Supporting Information

The G protein-coupled receptor GPR35 suppresses lipid accumulation in hepatocytes

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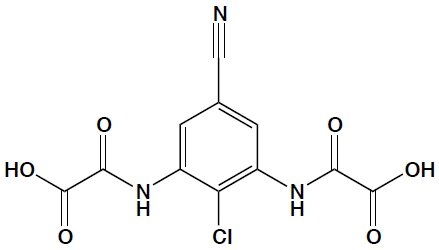

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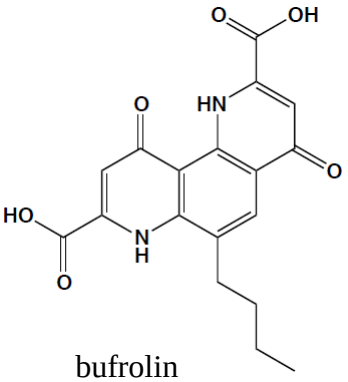

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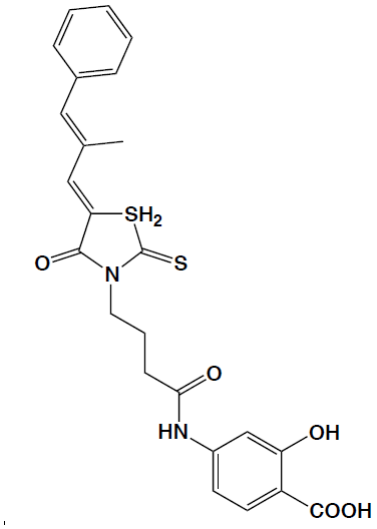

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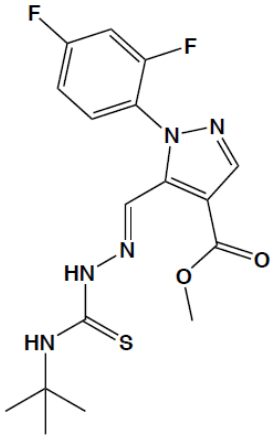

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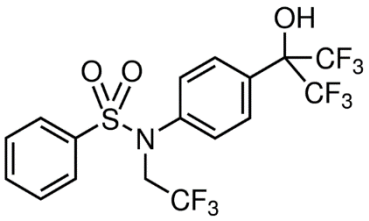

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Figure S1. Chemical structure of GPR35 agonists, antagonists and LXR activator.

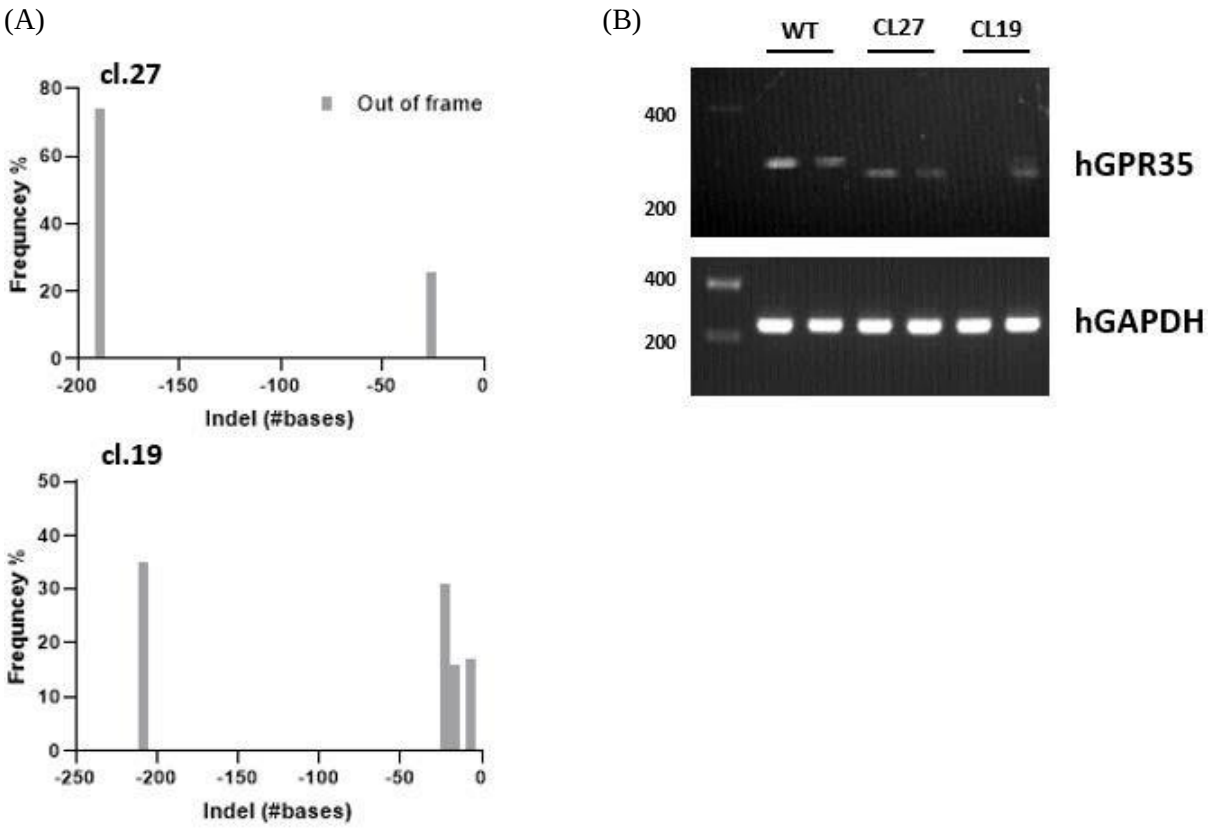

Figure S2. CRISPR-Cas9 genome editing produces HepG2 clones lacking expressing of GPR35

HepG2 cells were subjected to CRISPR-Cas9-mediated genome-editing targeting the GPR35 gene. Next generation sequencing of various clones identified disruption of the GPR35 gene sequence within the open-reading frame. **A.** clone 27 contains 1 larger deletion and 1 smaller deletion in the exon and which is out of frame (**upper**) whilst clone 19 contains 1 large deletion and 3 smaller deletions (**lower**) that all are in within the exon and out-of-frame. (**B**). RT-PCR confirmed deletion of sequence of GPR35b in both these clones but was able to detect a fragment of smaller size than for full length GPR35b. hGAPDH served as a control.

Table S1

gRNA sequence	
gRNA pair	AGGTCAGCAGAGAGTGAGCAGGG
	TCTCCGTCCACTGCTGCATGCGG
PCR primers for genome deletion efficiency check	
F'	CACCCATGCTTTCTTTGAGGAGTT (FAM labelled)
R'	AGCGGCGTGTCTGAGGTGTC
wt size: 412 bp expected deletion size: 189 bp	
NGS primers	
F'	TCGTCGGCAGCGTCAGATGTGTATAAGAGACAGGGCTCACCTCCTCCCACATC
R'	GTCTCGTGGGCTCGGAGATGTGTATAAGAGACAGGCACAGCAGGCAGAGGTC
RT-PCR primers	
F'	GATCAAGCTGGGCTTCTACG
R'	CAGGCTGATGCTCATGTACC

Table S2

qRT-PCR primers	
human GPR35-specific	
F'	GTGCCCTCCTGGAGACGAT
R'	GCAGCAGTTGGCATCTGAGA
mouse GPR35-specific	
F'	CCAAGATTCCCAGATCCTGA
R'	CTTGCTCACATCACAGGTTCC
actin	
F'	GACAGGATGCAGAAGGAGATTACTG
R'	CTCAGGAGGAGCAATGATCTTGAT
RT-PCR primers	
human GPR35a	
F'	ATGCTGGCTCTTCAGAGGTG
R'	GATGACCAGCACCCAGAGG
human GPR35b	
F'	TGCTTCATAGTCCTTGCGTCTC
R'	GATGACCAGCACCCAGAGG
human GAPDH	
F'	GAGTCAACGGATTTGGTCGT
R'	TTGATTTTGGAGGGATCTCG
Genotyping primers	
humanised GPR35 transgene	
F'	CGGCACAATTTCAACTCCATGG
R'	GGGGAGGGGTGTATCCTAAA
wild type allele	
F'	TGAACCTCAATACCTGTGCTGC
R'	GGGGAGGGGTGTATCCTAAA