

Cyp27a1 Cas9-CKO Strategy

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Project Overview

Project Name

Cyp27a1

Project type

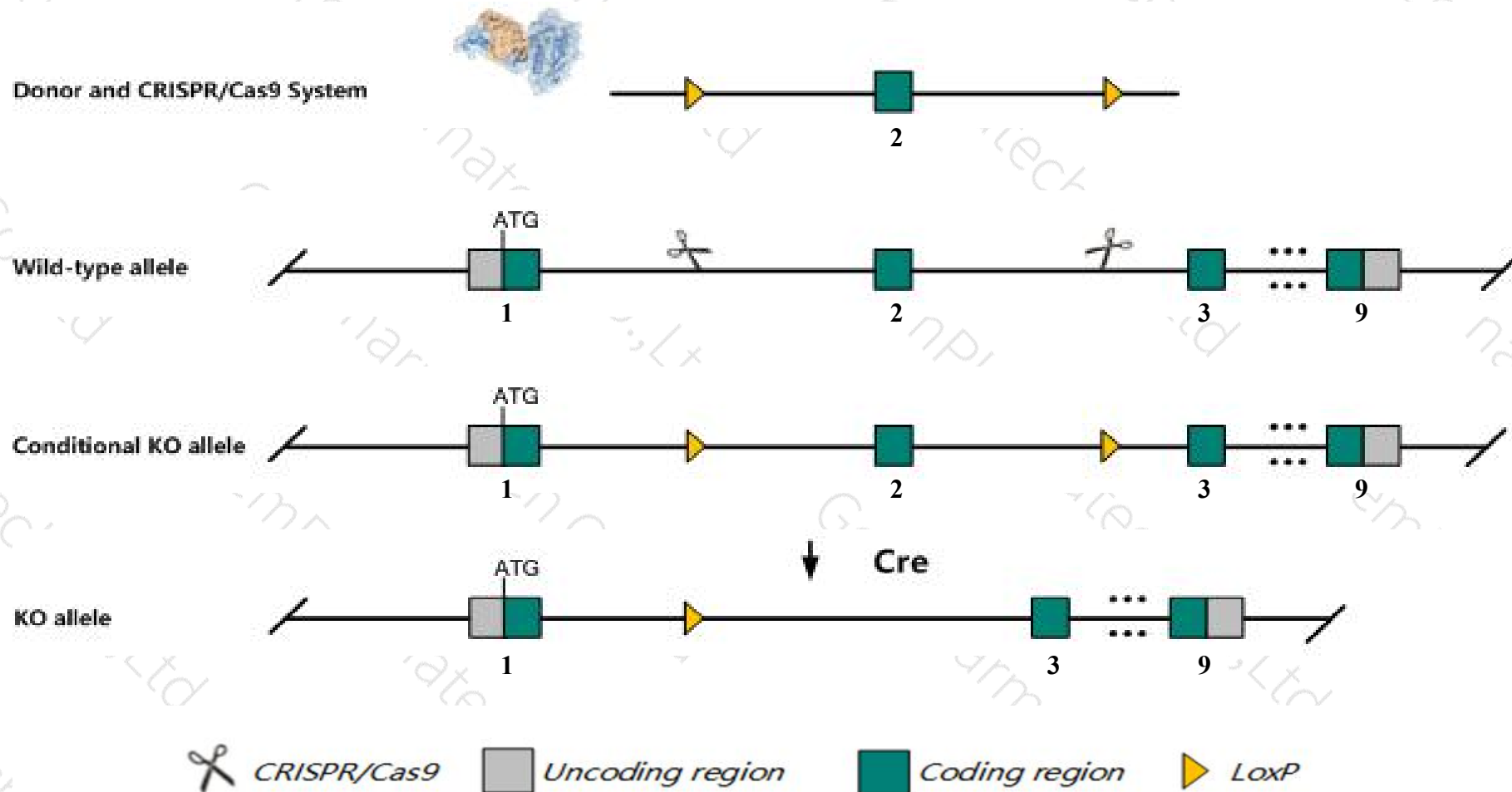
Cas9-CKO

Strain background

C57BL/6JGpt

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Cyp27a1* gene. The schematic diagram is as follows:



Technical routes

- The *Cyp27a1* gene has 3 transcripts. According to the structure of *Cyp27a1* gene, exon2 of *Cyp27a1-201* (ENSMUST00000027356.6) transcript is recommended as the knockout region. The region contains 191bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Cyp27a1* gene. The brief process is as follows: CRISPR/Cas9 system and Donor were microinjected into the fertilized eggs of C57BL/6JGpt mice. Fertilized eggs were transplanted to obtain positive F0 mice which were confirmed by PCR and sequencing. A stable F1 generation mouse model was obtained by mating positive F0 generation mice with C57BL/6JGpt mice.
- The flox mice will be knocked out after mating with mice expressing Cre recombinase, resulting in the loss of function of the target gene in specific tissues and cell types.

- According to the existing MGI data, Homozygous null allele show hepato- and adrenomegaly, reduced bile acid synthesis, increased cholesterol 7alpha-hydroxylase activity and 7alpha-hydroxycholesterol levels, slightly higher 25-hydroxyvitamin D levels, and altered hepatic fatty acid, triacylglycerol, and adrenal cholesterol homeostasis.
- The N-terminal of *Cyp27a1* gene will remain 87aa, it may remain the partial function of *Cyp27a1* gene.
- Transcript *Cyp27a1*-202&203 may not be affected.
- The *Cyp27a1* gene is located on the Chr1. If the knockout mice are crossed with other mice strains to obtain double gene positive homozygous mouse offspring, please avoid the two genes on the same chromosome.
- This Strategy is designed based on genetic information in existing databases. Due to the complexity of biological processes, all risk of loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at existing technological level.

Gene information (NCBI)

Cyp27a1 cytochrome P450, family 27, subfamily a, polypeptide 1 [Mus musculus (house mouse)]

Gene ID: 104086, updated on 19-Mar-2019

Summary



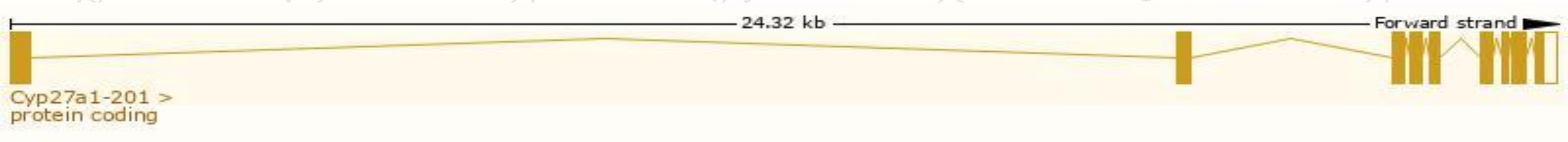
Official Symbol	Cyp27a1 provided by MGI
Official Full Name	cytochrome P450, family 27, subfamily a, polypeptide 1 provided by MGI
Primary source	MGI:MGI:88594
See related	Ensembl:ENSMUSG00000026170
Gene type	protein coding
RefSeq status	VALIDATED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	1300013A03Rik, Cyp27
Expression	Biased expression in liver adult (RPKM 72.0), bladder adult (RPKM 14.6) and 14 other tissues See more
Orthologs	human all

Transcript information (Ensembl)

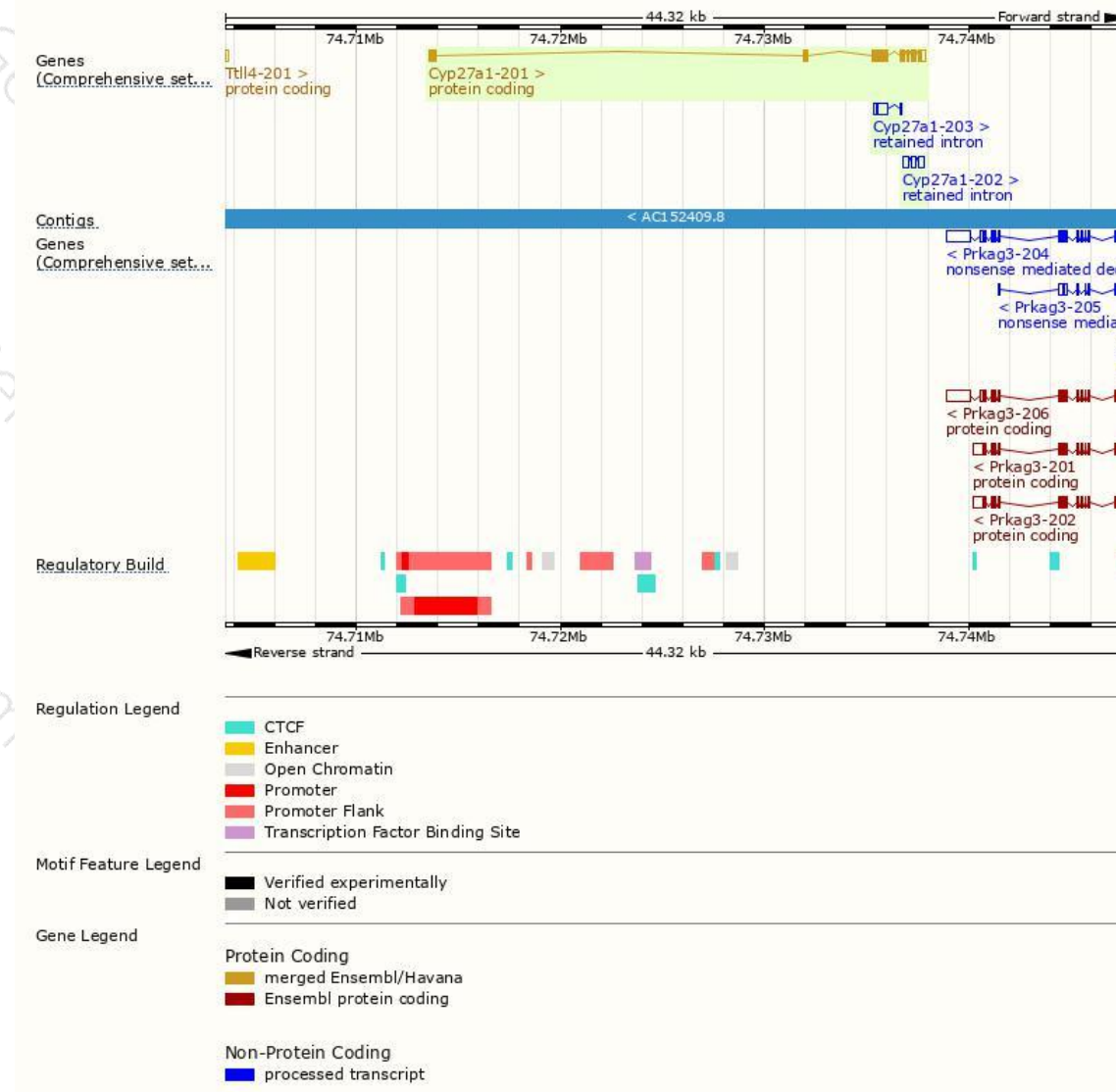
The gene has 3 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Cyp27a1-201	ENSMUST00000027356.6	1892	533aa	Protein coding	CCDS15054	Q9DBG1	TSL:1 GENCODE basic APPRIS P1
Cyp27a1-202	ENSMUST00000189083.1	784	No protein	Retained intron	-	-	TSL:2
Cyp27a1-203	ENSMUST00000190781.1	645	No protein	Retained intron	-	-	TSL:3

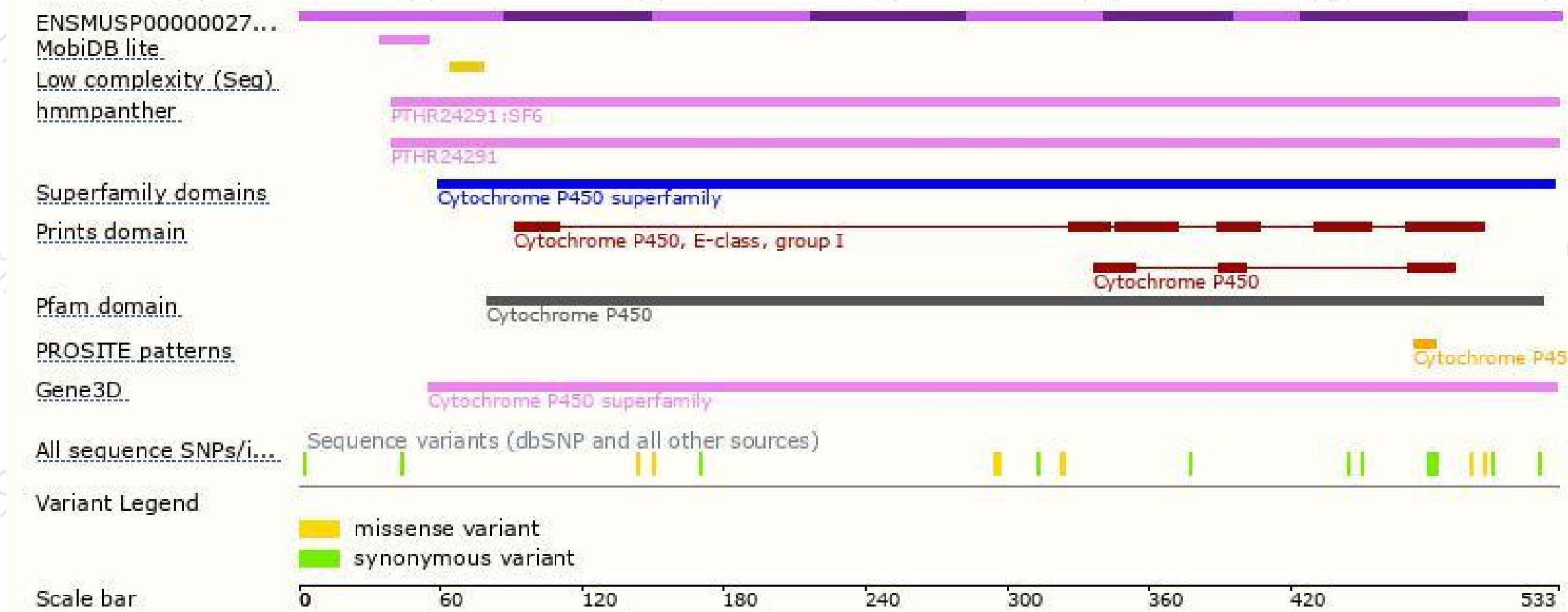
The strategy is based on the design of *Cyp27a1-201* transcript,The transcription is shown below



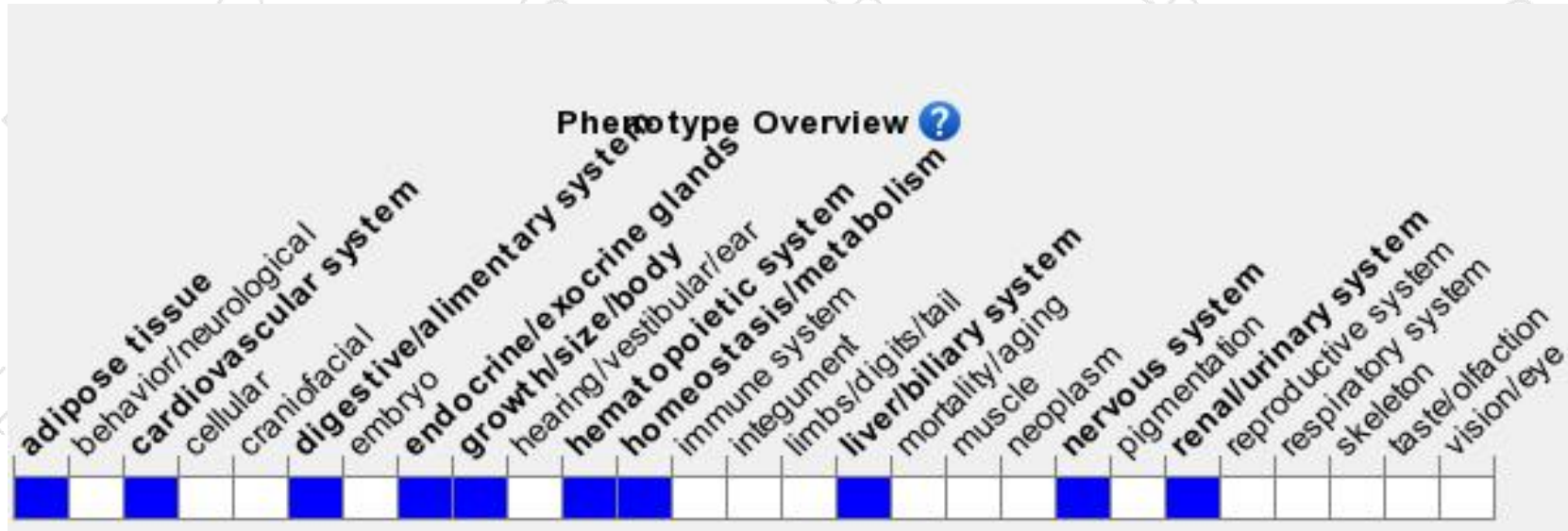
Genomic location distribution



Protein domain



Mouse phenotype description(MGI)



Phenotypes affected by the gene are marked in blue. Data quoted from MGI database(<http://www.informatics.jax.org/>).

According to the existing MGI data, Homozygous null allele show hepato- and adrenomegaly, reduced bile acid synthesis, increased cholesterol 7 α -hydroxylase activity and 7 α -hydroxycholesterol levels, slightly higher 25-hydroxyvitamin D levels, and altered hepatic fatty acid, triacylglycerol, and adrenal cholesterol homeostasis.

If you have any questions, you are welcome to inquire.

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