Supplemental Table 1

Patients and plasma parameters.

Plasma samples of 20 patients suffering from obstructive jaundice of different etiology were assayed for Lp(a), total bile acids and other variables of cholestasis before and after treatment of biliary obstruction. For details see Results section.

Patient	sex	age	Diagnosis	Bilirubin	[7]TBA (µmol/l)		LP-X (mg/dl) [§]		Lp(a) number	Lp(a) (mg/dl) [†]	
	m/f	(y)		µmol/l	before therapy	after therapy	before therapy	after therapy	of K-IV repeats	before therapy	after therapy
1	m	52	PC[2]	634	135,4	12,8	744	5,8	26	0	14,9
2	m	66	PC	488	121,6	5,7	268	0	16/22	9,6	64,4
3	f	49	PC	701	177,3	9,6	135	0	20	0	35,8
4	m	61	PC	285	85,3	5,5	449	0	22/26	0	5,3
5	m	55	PC	322	96,7	11,4	220	4,2	25	0	11,5
6	f	69	PC	680	135,1	6,6	376	5,5	27	2,4	17,6
7	m	72	PC	726	186,8	8,9	821	0	17/19	5,6	48,6
8	f	58	GBC[3]	189	78,6	4,3	209	0	17/30	0	5
9	m	52	GBC	112	56,4	5,2	283	0	22/28	0	11,9
10	m	70	GBC	398	145,9	8,7	645	5,1	28/37	0	6,4
11	m	39	GBC	188	79,5	6,3	489	0	17	13,3	54
12	f	73	BDC[4]	95	67,8	10	377	0	29/40	2,2	11,8
13	f	62	BDC	174	93,3	5,3	245	0	23/28	0	8,2
14	m	67	BDC	229	117,5	6,1	513	4,7	24	0	20,4
15	f	4w[1]	CBA[5]	183	45,8	2,8	75	8,6	22	0	4,7
16	m	42	CD[6]	198	65,9	4,2	281	0	20/29	4,4	11,3
17	m	39	CD	331	111,3	5	657	0	20	0	6,7
18	f	51	CD	117	48,2	3,9	125	0	26	0	3,8
19	m	48	CD	98	59,6	11,4	268	4,9	18/28	15,7	57,3
20	m	44	CD	176	70,3	8,7	224	0	22	0	6,3
Mean				**316,20**	**98,915**	**7,1‡**	**370,2**	**1,9‡**		**2,7**	**20,3‡**
SEM				**48,0**	**9,2**	**0,6**	**47,7**	**0,6**		**1,1**	**4,4**

[1] Newborn child at 4 weeks of age; [2]PC: pancreas carcinoma; [3]GBC: gallbladder carcinoma; [4]BDC: bile duct carcinoma; [5]CBA: congenital biliary atresia; [6]CD: choledocholithiasis; [7]TBA: total plasma bile acids.

[§]Values below the accuracy limit of the LP-X assay (3 mg/dl) were set zero; [†]Values below 1 mg/dl were set zero; [‡]significantly different from values before therapy ($p<0.001$).

Supplemental Table 2

Effect of common bile duct ligation (CBDL) on liver enzymes in tg-*APOA* mice.

Tg-*APOA* mice were subjected to biliary obstruction by common bile duct ligation (CBDL) (n=3 per group) or sham operation (n=4 per group) for 3 days. Serum levels of liver enzymes were measured as indicators of cholestasis. CBDL resulted in significantly elevated serum liver enzymes compared to sham operated mice.

Variable	Sham	CBDL
Alanine aminotransferase (U/L)	31 ± 1.7	2230 ± 560[a]
Alkaline phosphatase (U/L)	73.5 ± 9	2858 ± 781[a]
aspartate aminotransferase (U/L)	90.7 ± 49	3289 ± 396[a]

Values are expressed as means ± SD .

[a] *P*<0.001, compared to sham operated mice.

Supplemental Table 3

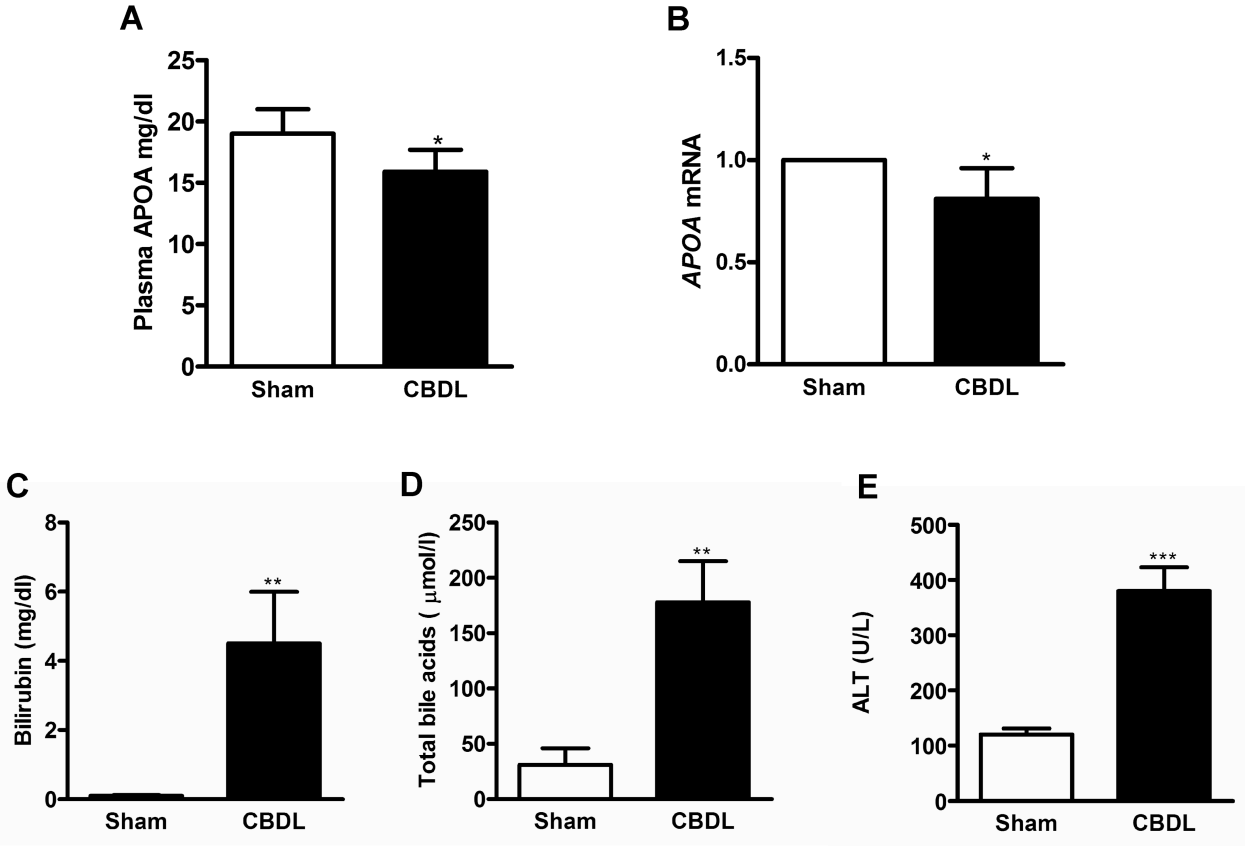
Primer Sequences used for real-time quantitative PCR

Gene Name	Forward primer (5'→3')	Reverse primer (5'→3')
Apoa1(Mouse)	GGCACGTATGGCAGCAAGAT	CCAAGGAGGAGGATTCAAACTG
Bsep(Mouse)	ACAGAAGCAAAGGGTAGCCATC	GGTAGCCATGTCCAGAAGCAG
Cyp7a1(Mouse)	GGGATTGCTGTGGTAGTGAGC	GGTATGGAATCAACCCGTTGTC
Cyp8b1(Mouse)	CCTCTGGACAAGGGTTTTGTG	GCACCGTGAAGACATCCCC
Ppia(Mouse)	TTCCAGGATTCATGTGCCAG	CCATCCAGCCATTCAGTCTT
Cyp3a11(Mouse)	AGCAGGGATGGACCTGG	CGGTAGAGGAGCACCAA
Fgf15(Mouse)	GCCATCAAGGACGTCAGCA	CTTCCTCCGAGTAGCGAATCAG
Hnf4a(Mouse)	CACGCGGAGGTCAAGCTAC	CCCAGAGATGGGAGAGGTGAT
*Il*b(Mouse)	GCAACTGTTCCTGAACTCAACT	ATCTTTTGGGGTCCGTCAACT
Il6(Mouse)	CTGCAAGAGACTTCCATCCAG	AGTGGTATAGACAGGTCTGTTGG
APOA(Human)	CAGCACGTTCATTCCAGGGA	CACCAGGGACCATTGATGTCA
*Lrh*1(Mouse)	GAACTGTCCAAAACCAAAAAAGG	CGTTTTCTCTGCGTTTTGTCA
Shp(Mouse)	TGGGTCCCAAGGAGTATGC	GCTCCAAGACTTCACACAGTG
Tnfa(Mouse)	GCCACCACGCTCTTCTGTCT	GGCTACAGGCTTGTCACTCG

Supplemental Figure 1

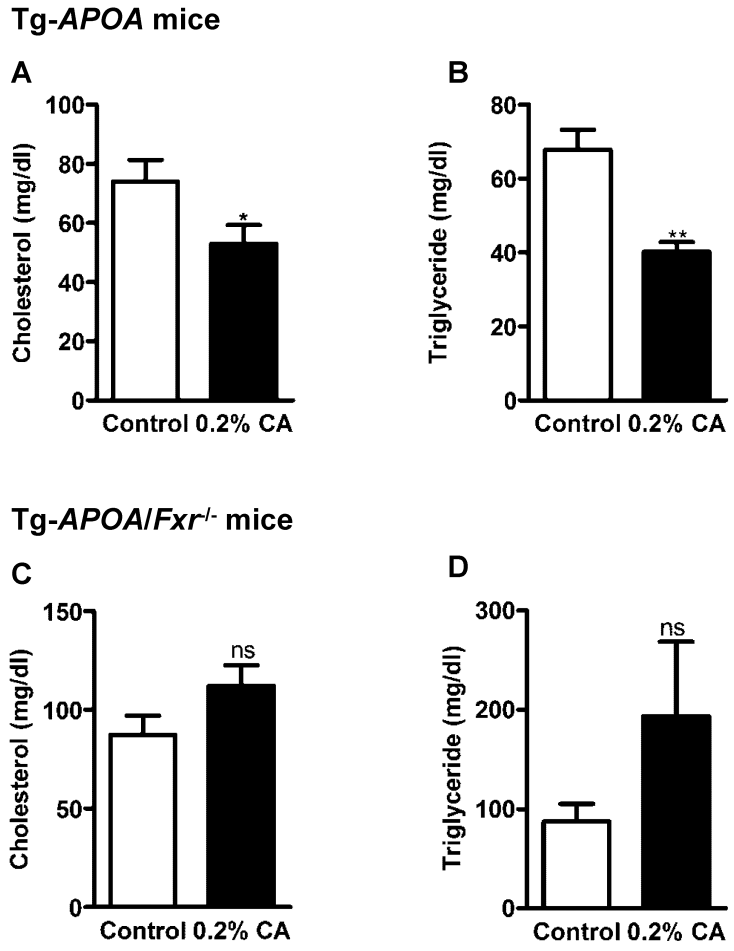
Common bile duct ligation in tg-*APOA*/*Fxr*$^{-/-}$ mice.

Tg-*APOA*/*Fxr*$^{-/-}$ mice were subjected to biliary obstruction by common bile duct ligation (CBDL) (n=3 per group) or sham operation (n=3 per group) for 3 days. (**A**) Plasma levels of APOA were measured by DELFIA and expressed as mean ± SD (* p≤0.05). (**B**) Liver *APOA* mRNA levels were analyzed by real-time quantitative PCR normalized to cyclophilin and expressed relative to sham operated mice. Results represent the mean ± SEM (* p≤0.05). Serum levels of bilirubin (**C**), total bile acids (**D**) and alanine aminotransferase (ALT) (**E**) were measured. Data are presented as mean ± SD. *** p≤0.001, ** p ≤0.01 when compared to sham operated mice.



Supplemental Figure 2

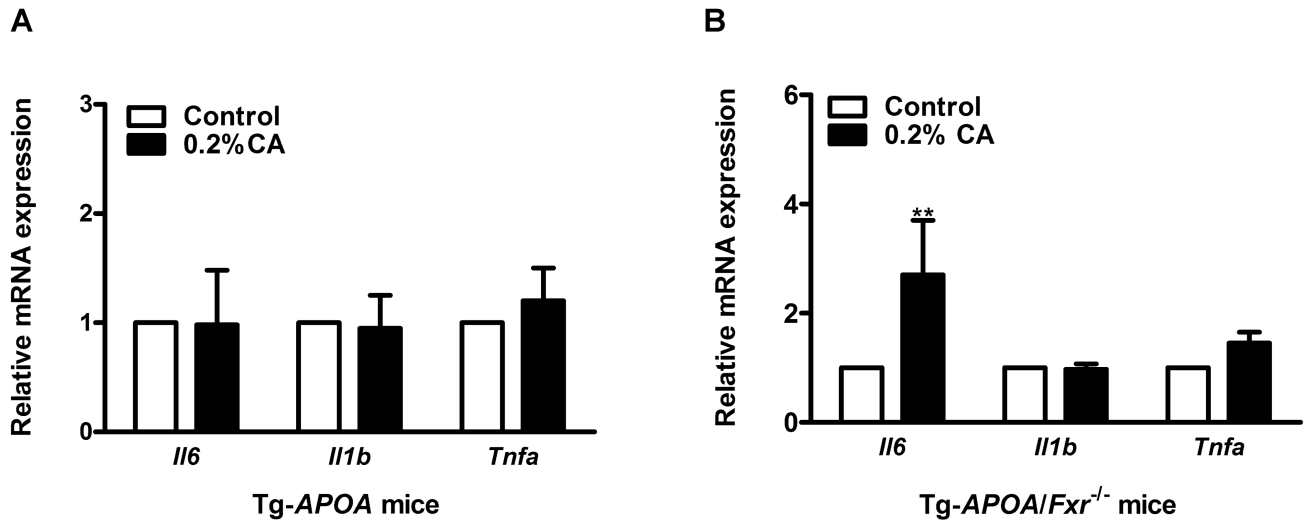
0.2% CA feeding decreases plasma total cholesterol and triglyceride levels in APOA transgenic mice. Tg-*APOA* and tg-*APOA*/*Fxr*$^{-/-}$ mice were fed a diet containing 0.2% CA (wt/wt) mixed in standard rodent chow for 5 days. (**A** and **C**) Total cholesterol, (**B** and **D**) triglycerides levels were measured in plasma after 4 h fasting period as described in *Materials and Methods*. Data are presented as mean ± S.D. (n = 8). ** p ≤0.01, * p<0.05, ns, statistically not significant when compared with chow fed control group.



Supplemental Figure 3

Influence of CA feeding on the expression of inflammatory genes.

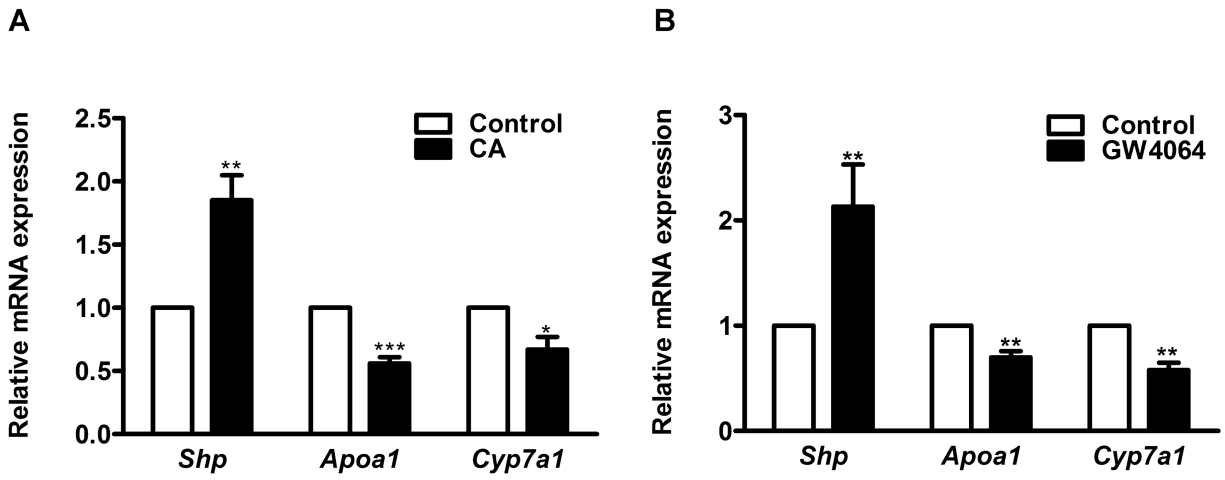
As bile acid signaling can result in hepatic inflammatory gene expression *in vivo*, we profiled hepatic expression of pro-inflammatory marker genes. The mRNA levels of *Il6*, *Il1b* and *Tnfa* were analyzed in tg-*APOA* and tg-*APOA*/*Fxr*$^{-/-}$ mice (**A** and **B**) upon 0.2% CA supplementation by real-time quantitative PCR and normalized to cyclophilin. Values are reported as mean ± SEM. Data indicate the relative mRNA expression in comparison with chow fed control mice (** $p \leq 0.01$).



Supplemental Figure 4

Gene expression profiling in primary mouse hepatocytes treated with natural and synthetic FXR agonists.

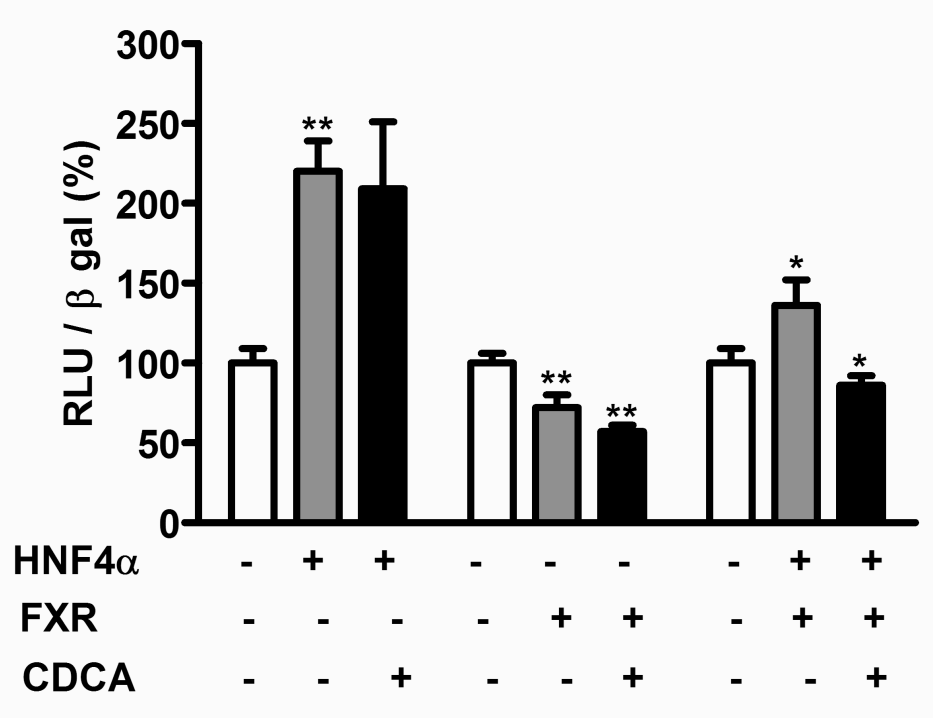
Primary mouse hepatocytes from tg-*APOA* mice were incubated with (**A**) CA (200 µM) or (**B**) GW4064 (5 µM) or vehicle (control) for 24 h. mRNA levels of previously reported FXR control genes *Shp, Apoa1 and Cyp7a1* were analyzed by real-time quantitative PCR normalized to cyclophilin and expressed relative to control treated cells. Values are expressed as mean ± SEM. (*** p≤0.001, ** p ≤0.01, * p<0.05).



Supplemental Figure 5

Effects of FXR and HNF4α overexpression on h*APOA* promoter activity in a non hepatic cell line.

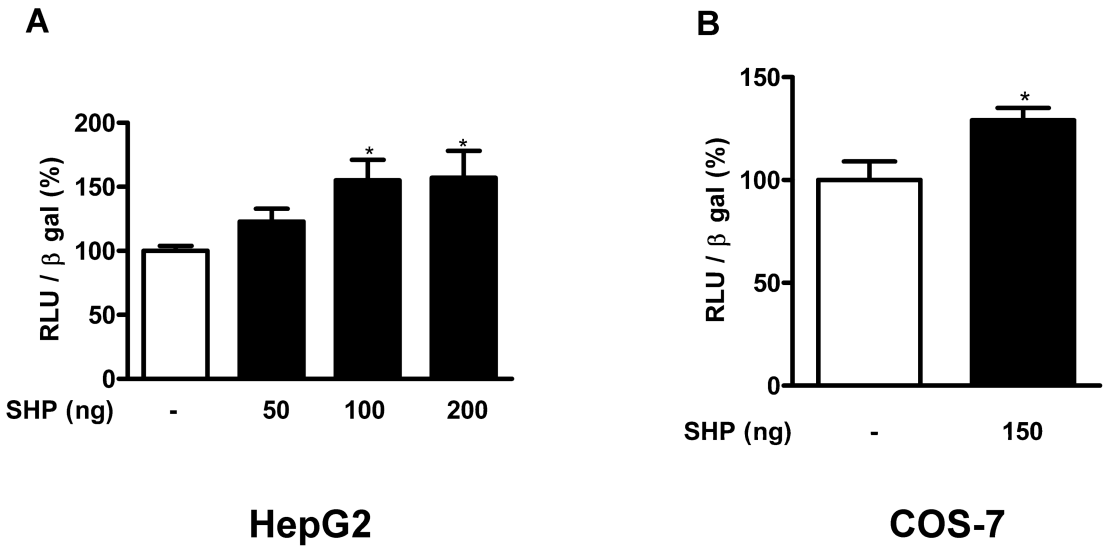
HNF4α mediated transactivation of the human *APOA* promoter is inhibited by cotransfection with FXR in COS-7 cells. COS-7 cells were co-transfected with the h*APOA* -1952/+52 promoter reporter plasmid (150 ng), expression vectors for HNF4α, FXR or HNF4α and FXR in the absence or presence of CDCA. Luciferase activity was measured, the values were normalized to internal control β-galactosidase and expressed in percentage. Values shown are mean ± SD of 3 replicates (** $p \leq 0.01$, * $p<0.05$).



Supplemental Figure 6

Effect of SHP on human *APOA* promoter activity.

(**A**) HepG2 cells were transfected with the h*APOA* -1952/+52 promoter reporter plasmid (150 ng) in the presence of either empty or increasing amount of SHP expression vector (50, 100, 200 ng). (**B**) COS-7 cells were transfected with the h*APOA* -1952/+52 promoter reporter plasmid (150 ng) in the absence or presence of SHP expression vector (150 ng). Values are expressed as percentage of control, normalized to internal control β-galactosidase activity. Data are presented as mean ± SD (* $p<0.05$).



Supplemental Figure 7

Proposed model for HNF4α and FXR counter regulation of *APOA* promoter activity.

(**A**) In basal conditions, HNF4α is bound to the DR-1 site and promotes *APOA* transcription by interacting with co-factors enhancing the polymerase II activity.

(**B**) During cholestasis or FXR activation, FXR displaced HNF4α from the DR-1, thereby lowering the polymerase II activity and in turn *APOA* gene expression.