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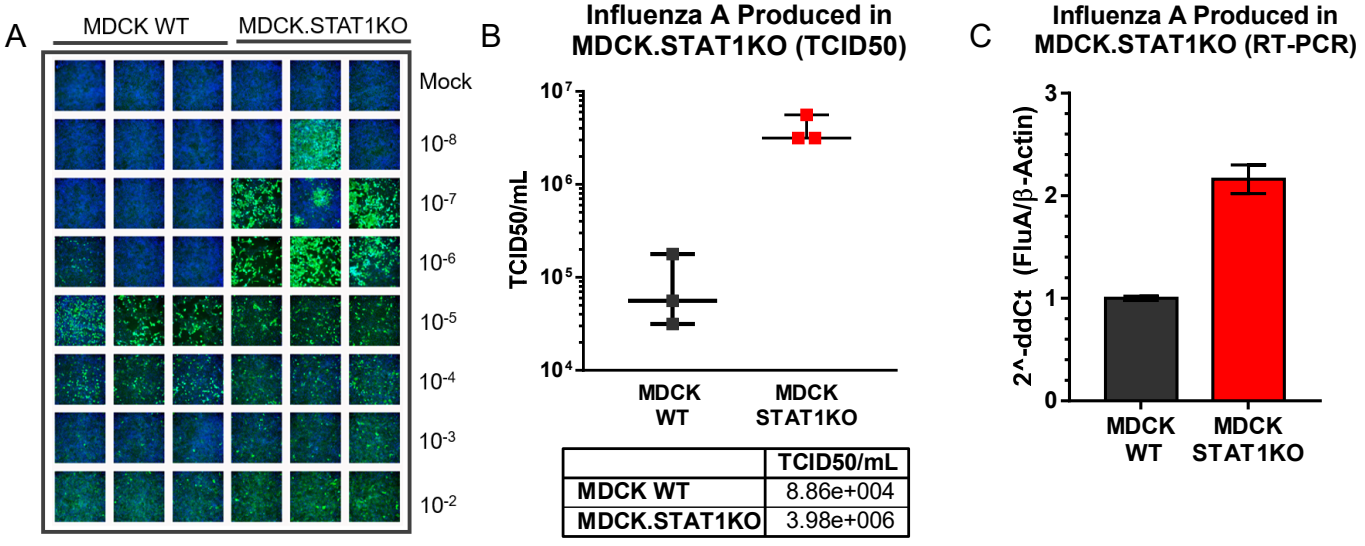
Technical Data Sheet: MDCK.STAT1KO

ATCC® Number	CCL-34-VHG™
Organism	*Canis familiaris*, dog
Tissue/Disease Source	Kidney, normal
Product Description	This STAT1 knockout MDCK cell line was derived from the parental MDCK cell line (ATCC® CCL-34™) at ATCC using CRISPR-Cas9 gene editing technology. This cell line carries short nucleotide insertions and deletions in the fourth exon of the STAT1 gene. This cell line does not express STAT1 protein.
Application	MDCK.STAT1 KO is an excellent cell model for virus propagation and viral vaccine production. It exhibits significant increased viral titer and enhanced virus production capability when compared to its parental cell line.

Increased Production of Influenza A (H1N1) in MDCK.STAT1KO Cells



	TCID50/mL
MDCK WT	8.86e+004
MDCK.STAT1KO	3.98e+006

Figure 1: Influenza A (H1N1; ATCC® VR-1736™) produced in STAT1 KO MDCK cells. (A) Immuno-stained $TCID_{50}$ of viral supernatants produced by WT parental and MDCK.STAT1 KO cells. Cells were infected with Influenza A at an MOI of 0.01. Supernatants were collected 48 hours after infection and used to re-infect WT MDCK cells at the indicated dilution. Green – anti-Influenza A stain, Blue – nuclear stain. Cells were infected with Influenza A at an MOI of 0.01 for 48h, then (B) calculated $TCID_{50}$ of Influenza A viral supernatants produced in MDCK.STAT1KO cells at 48h post-infection. (C) RT-PCR quantification of Influenza A viral genomes produced in MDCK.STAT1KO cells at 48h post-infection.

Cell Morphology

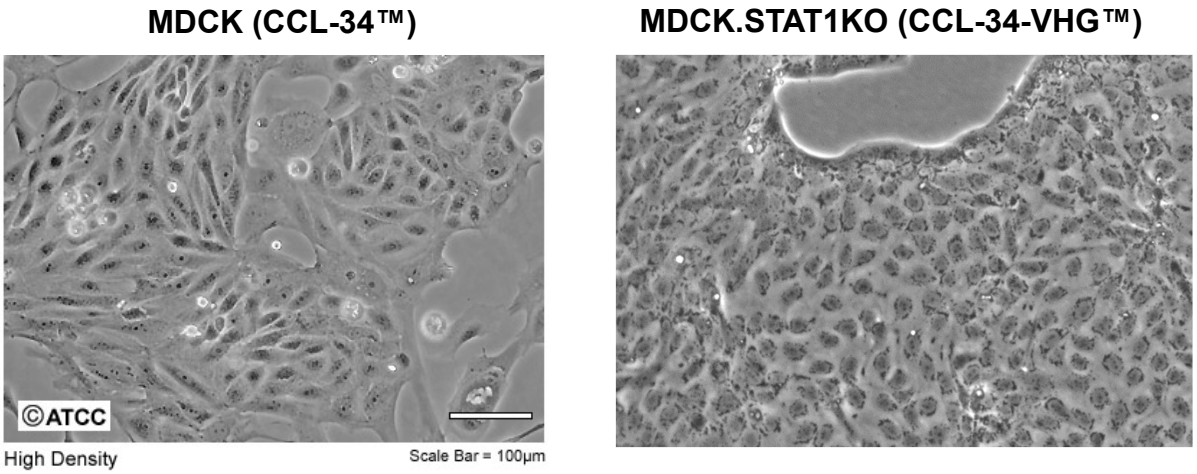


Figure 2: Cell morphology of parental MDCK and MDCK.STAT1KO cells. Cells were maintained in ATCC recommended culture conditions to maximum recommended cell density and then imaged by light microscopy.

Characterization of STAT1 Knockout

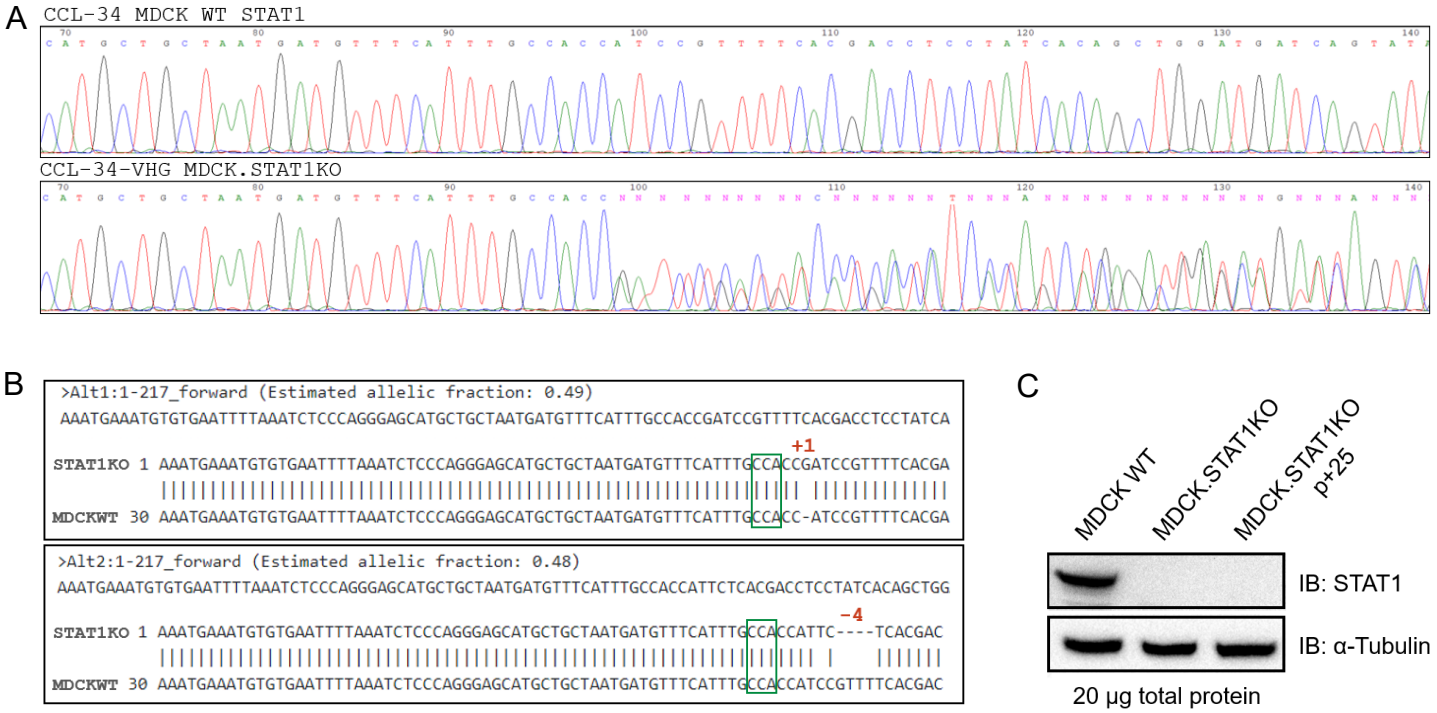


Figure 3: Molecular characterization of STAT1 KO MDCK cells. (A) Sanger sequencing of the STAT1 gene of MDCK.STAT1KO cells confirms STAT1 gene disruption. (B) Deconvolution of the mixed Sanger sequence chromatogram from MDCK.STAT1KO demonstrates that the knockout genotype is heterozygous, with one STAT1 allele carrying a single nucleotide insertion and the other carrying a four nucleotide deletion (red text) proximal to the PAM motif (green box) used for gene editing. (C) STAT1 immunoblot of total cellular protein from parental MDCK WT and MDCK.STAT1KO cells confirms absence of STAT1 protein expression in MDCK.STAT1KO at both high and low passages.