, and for the qPCR, at least one primer was located at an exon-exon junction. For each target gene, the product sizes in base pairs, the exon(s) location and the RefSeq (mRNA) accession # are shown.

F, forward primer; R, reverse primer; n, nucleotide where primer starts; RT-PCR, reverse transcription polymerase chain reaction; qPCR, quantitative real-time polymerase chain reaction.

Supplementary References

1. Untergrasser A, I Cutcutache, T Koressaar, J Ye, BC Faircloth, M Remm and SG Rozen. (2012). Primer3—new capabilities and interfaces. Nucleic Acids Res 40:e115.
2. Koressaar T and M Remm. (2007). Enhancements and modifications of primer design program Primer3. Bioinformatics 23:1289–1291.
3. Müller FJ, B Brändl and JF Loring. (2012). Assessment of human pluripotent stem cells with PluriTest. *StemBook* [Internet]. Harvard Stem Cell Institute, Cambridge, MA, pp. 1–17.