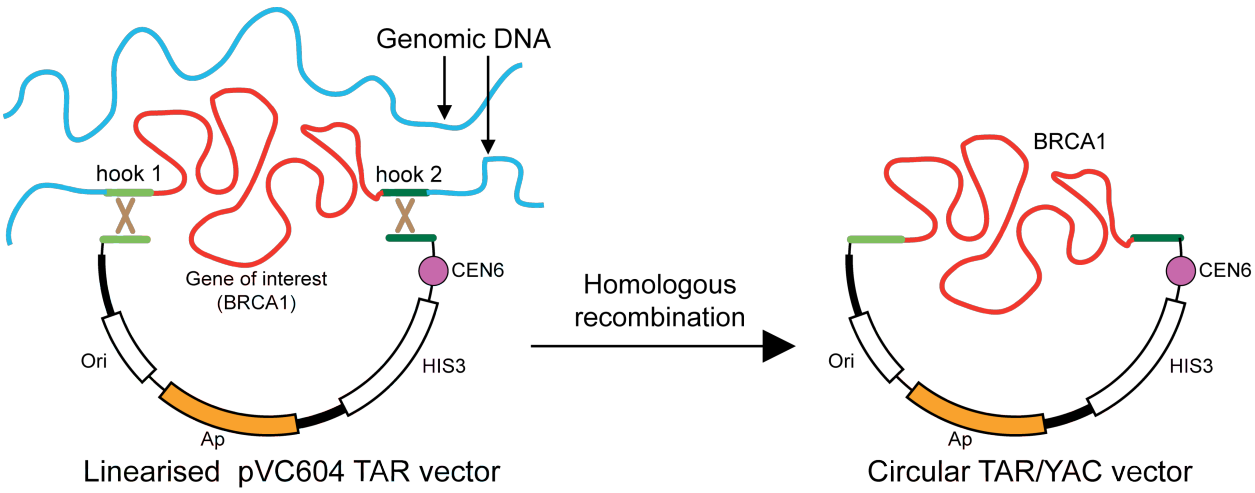


Figure S1. The diagram shows TAR cloning of the BRCA1 gene from total human genomic DNA with a linearized pVC604 TAR vector containing two unique targeting sequences (hooks) homologous to a gene of interest. After transformation into yeast *Saccharomyces cerevisiae* cells, recombination between targeting sequences in the vector and the targeted sequences of the genomic DNA fragment carrying *BRCA1* leads to the rescue of the gene as a circular TAR/YAC molecule. For TAR cloning experiments, the TAR vector DNA is linearized at a unique restriction site located between the hooks to expose targeting sequences (hooks).

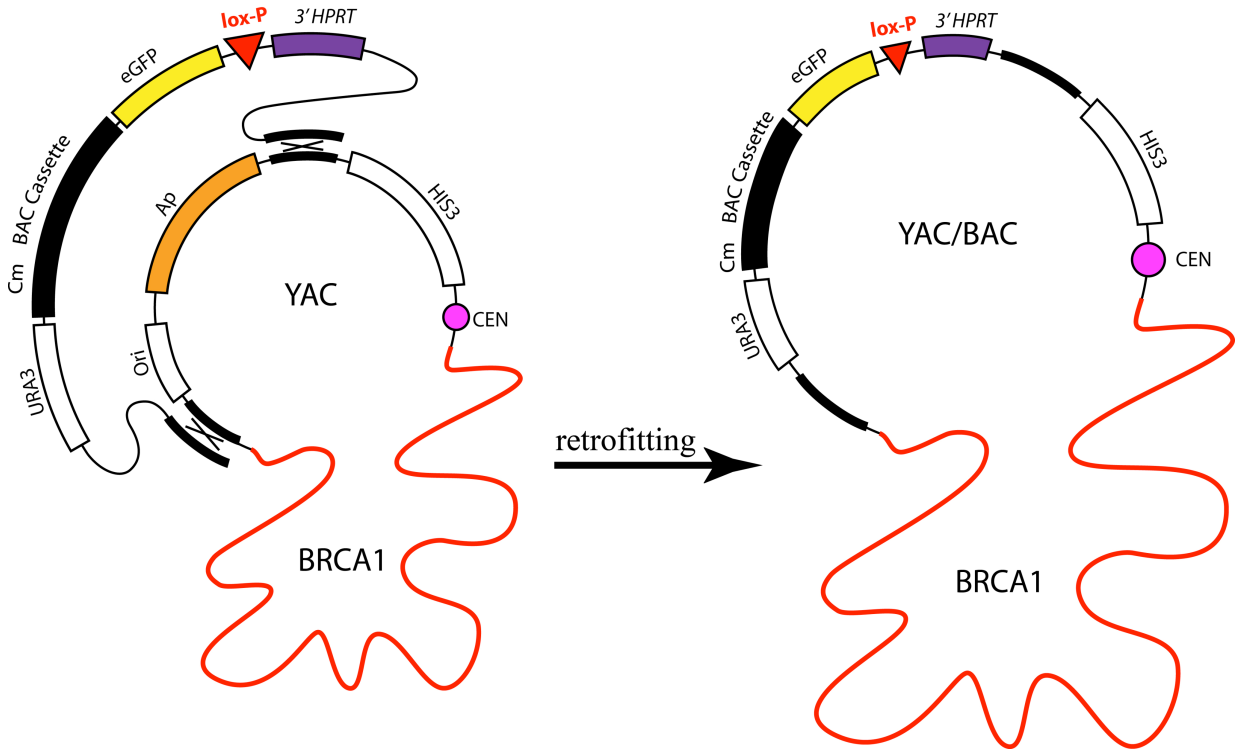


Figure S2. Schematic representation of retrofitting a circular TAR/YAC carrying the full-length *BRCA1* gene into a YAC/BAC using the pJBRV1vector containing the *URA3* yeast selectable marker, a BAC cassette containing the F′ factor origin of replication (*F'*), the chloramphenicol acetyltransferase (*Cm*) gene and a 3′HPRT-loxP cassette allowing gene loading into a unique loxP site of the alphoid^{tetO} -HAC in Hprt-deficient hamster CHO cells by Cre-loxP mediated recombination. A standard lithium acetate transformation procedure was used for retrofitting of a YAC into a BAC/YAC form. Recombination between targeting sequences in the vector and homologous regions in the YAC molecule replaces the *ColEI* origin of replication bat the *F'*-factor origin of replication that allows propagation of a molecule in bacterial cells. The YAC/BACs were moved to *Escherichia coli* by electroporation.

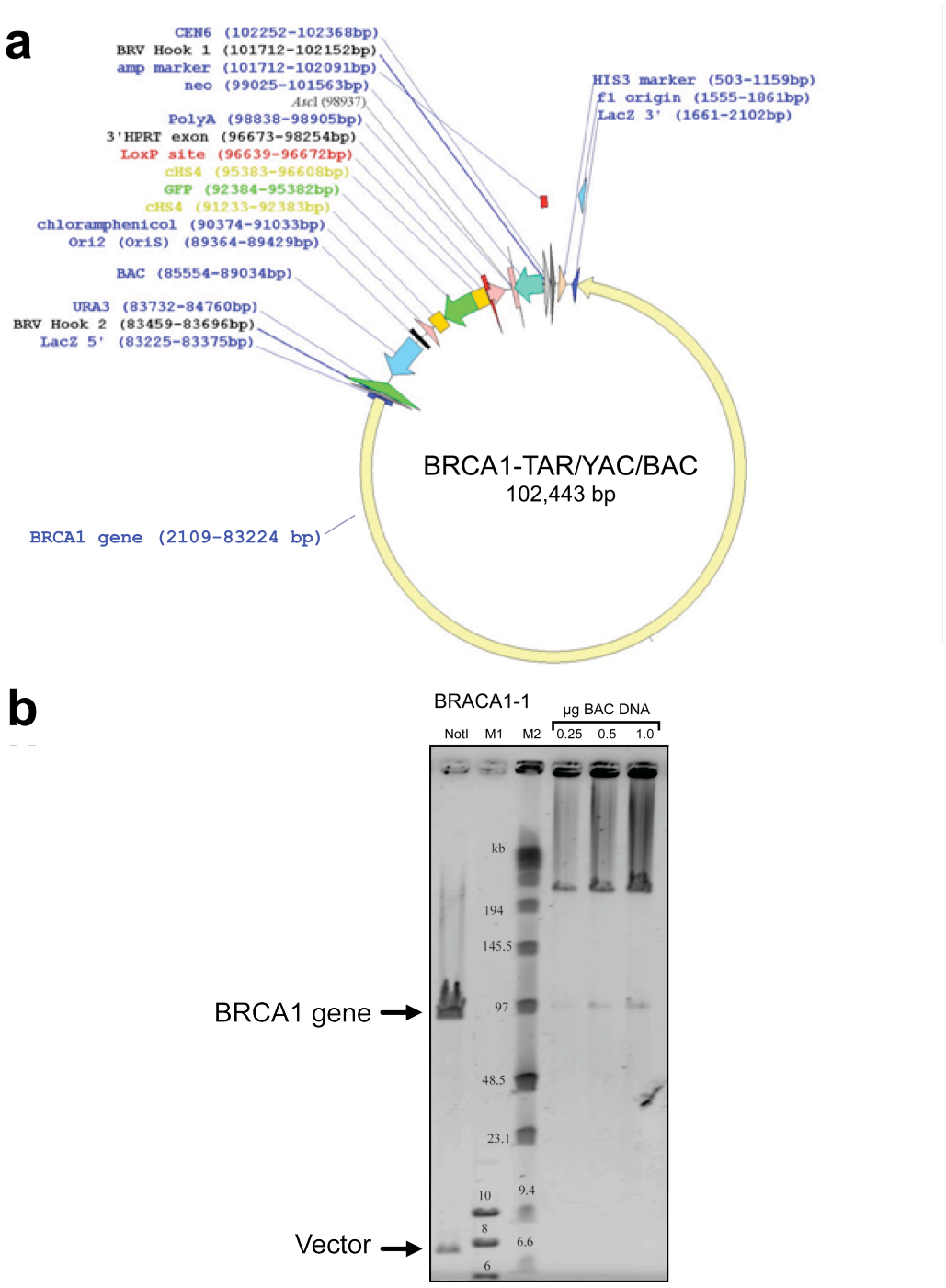
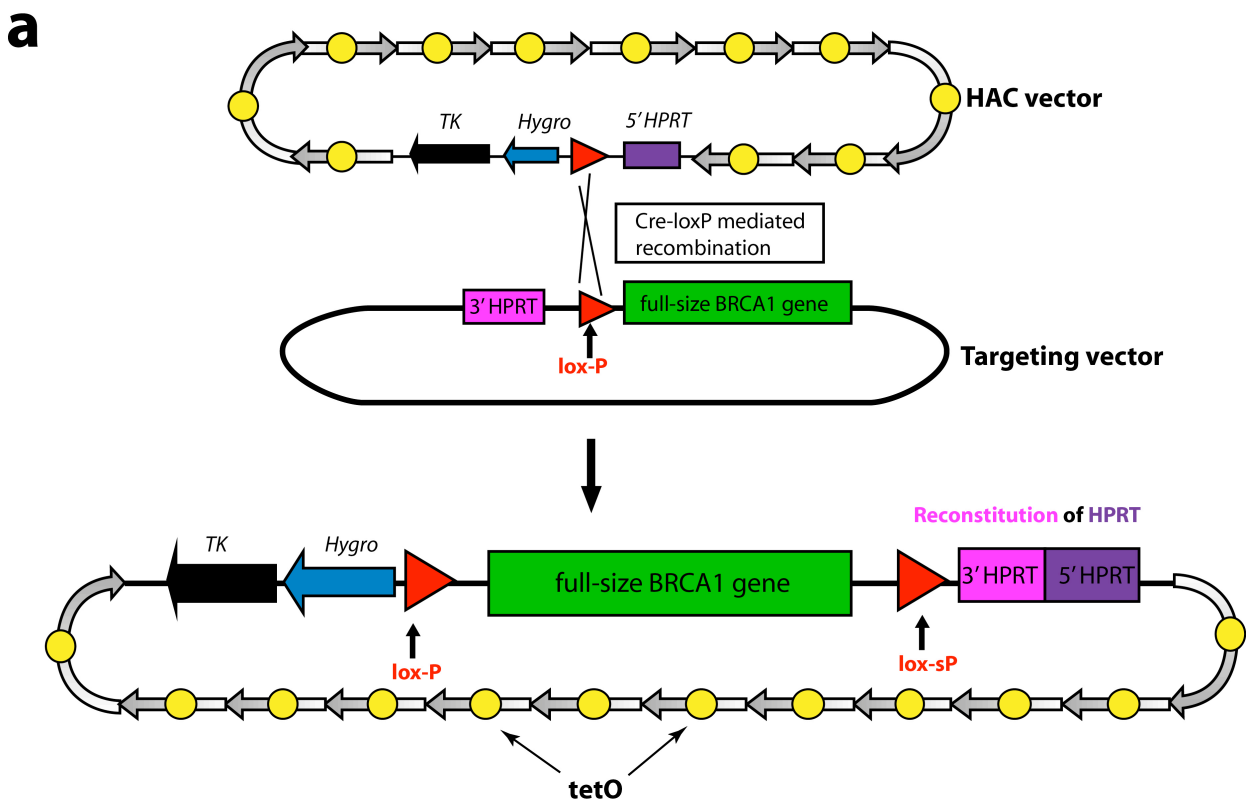


Figure S3. (a) Map of BRCA1-TAR/YAC/BAC used for loading the gene into alphoid ^{tetO}-HAC. **(b)** CHEF gel to verify the quality and integrity of BRCA1-YAC/BAC preparation. BRCA1-1 is a randomly selected BRCA1-TAR/YAC/BAC *E. coli* clone. The first sample lane is NotI digested BRCA1-1 BAC DNA. 0.25µg, 0.5µg and 1µg of uncut BRCA1-1 BAC were loaded onto the gel. As seen, a purified BRCA1 BAC DNA is predominately presented by covalently closed circular (ccc) molecules. M1 is the 2-log DNA ladder (NEB). M2 is the Pulse Marker 0.1-200kb (Sigma).



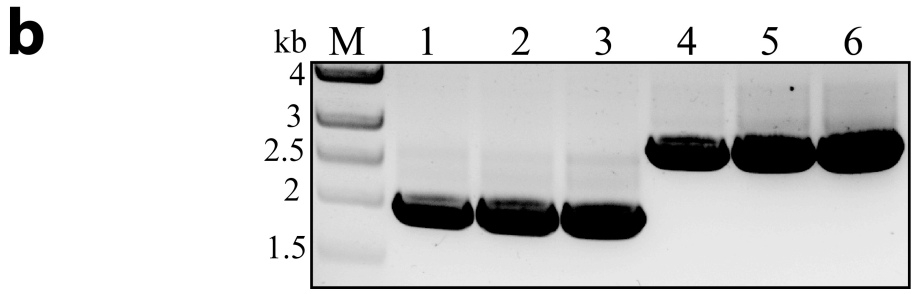


Figure S4. (a) The *BRCA1* gene loading into the alphoid^{tetO} -HAC vector propagated in Hprt-deficient hamster CHO cells by Cre-loxP mediated recombination. Loading of the targeting vector along with the *BRCA1* gene into the loxP site of the alphoid^{tetO} –HAC is accompanied by reconstitution of the *HPRT* gene allowing cells selection on HAT medium. **(b)** PCR reactions confirming reconstitution of the *HPRT* gene. Lanes 1, 2, 3, and 4, 5, 6 correspond to PCR products obtained with the genomic DNA isolated from alphoid^{tetO} -HAC /BRCA1-containing clones of CHO cells using a set of specific primers (Table S1). One clone was chosen for further MMCT transfer to human Brca1-deficient UWB1.289 cells.

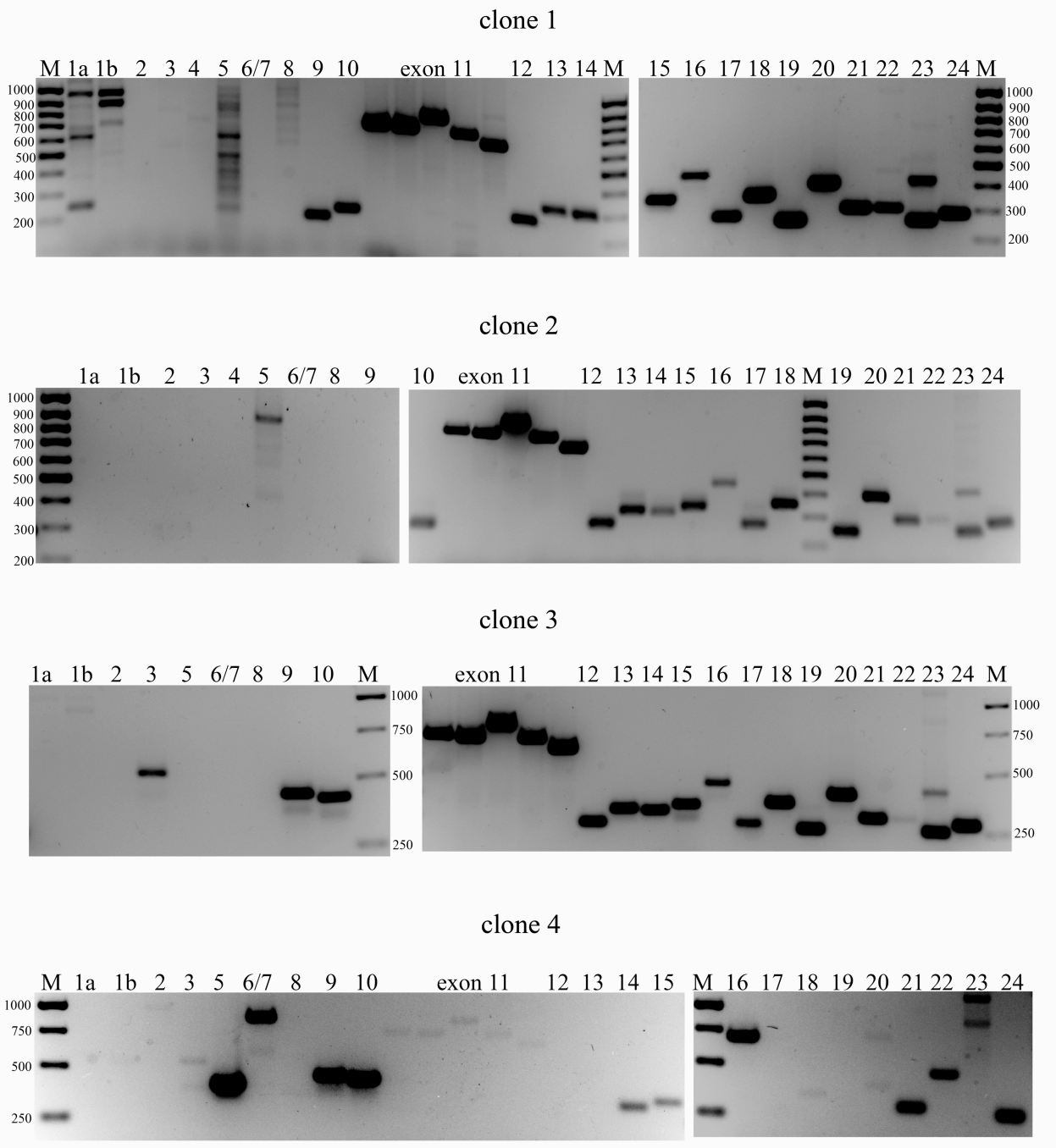


Figure S5. Integrity of the *BRCA1* gene after its loading into alphoid^{tetO} -HAC in CHO cells. PCR analysis of *BRCA1* exons is shown for four randomly chosen CHO clones selected on HAC medium. As seen, each clone contains a *BRCA1* sequence with a different size of internal deletions.

Figure S6. Sequence alignment of *BRCA1* mRNA obtained by RT-PCR from four CHO clones (1-4) containing the not arranged *BRCA1* sequences. As seen, the sequences are identical to a wild type mRNA of BRCA1. The sequence of the hamster *BRCA1* mRNA is also included.

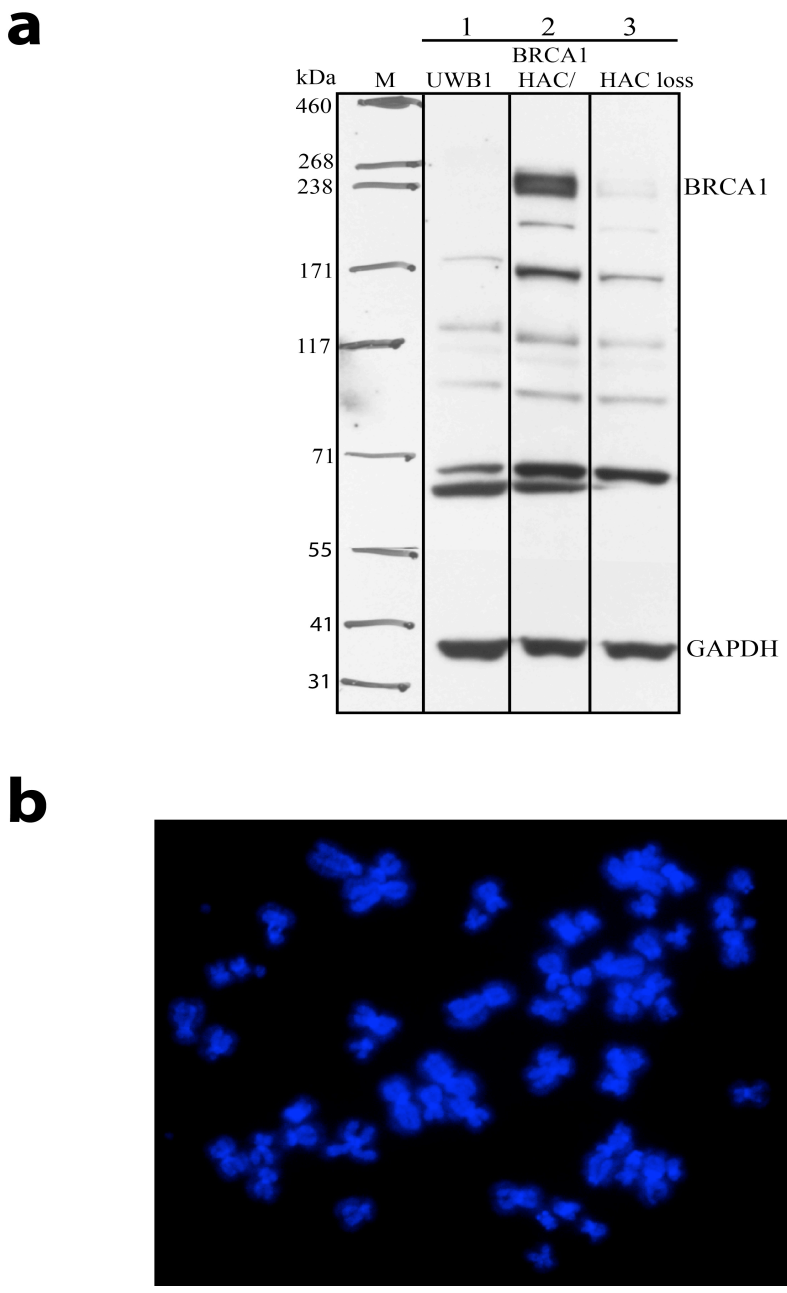


Figure S7. (a) Western blot analysis of alphoid^{tetO} -HAC/BRCA1-containing UWB1.289 cells (lane 2) and the same cells after elimination of the HAC (lane 3) by expression of the *tTS* fusion construct using human-specific Abs against BRCA1. Lanes 1 corresponds to the original UWB1.289 cell line. M-ladder. **(b)** FISH analysis of human UWB1.289 cells after expression of the tTS-tet-R fusion construct. FISH was performed using the labeled probe for tetO-alphoid sequences of the HAC. Chromosomal DNA was counterstained with DAPI (blue). As seen, the alphoid^{tetO} -HAC/BRCA1 is lost from the cells.

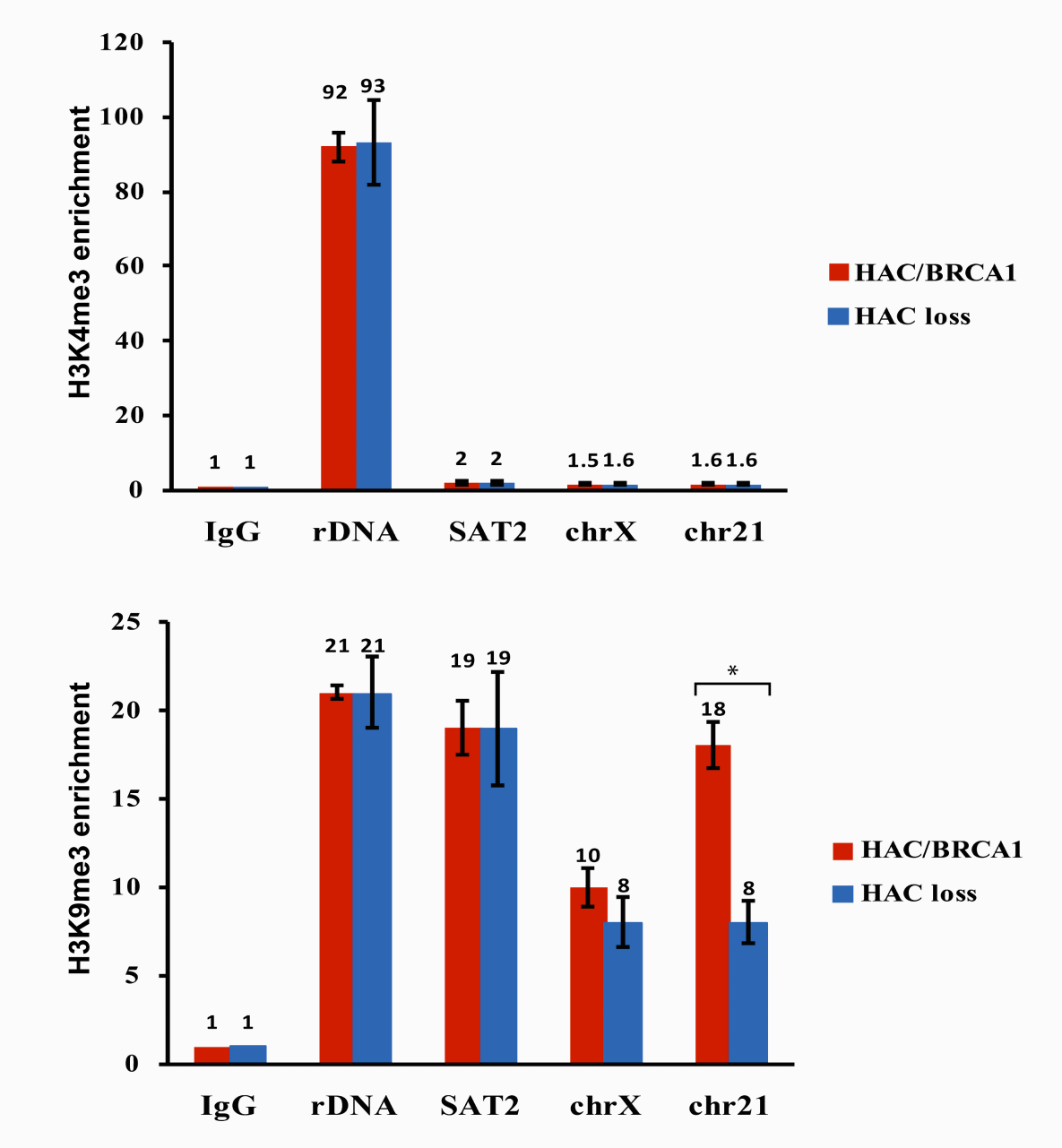


Figure S8. Chromatin immunoprecipitation (ChIP – PCR) analysis of HORs in chromosomes 21 and X using antibodies against H3K4me3 (upper) and H3K9me3 (lower) in the in Brca1-deficient UWB1.289 cells containing alphoid^{tetO} -HAC /BRCA1 and in the same cells that have lost the HAC.

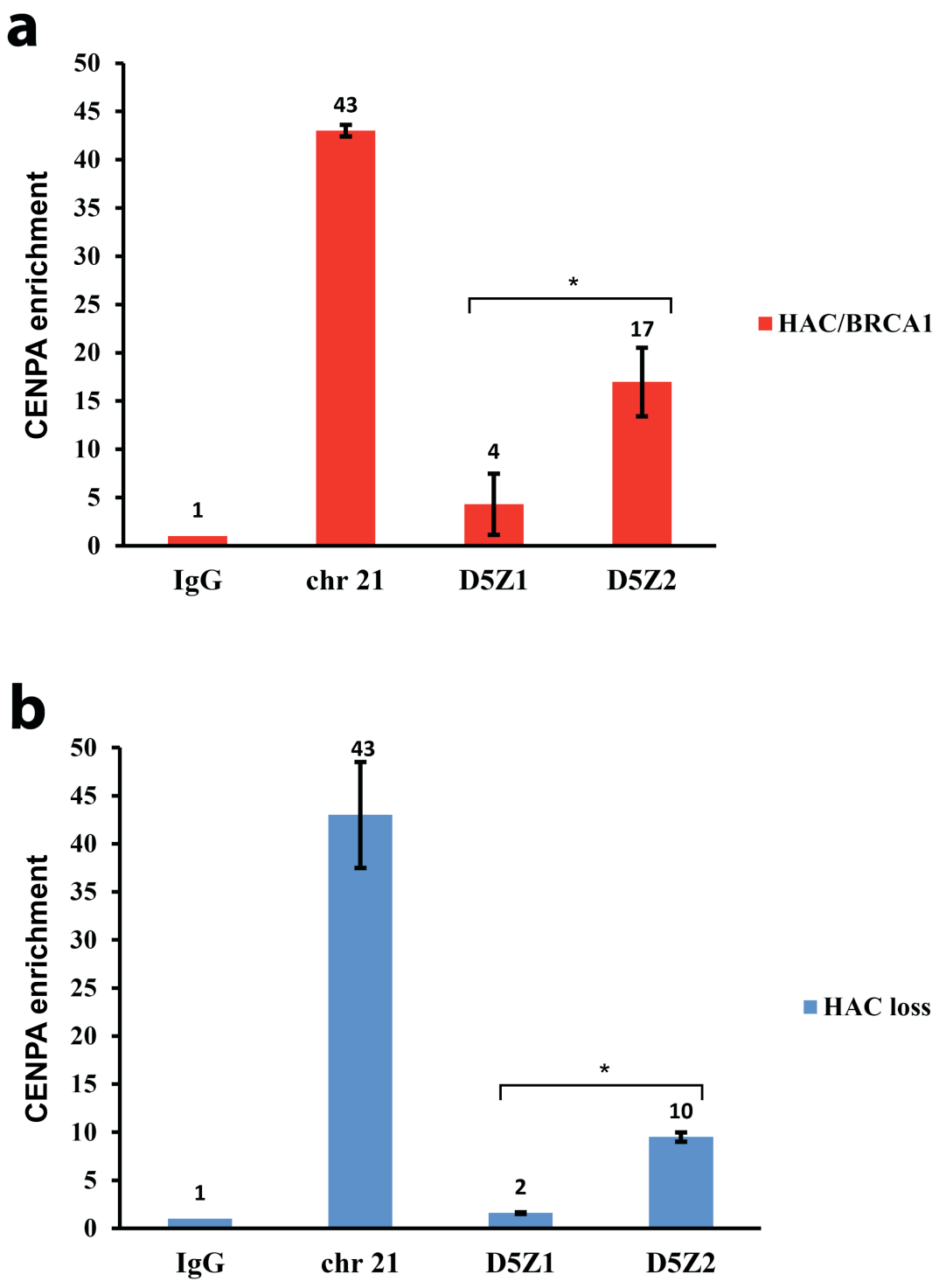


Figure S9. Chromatin immunoprecipitation (ChIP –PCR) analysis of different HORs using antibodies against CENP-A (centromeric protein A) in BRCA1-deficient UWB1.289 cells containing alphoid^{tetO} -HAC/BRCA1 and in the same cells that have lost the HAC. As seen, CENP-A chromatin is enriched on D5Z2 arrays.

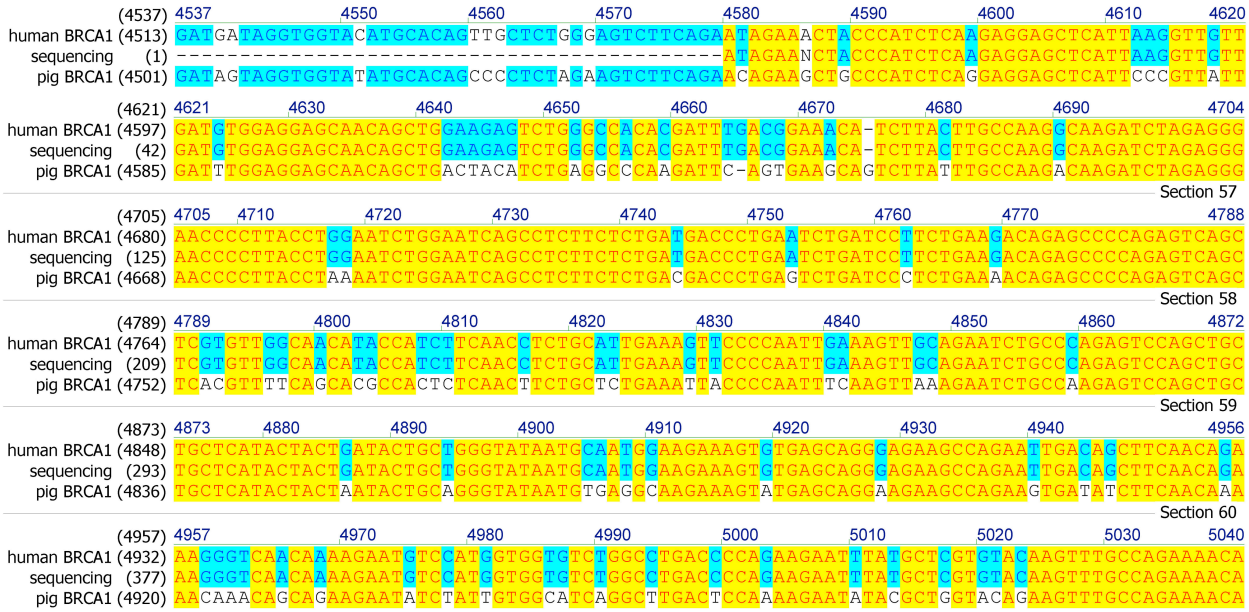


Figure S10. Alignment of human and porcine *BRCA1* mRNA sequences. For simplicity, the sequences corresponding to exons 15-17 are shown. A middle line corresponds to RT-PCR product risen from total mRNA isolated from ST porcine cells carrying the alphoid^{tetO} - HAC/BRCA1.

Table S1 Primers used in this study

Amplification of exons of the human BRCA1 gene	
Exon 1a_F	5’ ATTGGGACCTCTTCTTACGAC 3’
Exon 1a_rev	5’ GTCAGCTTCGGAAATCCAC 3’
Exon 1b_F	5’ AGAGTGGATTTCCGAAGCTG 3’
Exon 1b_rev	5’ CAAATGGATCCAGTTCTTAAGTTC 3’
BRCA1 Exon 2f-1	5’ TTTAAAATGATAAAATGAAGTTGTC 3’
BRCA1 Exon 2rev-1	5’ ATCATTTTCTATATTTTTAAAGTTCTTC 3’
BRCA1 Exon 3f-1	5’ TTGATATCTGAAAGCTCACTG 3’
BRCA1 Exon 3rev-1	5’ TTTCGTTCTCACTTAATTGAAG 3’
Exon 4F	5’ TCAAAGAGATAGAATGTGAGC 3’
Exon 4R	5’ CCCGTCTCTACAGAAAACAC 3’
BRCA1 Exon 5f-1	5’ AACAATTAGGAAACTATTGCTTG 3’
BRCA1 Exon 5rev-1	5’ AAAACTTTCAGGAAAATAACTTTG 3’
BRCA1 Exon 6/7f-1	5’ GTTTCTCAAACAATTTAATTTCAG 3’
BRCA1 Exon 6/7rev-1	5’ AAGAAGAAAACAAATGGTTTTAC 3’
BRCA1 Exon 8f-1	5’ GACATTTTAGTGTGTAAATTCCTG 3’
BRCA1 Exon 8rev-1	5’ ACAAATTATGACCAAGATTTTTG 3’
BRCA1 Exon 9f-1	5’ TGAACTCCTAACCTCAAATAATC 3’
BRCA1 Exon 9rev-1	5’ TTTACCTATATAACAAACTGCACATAC 3’
BRCA1 Exon 10f-1	5’ TTCATTTCAACTAGAAGTTTCTAAAG 3’
BRCA1 Exon 10rev-1	5’ TAGTCTAGTACCATTTAAATCTATCAGAC 3’
Exon 11_1F	5’ GTTTTTGAGTACCTTGTTATTTTTG 3’
Exon 11_1R	5’ ATTTTGTCTTCAATATTACTCTCTACTG 3’
Exon 11_2F	5’ CTCCAAATCAGTAGAGAGTAATATTG 3’
Exon 11_2R	5’ CTGTCATGTCTTTTACTTGTCTG 3’
Exon 11_3F	5’ GTAACAAGCCAAATGAACAG 3’
Exon 11_3R	5’ CCTCTGAACTGAGATGATAGAC 3’
Exon 11_4F	5’ TCTAGGTTTTGTCTATCATCTCAG 3’
Exon 11_4R	5’ CTAGGACTCCTGCTAAGCTC 3’

Exon 11_5F	5’ ACATTAAGGAAAGTTCTGCTG 3’
Exon 11_5R	5’ GCATAAACATTTAGCTCACTTC 3’
Exon 12F	5’ GTCCTGCCAATGAGAAGAAA 3’
Exon 12R	5’ TGTCAGCAAACCTAAGAATGT 3’
Exon 13F	5’ AATGGAAAGCTTCTCAAAGTA 3’
Exon 13R	5’ ATGTTGGAGCTAGGTCCTTAC 3’
Exon 14F	5’ CTAACCTGAATTATCACTATCA 3’
Exon 14R	5’ GTGTATAAATGCCTGTATGCA 3’
Exon 15F	5’ TGGCTGCCCAGCAAGTATG 3’
Exon 15R	5’ AACCAGAATATCTTTATGTAGGA 3’
Exon 16F	5’ AATTCTTAACAGAGACCAGAAC 3’
Exon 16R	5’ AAAACTCTTTCCAGAATGTTGT 3’
Exon 17F	5’ GTGTAGAACGTGCAGGATTG 3’
Exon 17R	5’ TCGCCTCATGTGGTTTTA 3’
Exon 18F	5’ GGCTCTTTAGCTTCTTAGGAC 3’
Exon 18R	5’ AGACCCATTTTCCCAGCATC 3’
Exon 19F	5’ CTGTCATTCTTCCTGTGCTC 3’
Exon 19R	5’ CATTGTTAAGGAAAGTGGTGC 3’
Exon 20F	5’ ATATGACGTGTCTGCTCCAC 3’
Exon 20R	5’ GGGAATCCAAATTACACAGC 3’
Exon 21F	5’ AAGCTCTTCCTTTTTGAAAGT 3’
Exon 21R	5’ GTAGAGAAATAGAATAGCCTCT 3’
Exon 22F	5’ TCCCATTGAGAGGTCTTGCT 3’
Exon 22R	5’ GAGAAGACTTCTGAGGCTAC 3’
Exon 23F	5’ CAGAGCAAGACCCTGTCTC 3’
Exon 23R	5’ ACTGTGCTACTCAAGCACCA 3’
Exon 24F	5’ ATGAATTGACACTAATCTCTG 3’
Exon 24R	5’ GTAGCCAGGACAGTAGAAGGA 3’

RT-PCR of the full-size human BRCA1 gene expressed from the HAC in CHO cells (the products were used for sequencing analysis)	
BRCA1RT1 (cDNA synthesis)	5’ ATGGATTTATCTGCTCTT 3’
BRCA1start	5’ ATGGATTTATCTGCTCTTCGCGTTG 3’
BRCA1RT1rev	5’ CATGTGAGTCATCAGAACCTAACAGTTC 3’
BRCA1RT2 (cDNA synthesis)	5’ ACAGAAAAAAAGGTAGATC 3’
BRCA1RT2f	5’ ACAGAAAAAAAGGTAGATCTGAATGCTG 3’
BRCA1RT2rev	5’ CAGCTCTGGGAAAGTATCGCTGTC 3’
BRCA1RT3: (cDNA synthesis)	5’ ATAAAGAAAAAAAAGTACAAC 3’
BRCA1RT3f	5’ CAACCAAATGCCAGTCAGGCAC 3’
BRCA1RT3rev	5’ ATTTCATTAATACTGGAGCCCACTTC 3’
BRCA1RT4: (cDNA synthesis)	5’ AAAACTTTGAGGAACATT 3’
BRCA1RT4f	5’ TTTGAGGAACATTCAATGTCACCTG 3’
BRCA1RT4rev	5’ GTTGCATGGTATCCCTCTGCTGAG 3’
BRCA1RT5: (cDNA synthesis)	5’ ATGATGAAGAAAGAGGAA 3’
BRCA1RT5f	5’ GAAAGAGGAACGGGCTTGGAAG 3’
Exons15-21rev	5’ AAGGGTGAATGATGAAAGC 3’
3': (cDNA synthesis)	oligo dT18 commercial primer (IDT)
Exons15-21f	5’ CATTAGATGATAGGTGGTA 3’
BRCA1stop	5’ TCAGTAGTGGCTGTGGGGGATC 3’
Sequence of BRCA1 cDNA	
BRCA1sec0	5’ CATCTTTTAGATGTTCAGGAGAG 3’
BRCA1sec2:	5’ GGGAGTCTGAATCAAATGCCAAAG 3’
BRCA1sec3	5’ GCAGCAGTATAAGCAATATGGAACTC 3’
BRCA1sec4	5’ GTATCTCGTTACTGGAAGTTAGCACTC 3’
BRCA1sec5	5’ TTTTACAAAACCCATATCGTATACCACC 3’
BRCA1sec6	5’ GGAAAGTTCTGCTGTTTTTAGCAAAAG 3’
BRCA1sec7	5’ AAGAGCAAAGCATGGATTCAAACTTAG 3’
BRCA1sec8	5’ GAGTCTGGGCCACACGATTTGAC 3’
BRCA1sec9	5’ CTGGGTGACCCAGTCTATTAAAGAAAG 3’
BRCA1rev1	5’ GCCTCACACATCTGCCCAATTG 3’

RT-PCR of hBRCA1 in CHO cells	
Exons 15–21f	5’ CATTAGATGATAGGTGGTA 3’
Exons 15–21 rev	5’ AAGGGTGAATGATGAAAGC 3’
ERCC2 f2	5’ CTTCAGTTGTATGGACGCCTCCTTG 3’
ERCC2 rev2	5’ CGAGCCACCGAGAGCAGAATG 3’
qPCR (SYBR green)	
h -cyclophilin1251F	5’ TGCAGTGAGTTCATGCATTTAGAG 3’
h-cyclophilin-1320R	5’ CGCAACAGATGTCTCAATTTTTG 3’
qPCR-PRKG1Bf	5’ GGGAAAAGATGCTTCTGGGAA 3’
qPCR-PRKG1Brev	5’ TTCGAAAGTGAAGCTCGGAAA 3’
ChXVl-F	5’ GTGACGATGGAGTTTAACTCAGGG 3’
ChXVl-R	5’ TGCTTCCGTTCAGTTATGGGAAG 3’
D5Z1forward	5’ TAGACAGAAATATTCTCACAATCGT 3’
D5Z1 reverse	5’ GCCCTCAAAGCGCTCCAAG 3’
D5Z2 forward	5’ TTTTTGTGCAATTGGCAAATGGAG 3’
D5Z2 reverse	5’ AGACTGTTTCCTCACTGCTCT 3’
21-IaF	5’ CTAGACAGAAGCCCTCTCAG 3’
21-IaR	5’ GGGAAGACATTCCCTTTTTCACC 3’
21-IbF	5’ GTAGTTTGTGGAAGTGGAC 3’
21-IbR	5’ CTGAGAATGCTGCTGTCTACC 3’
Satellite 2 F	5’ TCGCATAGAATCGAATGGAA 3’
Satellite 2 R	5’ GCATTCGAGTCCGTGGA 3’
qPCR (TaqMan)	
GAPDH	Hs.PT.39a.22214836 (IDT)
SPANXBf	5’ CCCCTGTGAATCCAACGAG 3’
SPANXBprobe	/56-FAM/CCG CAA CCT /ZEN/GCT CCT AAA AAA ATG /3IABkFQ/
SPANXBrev	5’ CCTGTAGCGAACCACTAGTATG 3’
PRB.DUX4	/56-FAM/TCA CCG GAT /ZEN/CCC AGA CCG C/3IABkFQ/

FOR.DUX4	5’ TTCAGAATGAGAGGTCACGC 3’
REV.DUX4	5’ CTTCTCAAAGGCTCGGAGG 3’
Pig specific primers	
Pig specific f	5’ TTAAGTGGAGGAAGAAGG 3’
Pig specific rev	5’ CATTATGACAGTTAAGCGG 3’
Porcine f	5’ GCCTAAATCTCCCCTCAATGGTA 3’
Porcine rev	5’ ATGAAAGAGGCAAATAGATTTTCG 3’
HPRT gene reconstitution	
Lox137-R	5’ AGCCTTCTGTACACATTTCTTCTC 3’
Rev #6	5’ GCTCTACTAAGCAGATGGCCACAGAACTAG 3’
SV40 PA term rev	5’ AATGGTTACAAATAAAGCAATAGCATCAC 3’
Hamster specific B2 repeats	
Cons B2-F	5’ CCATCTGTAATGAGATCTGATGC 3’
Ham B2-F	5’GCTCAGAGGTTAAGAGCACTGAC 3’
Ham B2-R	5’TGCTTCCATGTATATCTGCACAC 3’