Supplementary Information:

3D Culture System for Liver Tissue Mimicking Hepatic Plates for Improvement of Human Hepatocyte (C3A) Function and Polarity

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Table S1. ELISA kit information

Name of reagent kit	Catalog number	Brand, country
Alpha 1 Antitrypsin Human ELISA Kit	ab108798	Abcam, UK
Apolipoprotein AI Human ELISA Kit	ab108804	Abcam, UK
Glucokinase Human ELISA Kit	ab125967	Abcam, UK
Testosterone free Human ELISA Kit	ab178663	Abcam, UK
Human Coagulation Factor II ELISA Kit	abx053005	Abbexa, UK
Human sTfR Quantikine IVD ELISA Kit	DTFR1	R&D Systems, USA
Amino Acid Assays	ab83362	Abcam, UK
Human ALB / Serum albumin ELISA Kit	RAB0603-1KT	Sigma, Japan
ELISA Kit for Coagulation Factor VII	EKU03266	Biomatic, Canada
ELISA Kit for Cytochrome P450 1A2	EKU03593	Biomatic, Canada
ELISA Kit for Glutathione S Transferase Alpha 1	EKU04486	Biomatic, Canada
Cytochrome P450 2D6 Activity Assay Kit	K703-200	Biovision, USA
Cytochrome P450 3A4 Human ELISA Kit	K7570-100	Biovision, USA
Nuclear receptor subfamily 1, group I, member 2 (NR1I2), ELISA Kit	MBS9326391	MyBioSource, USA

Human Coagulation Factor V (F5) ELISA Kit	EKU03262	Biomatik, Canada
REAGEN™ Diazepam (DIA) ELISA Test Kit	RNH94016	REAGEN, USA
Blood Ammonia assay kit	A086	Nanjing Jiancheng Bioengineering Institute, China

Table S2. Hepatocyte functional genes and primer sequences

Gene	Forward primer (5′–3′)	Reverse primer (5′–3′)
UGT1A1	TGTTGTGCTTATGGCTACCG	TTCCCACCCACTTCTCAAT
GCK	TACGAAGACCATCAGTGCG	CTCCATGTAGCAGGCATTG
ALB	GTGGGCAGCAAATGTTGTAA	GAGTTCAGGTGTGAAATAGCG
TF	GAAGCCTGCACTTTCCGTA	TCATGCAACACACATAACTGG
A1	GTACGTGGATGTGCTCAAAGA	GTGAAACCAGGGCAAACT
AAT	GCCAAGAAACAGATCAACGA	AGTTGTCAAGGAGCTTTAGG
CPS1	ATGAGTGTGACAAACTGTACTT	CGAAGAGACCCAATAGGTAG
GLUL	TCCACACAGTTCCTATTGTCA	CTCCAACTGATATGATGCAGC
CYP3A4	CCGTTGTTCTAAAGGTTGAGT	ACAATGCAGGTAACTCAATCT
CYP2D6	ATCCCGTGACATCGAAGT	AGGCACAGATTTCTTGAAGAG
CYP1A2	ACATCTTTGGAGCAGGATTT	ACAGGTTGGTGATGAGTG
GSTA1	GCTGACATTCATCTGGTGG	CAGGCTTGGTCACAAGGTA
F2	TCAGACAGGGCCATCGAA	AATGGGTCACCTTCAGCA
F5	CAAATTACAGGCCAGTCTCG	CCAAAGGTCCTCGGATTGA
F7	CCTTCATTGCTGGAGACAGTA	GGTATGGGCAACAGGTAAC

PXR	GTGATTTGGTGTAGGTAGGTC	ATCGGTGTGTTTGCGTAT
β-actin	CCATCATGAAGTGTGACG	AACTGATTGTCAGCGTAGC

UGT1A1: UDP-glucuronosyltransferase; *GCK*: glucokinase; *ALB*: albumin; *TF*: transferrin; *APOA1*: apolipoprotein A1; *AAT*: α1-antitrypsin; *CPS1*: carbamoyl phosphate synthetase; *GLUL*: glutamine synthetase; *GSTA1*: glutathione-S-transferase; *F2*: Coagulation Factor II; *F5*: Coagulation Factor V; *F7*: Coagulation Factor VII; *PXR/NR1I2*: pregnane X receptor

Table S3. Hepatocyte polarity genes and primer sequences

Gene	**Forward primer** (5′–3′)	**Reverse primer** (5′–3′)
ZO1	TTGACGGGACTGTTGGTATTG	GTGAAGTTAAGCAGTGACGA
Occludin	TGCCACTTTGGCATTATGAG	CACCATTCTGCATTTCGTTG
MRP2	GAGAGAACAGCTTTCGTCG	CTTCAGGCTATTCACATTCCG
DPPIV	GTGGGCAGTGATGTCACTA	GTATGCTTGGACTTGGGTTC
NTCP	CTGAAAGCCAGTGTGGTAA	CGGCCAAGACTTGATGATT
CD147	TGCCTGTCTGAAGCCAAT	TGAGGCTGAGTCGAACAC
β-actin	CCATCATGAAGTGTGACG	AACTGATTGTCAGCGTAGC

ZO-1, zona occludens protein 1; *MRP2*, multidrug-resistance associated protein 2; *DPPIV*, dipeptidyl peptidase IV; *NTCP*, (SLC10A1) Na+-dependent taurocholate cotransporting polypeptide; *CD147*, cluster of differentiation 147

Table S4. Immunofluorescence antibody information

Name of antibody	Brand	Dilution
Anti-CD147 antibody [EPR4052]	Abcam, UK	1:100
Anti-MRP2 antibody [EPR10998]	Abcam, UK	1:250
Anti-Occludin (C-terminal) produced in rabbit, affinity isolated antibody	Sigma, Japan	1:1000
Anti-ZO-1 tight junction protein antibody	Abcam, UK	1:1000
Donkey Anti-Rabbit IgG H&L (Alexa Fluor® 647)	Abcam, UK	1:500
Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488)	Abcam, UK	1:500
DAPI Solution	Dojindo Laboratories, Japan	1:1000

Table S5. Primers used to validate differentially expressed genes associated with bile secretion

Gene	**Forward primer** (5′–3′)	**Reverse primer** (5′–3′)
AQP8	GCGTGTTTCTTGAGAGGAAT	CGGGAAATGAGCTGATGTG
MDR1	TGGTGTCACAGGAAGAGATT	TGACTCGATGAAGGCATGTA
MDR3	ATCAGTGAAGGAATTGGTGAC	CCTCTGATGAATCCCACTATG
FXR	TCAGGAGCCACTTCTTGAT	AGACAGGCAAAGTGTTGAG
RXRA	CTCAAATGCCTGGAACATCTC	CTCCATAAGGAAGGTGTCAAT
SHP	ACCCTCAAGTCCATTCCGA	CCAGCGATGTCAACATCTC
OSTA	ACCGAAGGAAAGACCACAA	GTCCAAGCCATCCACCTTA
MRP2	GAGAGAACAGCTTTCGTCG	CTTCAGATGCCTGCCATTG
NTCP	AAGCCAGTGTGGTAAACTAGA	ACTGGAAATGCTGGAGAAAG
ACTB	CCATCATGAAGTGTGACG	GCCGATCCACACGGAGTA

FXR: farnesoid X receptor; *SHP*: small heterodimer partner; *RXRA*: retinoid X receptor α; *MRP2*: multidrug resistance-associated protein; *MDR1*: multidrug resistance 1; *MDR3*: multidrug resistance 3; *NTCP*: Na+-dependent taurocholic cotransporting polypeptide; *OSTA*: organic solute transporter-alpha; *AQP8*: aquaporin-8

Table S6. Viability trends of the four groups (mean±standard deviation, n=3)

Time (day)	Viability(OD/10^4cells)			
	P	S	M	H
1	0.004318658±0.0001780362	0.0035±0.0002795782$^{▲}$	0.003330909±0.0006248096$^{■}$	0.004550349±0.0007435413
3	0.003850299±0.0001351292***	0.003930041±0.0003974338$^{▲▲▲}$	0.003820144±0.00002855132$^{■■■}$	0.005552529±0.0008242053
5	0.004329147±0.0004472979***	0.004982825±0.0006998997$^{▲▲}$	0.005367048±0.001574741	0.006275122±0.001294016
7	0.004069356±0.0001734511***	0.005279788±0.00005051773$^{▲▲}$	0.005716854±0.0005443209	0.006551211±0.001029228
9	0.003031145±0.0003739792***	0.005363278±0.0001955372	0.004611268±0.0003973028$^{■}$	0.005858899±0.000649709
11	0.002896848±0.0004468536***	0.004613072±0.0004741096	0.003632±0.001180509$^{■■■}$	0.005528369±0.0006200706
13	0.002598022±0.0002967114***	0.003247458±0.0002630342$^{▲▲▲}$	0.003666879±0.00127014$^{■■■}$	0.005850794±0.0004796381

H, hepatic plate-mimetic group; M, mixed 3D group; S, sandwich group; P, mixed 2D/plate group. H Vs P: $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$; H Vs S: $^{▲}P < 0.05$, $^{▲▲}P < 0.01$, $^{▲▲▲}P < 0.001$; H Vs M: $^{■}P < 0.05$, $^{■■}P < 0.01$, $^{■■■}P < 0.001$).

Table S7. Dynamic testing of hepatocyte function (mean±standard deviation, n=3)

Time (day)	CYP1A2(ng/mg)			
	P	S	M	H
1	3.7015±0.3514321***	2.0925±0.1378859▲▲▲	40.6965±0.8251935	39.325±0.07495261
3	3.2375±0.07000355***	16.6055±0.2877922▲▲	32.3525±1.509672▪▪▪	23.6285±0.6611458
5	10.058±0.6420529***	1.949±0.1626345▲▲▲	35.7295±0.8393359	34.1605±1.810901
7	1.326±0.09758075***	2.6735±0.1421284▲▲▲	29.582±0.4285074▪▪▪	59.8605±6.715394
9	6.6535±0.04737609***	3.8165±0.2793072▲▲▲	28.813±0.2503159▪▪▪	74.3775±1.403607
11	15.2955±0.1463709***	4.674±0.4440629▲▲▲	27.802±0.4525479▪▪▪	55.6115±3.380676
13	5.2775±0.8167085***	9.863501±0.751655▲▲▲	26.572±0▪▪▪	39.1735±1.344211

H, hepatic plate-mimetic group; M, mixed 3D group; S, sandwich group; P, mixed 2D/plate group. H Vs P: $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$; H Vs S: ▲$P < 0.05$, ▲▲$P < 0.01$, ▲▲▲$P < 0.001$; H Vs M: ▪$P < 0.05$, ▪▪$P < 0.01$, ▪▪▪$P < 0.001$).

Time (day)	CYP2D6（pU/mg）			
	P	S	M	H
1	6.1935±0.1958685	69.3125±2.170111▲▲▲	10.0525±2.492551	13.4945±3.693219
3	8.7155±3.120462**	16.7895±0.6526598	18.5785±0.1647557	31.578±1.885146
5	9.967±2.231629***	13.7675±1.221173▲▲▲	81.586±8.67903■■■	146.44±5.176027
7	2.6845±0.2029398***	19.36±0.8994394▲▲▲	51.7155±1.262185■■■	124.16±13.56089
9	3.6745±0.1605132***	15.9155±3.379264▲▲▲	48.06±1.65463■■■	93.98351±24.26295
11	3.7815±0.3981012***	6.787±0.7537758▲▲▲	47.377±4.310524	44.7325±3.127533
13	3.238±0.2022325***	5.631±0.5218447▲▲▲	36.5825±2.396385■■■	89.8705±3.59281

H, hepatic plate-mimetic group; M, mixed 3D group; S, sandwich group; P, mixed 2D/plate group. H Vs P: $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$; H Vs S: $^{▲}P < 0.05$, $^{▲▲}P < 0.01$, $^{▲▲▲}P < 0.001$; H Vs M: $^{■}P < 0.05$, $^{■■}P < 0.01$, $^{■■■}P < 0.001$).

Time (day)	CYP3A4（ng/mg）			
	P	S	M	H
1	0.8285±0.0007070977	1.0495±0.06576089	1.213±0.02828433	1.123±0.08768121
3	0.59±0.04666908	0.7065±0.07566042	2.942±0.3337543▪▪▪	1.2455±0.3528463
5	0.3195±0.009192396***	0.4545±0.1081874▲▲▲	1.867±0.2489016▪	2.89±0.7509474
7	0.4485±0.006363963	0.8435±0.08838835	1.2475±0.1251578	0.623±0.05091166
9	0.173±0.02404163	0.4475±0.07566042	1.3545±0.3966869	0.6255±0.01767769
11	0.7555±0.002121335	4.8975±0.3132482▲▲▲	7.7805±0.7912524▪▪▪	1.3175±0.4080006
13	0.7505±0.1180868*	0.7065±0.02899138	2.1825±0.2326381▪	1.7685±0.171827

H, hepatic plate-mimetic group; M, mixed 3D group; S, sandwich group; P, mixed 2D/plate group. H Vs P: $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$; H Vs S: ▲$P < 0.05$, ▲▲$P < 0.01$, ▲▲▲$P < 0.001$; H Vs M: ▪$P < 0.05$, ▪▪$P < 0.01$, ▪▪▪$P < 0.001$).

Time (day)	GST(pg/mg)			
	P	S	M	H
1	591.605±86.3448*	90.35001±18.29709▲▲▲	4158.862±530.8326■■■	1806.132±264.2621
3	649.244±77.46919	144.728±4.710747	1547.7±205.2767	900.0795±87.89552
5	659.7305±66.39238***	345.688±28.83158▲▲▲	1403.817±84.50064■■■	5637.073±607.0764
7	216.0165±6.111517***	374.615±59.54546▲▲▲	833.783±21.56533■■■	7208±166.8772
9	266.615±11.67151***	874.714±43.76▲▲▲	3004.685±214.0433■■■	11074.01±1251.596
11	524.7104±9.00219***	581.3365±30.03296▲▲▲	5631.476±602.7795■■■	8017±687.3078
13	180.9035±6.549931***	540.233±6.59024▲▲▲	3268.76±126.2279	3916.537±73.46556

H, hepatic plate-mimetic group; M, mixed 3D group; S, sandwich group; P, mixed 2D/plate group. H Vs P: $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$; H Vs S: $^{▲}P < 0.05$, $^{▲▲}P < 0.01$, $^{▲▲▲}P < 0.001$; H Vs M: $^{■}P < 0.05$, $^{■■}P < 0.01$, $^{■■■}P < 0.001$).

Time (day)	△DIA（ppb/10^4cells）			
	P	S	M	H
1	0.0007421385±0.0004447213	0.00157783±0.0001701059▲▲▲	0.0004145455±0.00003085602	0.000442672±0.0002030372
3	0.0006691615±0.00006986425	0.001549382±0.0001833238▲	0.000582734±0.0000763067	0.00103891±0.00004952502
5	0.0005353605±0.0001261853**	0.0009589905±0.00002379342	0.0006019575±0.0001038167■	0.00115317±0.0003559682
7	0.000368517±0.00008793298**	0.000484554±0.0002958238▲	0.000710112±0.0001144085	0.001016759±0.0002054166
9	0.0003093435±0.00003749858***	0.00022695±0.0001002989▲▲▲	0.000701408±0.000119511■■■	0.001436505±0.0003784966
11	0.000381089±0.0001215658***	0.00012549±0.00001663823▲▲▲	0.000648±0.00001131371■■	0.00137234±0.0001654934
13	0.0002146595±0.00004195759**	0.0001101695±0.00001198475▲▲▲	0.0006484425±0.00009709778	0.0009380955±0.00002020275

H, hepatic plate-mimetic group; M, mixed 3D group; S, sandwich group; P, mixed 2D/plate group. H Vs P: $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$; H Vs S: ▲$P < 0.05$, ▲▲$P < 0.01$, ▲▲▲$P < 0.001$; H Vs M: ■$P < 0.05$, ■■$P < 0.01$, ■■■$P < 0.001$).

Time (day)	△NH3(mmol/L/10^4cells)			
	P	S	M	H
1	0.0133522±0.001325269***	0.00825±0.001841147	0.003578182±0.0002622723	0.005842936±0.0007869669
3	0.006649492±0.0004923457	0.007751939±0.001421116	0.012088±0.0001183056	0.009818116±0.0001279717
5	0.000974884±0.0001972672***	0.005696166±0.001913607▲▲▲	0.01269984±0.000339134▪	0.01592195±0.0006622653
7	0.002527551±0.0002327576***	0.003222561±0.0001161119▲▲▲	0.01537978±0.0002669522	0.01770279±0.001144012
9	0.002239449±0.00003875798***	0.00375685±0.0005131565▲▲▲	0.01235239±0.0004625083▪▪▪	0.01809012±0.001019625
11	0.002021279±0.0006596585***	0.005213559±0.0002794458▲▲▲	0.01314729±0.001490956▪▪▪	0.02287234±0.002256724
13	0.001259522±0.0003883876***	0.001697542±0.00002229716▲▲▲	0.009028608±0.0006958689	0.01095238±0.002020305

H, hepatic plate-mimetic group; M, mixed 3D group; S, sandwich group; P, mixed 2D/plate group. H Vs P: $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$; H Vs S: ▲$P < 0.05$, ▲▲$P < 0.01$, ▲▲▲$P < 0.001$; H Vs M: ▪$P < 0.05$, ▪▪$P < 0.01$, ▪▪▪$P < 0.001$).

Time (day)	Urea(nmol/10^4cells)			
	P	S	M	H
1	0.1425472±0.008173971	0.1755283±0.006123811▲▲	0.1369855±0.002360455	0.1300857±0.01554929
3	0.1004102±0*	0.1720494±0▲	0.1008075±0.02337268▪	0.135858±0.01263713
5	0.06908129±0***	0.1030578±0.01639358	0.1184144±0.012721	0.13536±0.02662309
7	0.03460418±0.007553625***	0.04362577±0.0009136857▲▲▲	0.08534382±0.005841179	0.1040056±0.004321648
9	0.01972222±0.001092801***	0.05575177±0.008194414▲▲▲	0.1035296±0.005485555	0.1143086±0.004402923
11	0.0184298±0.004343946***	0.03663334±0.001699827▲▲▲	0.07718±0.003467653▪▪▪	0.1319628±0.02304115
13	0.01583944±0.002853108***	0.04360508±0.0007358708▲▲▲	0.1016376±0.02313089	0.1170952±0.0005252762

H, hepatic plate-mimetic group; M, mixed 3D group; S, sandwich group; P, mixed 2D/plate group. H Vs P: $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$; H Vs S: ▲$P < 0.05$, ▲▲$P < 0.01$, ▲▲▲$P < 0.001$; H Vs M: ▪$P < 0.05$, ▪▪$P < 0.01$, ▪▪▪$P < 0.001$).

Time (day)	△TT（pg/ml/10^4cells）			
	P	S	M	H
1	0.009759748±0.0002490437***	0.009642452±0.0003502186$^{▲▲▲}$	0.005574545±0.0009102395$^{■■■}$	0.008008774±0.0003130161
3	0.006157185±0.0001841863***	0.009560494±0.000218243$^{▲▲▲}$	0.007202158±0.00009919886$^{■■■}$	0.008385993±0.0002393704
5	0.003169531±0.0001507219***	0.005605889±0.0002795709$^{▲▲▲}$	0.006341599±0.0002214758$^{■■}$	0.007100488±0.0002897411
7	0.002501972±0.0000514725***	0.003899735±0.0001310615$^{▲▲▲}$	0.005855281±0.00022246$^{■■■}$	0.004793296±0.0002686213
9	0.00192399±0.00004642574***	0.003479433±0.0001136724$^{▲▲▲}$	0.005411831±0.000473264$^{■■■}$	0.006609012±0.0003862209
11	0.001855015±0.00007496533***	0.002862353±0.00009428135$^{▲▲▲}$	0.0055312±0.0001187939$^{■■■}$	0.007722341±0.0001955825
13	0.002402792±0.0000962555***	0.002398136±0.00003140056$^{▲▲▲}$	0.005205849±0.0001780128$^{■■■}$	0.007651429±0.0001683585

H, hepatic plate-mimetic group; M, mixed 3D group; S, sandwich group; P, mixed 2D/plate group. H Vs P: $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$; H Vs S: $^{▲}P < 0.05$, $^{▲▲}P < 0.01$, $^{▲▲▲}P < 0.001$; H Vs M: $^{■}P < 0.05$, $^{■■}P < 0.01$, $^{■■■}P < 0.001$).

Time (day)	PXR（pg/ml/10^4cells）			
	P	S	M	H
1	0.2128491±0.03967802***	0.1054316±0.01113693	0.07521273±0.03433454	0.09579661±0.01883174
3	0.07191917±0***	0.09198149±0.009716171▲▲▲	0.1224604±0.03400217■■■	0.2170564±0.02762669
5	0.03141524±0.003901239***	0.0533123±0.006620483▲▲▲	0.269168±0.03460554■■	0.2046263±0.009230329
7	0.02597361±0.002387049***	0.04240247±0.008335498▲▲▲	0.2223371±0	0.1871508±0.01185096
9	0.01600505±0.00596041***	0.09583215±0.01492781	0.08178169±0.01996431	0.1300501±0.02581502
11	0.02453868±0.006767153**	0.03576667±0.006175401▲▲	0.059604±0.006296081	0.1029468±0.008379968
13	0.01780105±0.0004936173	0.01959661±0	0.03137699±0.005998495	0.06190477±0.006734353

H, hepatic plate-mimetic group; M, mixed 3D group; S, sandwich group; P, mixed 2D/plate group. H Vs P: $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$; H Vs S: ▲$P < 0.05$, ▲▲$P < 0.01$, ▲▲▲$P < 0.001$; H Vs M: ■$P < 0.05$, ■■$P < 0.01$, ■■■$P < 0.001$).

Time (day)	ALB（pg/ml/10^4cells）			
	P	S	M	H
1	7.830189±0.2045717***	3.870283±0.2829161	6.381818±0.2314169■	4.415779±0.373327
3	14.43413±0.3429681***	8.350877±0.5149661▲	10.95814±1.358897■■■	6.63656±0.8095354
5	14.82155±0.07010268***	7.53931±0.4573725▲▲▲	10.19464±0.4828653	11.41556±1.261252
7	8.423335±0.4453998***	9.696382±0.2920797▲▲▲	10.4±0.2669528■■	12.95562±0.5138732
9	6.807367±0.1772356***	8.085107±0.1337319▲▲▲	10.99126±0.3777922■■	13.06845±0.02193727
11	6.84962±0.1131592***	7.294118±0.1663779▲▲▲	12.28059±0.05012576	13.62264±0.4313578
13	6.461136±0.1287229***	7.21017±0.2061396▲▲▲	12.36738±0.01609672■■	14.6085±0.954533

H, hepatic plate-mimetic group; M, mixed 3D group; S, sandwich group; P, mixed 2D/plate group. H Vs P: $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$; H Vs S: ▲$P < 0.05$, ▲▲$P < 0.01$, ▲▲▲$P < 0.001$; H Vs M: ■$P < 0.05$, ■■$P < 0.01$, ■■■$P < 0.001$).

Time (day)	AAT（μg/ml/10^4cells）			
	P	S	M	H
1	0.009471698±0.001956773	0.01412264±0.002701682▲▲▲	0.006987273±0.0004088366	0.007932204±0.0004906753
3	0.00847006±0.0003810753	0.01458642±0.002173699▲▲▲	0.01040288±0.001144597	0.01042996±0.000586046
5	0.006986946±0.0002278347***	0.01285804±0.00008922488	0.01341436±0.000159186	0.01335805±0.0008112754
7	0.006085835±0.0002981125***	0.008316858±0.0004456087▲▲▲	0.01337977±0.001061455▪▪▪	0.01851955±0.0004345353
9	0.004233586±0.00003035594***	0.008879432±0.000869257▲▲▲	0.01238451±0.0007827973▪▪▪	0.01649522±0.0001544879
11	0.003965616±0.00008509614***	0.005033334±0.000646101▲▲▲	0.010868±0.0001527349▪▪▪	0.01519681±0.0003535534
13	0.005345549±0.0001752344***	0.00359661±0.0003451642▲▲▲	0.009291799±0.000172619▪▪	0.01228571±0.0001346868

H, hepatic plate-mimetic group; M, mixed 3D group; S, sandwich group; P, mixed 2D/plate group. H Vs P: $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$; H Vs S: $^{▲}P < 0.05$, $^{▲▲}P < 0.01$, $^{▲▲▲}P < 0.001$; H Vs M: $^{▪}P < 0.05$, $^{▪▪}P < 0.01$, $^{▪▪▪}P < 0.001$).

Time (day)	APOA1(ug/ml/10^4cells)			
	P	S	M	H
1	0.04±0.006759762***	0.02969575±0.001390866$^{▲▲▲}$	0.01735091±0.001056803	0.01756929±0.0002791768
3	0.04935629±0.005017495***	0.02604321±0.002034023$^{▲▲▲}$	0.01565827±0.002685988	0.01421206±0.0009492287
5	0.03104098±0.0008061849***	0.02154364±0.0004758656	0.01634095±0.0005052406	0.01641366±0.001374201
7	0.01488778±0.0002037464*	0.01158958±0.000542968$^{▲▲}$	0.01342921±0.001576291$^{▪}$	0.02148045±0.003705398
9	0.01015657±0.000778532**	0.007413711±0.00002005992$^{▲▲▲}$	0.01837183±0.001613398	0.01827583±0.001112317
11	0.009723496±0.001237943***	0.005988235±0.00068215$^{▲▲▲}$	0.014532±0.002720947$^{▪}$	0.02246809±0.0005566584
13	0.007921467±0.0002542133**	0.006376271±0.001107402$^{▲▲}$	0.01368214±0.002195493	0.01628571±0.001912556

H, hepatic plate-mimetic group; M, mixed 3D group; S, sandwich group; P, mixed 2D/plate group. H Vs P: $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$; H Vs S: $^{▲}P < 0.05$, $^{▲▲}P < 0.01$, $^{▲▲▲}P < 0.001$; H Vs M: $^{▪}P < 0.05$, $^{▪▪}P < 0.01$, $^{▪▪▪}P < 0.001$).

Time (day)	TF（nM/10^4cells）			
	P	S	M	H
1	0.6903774±0.03475943***	0.2334056±0.009205725	0.4169291±0.01773424■■■	0.2502832±0.01846796
3	1.189347±0.01825355***	0.4179074±0.02408528▲	0.5091798±0.05437599■■■	0.3421459±0.03510495
5	0.9372794±0.01893835	0.3553438±0.01504331▲▲▲	0.4629592±0.009551129	0.5079922±0.03615972
7	0.3392144±0.0130655***	0.1494334±0▲▲▲	0.5605079±0.01899816	0.5798491±0.01816356
9	0.2761818±0.005746133***	0.09402128±0.0003844794▲▲▲	0.5007253±0.01098901■■	0.5957943±0.003553232
11	0.2822521±0.003959001***	0.04034118±0.0006377819▲▲▲	0.541608±0.004548109■	0.6120053±0.01210356
13	0.246534±0.002838295***	0.03078135±0.001378259▲▲▲	0.5436542±0.01054055■■■	0.6431286±0.03715337

H, hepatic plate-mimetic group; M, mixed 3D group; S, sandwich group; P, mixed 2D/plate group. H Vs P: $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$; H Vs S: ▲$P < 0.05$, ▲▲$P < 0.01$, ▲▲▲$P < 0.001$; H Vs M: ■$P < 0.05$, ■■$P < 0.01$, ■■■$P < 0.001$).

Time (day)	GCK(ng/ml/10^4cells)			
	P	S	M	H
1	0.008446541±0.001378636	0.01023113±0.0005203239	0.01128±0.002005612	0.009732801±0.001091327
3	0.01400748±0.006675173	0.01260494±0.003858533	0.02321763±0.001823726	0.01374514±0.004085811
5	0.01667796±0.0006694855	0.01877182±0.0001606045	0.03097879±0.00903897	0.02099707±0.002624229
7	0.01155293±0.002899631**	0.01005649±0.0003969285▲▲	0.0363236±0.005644143	0.02657542±0.002109469
9	0.01307197±0.0005589001***	0.01812293±0.002306873▲▲▲	0.03837465±0.001673156	0.04780883±0.009060751
11	0.01652292±0.002336086***	0.02021177±0.004176089▲▲▲	0.050512±0.0003280974▪▪	0.06393085±0.002835951
13	0.02332286±0.001525277***	0.02196271±0.00288116▲▲▲	0.05791354±0.005194741	0.06287619±0.003878988

H, hepatic plate-mimetic group; M, mixed 3D group; S, sandwich group; P, mixed 2D/plate group. H Vs P: $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$; H Vs S: ▲$P < 0.05$, ▲▲$P < 0.01$, ▲▲▲$P < 0.001$; H Vs M: ▪$P < 0.05$, ▪▪$P < 0.01$, ▪▪▪$P < 0.001$).

Time (day)	F2（ng/ml/10^4cellsl）			
	P	S	M	H
1	0.02425786±0.00146758	0.0280684±0.0005503417▲	0.01594364±0.001689342	0.0210987±0.0004652946
3	0.02028593±0.0003493197	0.02584568±0.0004801343	0.01725±0.0004196865	0.02186965±0.001361937
5	0.01228354±0.0005748461***	0.02431125±0.0006543153	0.01672268±0.0003806611▪	0.02354342±0
7	0.008765545±0.000235917***	0.02184996±0.003085556	0.01782921±0.001747905	0.0230838±0.0008690694
9	0.007506314±0.00009820907***	0.03275887±0.0001838814	0.01908591±0.0009859663▪▪▪	0.03787346±0.009763673
11	0.008911175±0.0002228699***	0.01402745±0.0004547671▲▲▲	0.02224±0.002489016▪▪▪	0.02957447±0.0008274651
13	0.01305934±0.000407234***	0.01100847±0.0001318334▲▲▲	0.02602924±0.001478047	0.02490476±0.0007407783

H, hepatic plate-mimetic group; M, mixed 3D group; S, sandwich group; P, mixed 2D/plate group. H Vs P: $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$; H Vs S: ▲$P < 0.05$, ▲▲$P < 0.01$, ▲▲▲$P < 0.001$; H Vs M: ▪$P < 0.05$, ▪▪$P < 0.01$, ▪▪▪$P < 0.001$).

Time (day)	F5(pg/ml/10^4cells)			
	P	S	M	H
1	0.3143899±0.2222717	0.3536887±0.2500556	0.4770873±0.2890858	0.07476969±0.1057403
3	1.513801±0.07919385	2.157648±0.2176753	2.894277±0.475008▪	2.040111±0.2058176
5	2.438506±0.2174668***	1.679912±0.1482619▲▲▲	4.815±0.2576383▪▪▪	3.722827±0.1029905
7	1.849239±0.2125267***	1.848622±0.1862531▲▲▲	6.425789±1.021895▪▪▪	4.733553±0.1962678
9	1.852159±0.1324647***	2.003088±0.08305407▲▲▲	7.659164±0.294768▪▪▪	5.644938±0.09581395
11	1.213878±0.1509988***	1.977812±0.03440135	5.125372±0.07003728▪▪▪	2.588356±0.0936916
13	0.9578255±0.2457968***	1.667575±0.02974396▲▲	4.084013±0.2675696▪▪▪	2.791481±0.083809

H, hepatic plate-mimetic group; M, mixed 3D group; S, sandwich group; P, mixed 2D/plate group. H Vs P: $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$; H Vs S: ▲$P < 0.05$, ▲▲$P < 0.01$, ▲▲▲$P < 0.001$; H Vs M: ▪$P < 0.05$, ▪▪$P < 0.01$, ▪▪▪$P < 0.001$).

Time (day)	F7(pg/ml/10^4cells)			
	P	S	M	H
1	0.5670252±0.08715644***	1.333054±0.1177633▲▲▲	0.9658527±0.0235968	1.27008±0.1027117
3	0.6475195±0.02795187	1.211389±0.1025655▲▲▲	0.4936996±0.04417382■■■	0.7849553±0.02184876
5	0.5455965±0.006070917	0.6968959±0.05992936	0.5217798±0.01535797	0.6577229±0.1174115
7	0.2771429±0.009359467***	0.4248111±0.01477997▲	0.6024629±0.01962741	0.6005642±0.04557878
9	0.2197121±0.00624611***	0.243565±0.06088139▲▲▲	0.4692338±0.02384245■■	0.6862667±0.09558206
11	0.1962751±0.006078281***	0.2411529±0.03678619▲▲▲	0.643424±0.07937697	0.7411649±0.009937089
13	0.1535777±0.009872341***	0.2107254±0.01801084▲▲▲	0.905569±0.02792106	0.9380952±0.04714045

H, hepatic plate-mimetic group; M, mixed 3D group; S, sandwich group; P, mixed 2D/plate group. H Vs P: $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$; H Vs S: ▲$P < 0.05$, ▲▲$P < 0.01$, ▲▲▲$P < 0.001$; H Vs M: ■$P < 0.05$, ■■$P < 0.01$, ■■■$P < 0.001$).

Table S8. Quantification of hepatocyte functional gene expression levels (mean±standard deviation, n=3)

Time (day)	*CYP3A4* Relative expression			
	P	S	M	H
1	1±0	0.7056401±0.1090831	0.384651±0.05200034	0.3896478±0.2103232
7	1±0**	2.240479±0.7401251*	3.165113±1.366681	5.159448±2.042862
13	1±0***	2.165119±0.120206***	14.06088±1.032818***	8.110536±0.5164277
Time (day)	*CYP2D6* Relative expression			
	P	S	M	H
1	1±0	0.506513±0.2540927	2.099574±0.03430062	1.730353±0.3813379
7	1±0***	0.8426434±0.1870459***	4.000867±1.370987***	8.349505±0.06820507
13	1±0**	0.609924±0.005978741***	1.587294±0.1501846*	3.160521±0.5700326
Time (day)	*CYP1A2* Relative expression			
	P	S	M	H
1	1±0**	0.5979288±0.06823251***	1.230192±0.096374	1.531559±0.002502006
7	1±0***	1.127623±0.07547683***	2.392805±0.1796575***	4.033256±0.1120052
13	1±0***	0.4945228±0.07085421***	1.576838±0.04644987*	2.063296±0.3688198
Time (day)	*GSTA1* Relative expression			
	P	S	M	H
1	1±0	0.9825979±0.2462008	1.43493±0.4673636	0.8245517±0.07935742
7	1±0	3.931367±0.2695513	1.704588±1.81742	1.259788±0.145805
13	1±0***	10.8378±5.59094***	47.38522±15.50688	52.10273±4.293867

Time (day)	*UGT1A1* Relative expression			
	P	S	M	H
1	1±0	1.042135±0.2196891	0.818848±0.001337783	1.027272±0.1055478
7	1±0	3.374777±0.313821***	0.8115302±0.5147988	0.942058±0.03692894
13	1±0	0.4141446±0.02704529	0.765632±0.04750228	0.5472754±0.2009406
Time (day)	*PXR* Relative expression			
	P	S	M	H
1	1±0	0.5288886±0.04573904	0.6965783±0.005690162	0.902119±0.0559703
7	1±0***	0.547569±0.03040069***	2.268824±0.06300616	2.039688±0.1963058
13	1±0***	1.192986±0.1070541***	4.96552±0.1378944	4.545526±0.5039532
Time (day)	*CPS1* Relative expression			
	P	S	M	H
1	1±0	0.3033684±0.01436794**	0.8501619±0.04026486	0.8600403±0.04073274
7	1±0**	0.1493876±0.005368223***	1.075775±0.1957557**	1.55248±0.06068455
13	1±0***	0.4146159±0.04798674***	1.587399±0.09733891*	1.968869±0.2470322
Time (day)	*GLUL* Relative expression			
	P	S	M	H
1	1±0***	0.9625646±0.1301275***	2.158501±0.4135661	2.654592±0.4060807
7	1±0	0.6850677±0.1640518	0.4513991±0.08358987	0.6314178±0
13	1±0	0.7130198±0.02212967	0.6863669±0.0224233	0.7292306±0.08084828

Time (day)	*ALB* Relative expression			
	P	S	M	H
1	1±0*	0.3121235±0.007138475	0.4784094±0.02890191	0.5428215±0.01330133
7	1±0	1.629671±0.09579464**	0.7721775±0.1193689	0.9822235±0.1692553
13	1±0	0.4493926±0.02421691*	0.7994345±0.02611714	0.9660175±0.4956045
Time (day)	*AAT* Relative expression			
	P	S	M	H
1	1±0***	0.5183659±0.09432557***	1.718029±0.07856451	1.656394±0.1890189
7	1±0***	0.4611424±0.03086622	0.2698752±0.0272899	0.4435935±0.03547374
13	1±0**	0.5554886±0.01179707	0.4514386±0.03830622	0.5974686±0.2120699
Time (day)	*TF* Relative expression			
	P	S	M	H
1	1±0***	0.4744391±0.09925693***	1.521426±0.05218834***	2.254255±0.2462642
7	1±0	0.426224±0.03894124**	0.5543753±0.02263669*	0.8673656±0.09897926
13	1±0	0.6115146±0.0469102**	0.7873337±0.009004042*	1.06672±0.1234597
Time (day)	*APOA1* Relative expression			
	P	S	M	H
1	1±0	0.7381235±0.1872802	0.7838294±0.02176725	0.8890349±0.02614074
7	1±0***	0.5106506±0.06241506*	0.2189659±0.1547011	0.2737854±0.04011261
13	1±0*	0.4064078±0.05230881**	0.4132321±0.003375583**	0.7526957±0.01475578

Time (day)	*F2* Relative expression			
	P	S	M	H
1	1±0***	0.5936894±0.06582111***	1.777728±0.01742604**	2.130004±0.2292309
7	1±0	0.5279561±0.08076297*	0.5226942±0.08248904*	0.8136721±0
13	1±0*	0.600176±0.07141063***	0.6872194±0.02581726***	1.264809±0.03839443
Time (day)	*F5* Relative expression			
	P	S	M	H
1	1±0	0.332691±0.05679326	0.7120463±0.1523783	0.9667831±0.2238352
7	1±0*	1.163835±0.1441385*	1.537236±0.56442*	3.241997±1.045898
13	1±0***	2.103039±0.3894399***	5.094015±0.1851157*	7.145596±1.20831
Time (day)	*F7* Relative expression			
	P	S	M	H
1	1±0***	0.3491078±0.005133064***	2.928524±0.7152218**	4.785264±0.5771259
7	1±0***	0.7042646±0.07579293**	1.16758±0.2628936*	2.645736±0.7794414
13	1±0***	1.964126±0.07699437***	4.196167±0.3423964**	6.255023±0.6020027

H, hepatic plate-mimetic group; M, mixed 3D group; S, sandwich group; P, mixed 2D/plate group. Comparison between H and other groups: $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$

Table S9. Comparison of gene expression levels of hepatocyte structural polarity proteins (mean±standard deviation, n=3)

Time (day)	*ZO-1* Relative expression			
	P	S	M	H
1	1±0	0.5073725±0.07433575	1.222372±0.11168	1.482834±0.012113
7	1±0***	1.516752±0.07926027***	2.522051±0.5839198*	3.796855±0.7213826
13	1±0***	2.741595±0.2638595***	8.06509±0.06588193***	5.293767±1.107504
Time (day)	*Occludin* Relative expression			
	P	S	M	H
1	1±0	0.4976248±0.0664674	1.027969±0.06545456	1.755333±0.02580946
7	1±0***	1.117287±0.0000001192093***	3.455173±0.1185203**	5.627337±1.71052
13	1±0***	1.108751±0.04527339***	4.069943±0.0199476*	5.656756±0.3787575
Time (day)	*MRP2* Relative expression			
	P	S	M	H
1	1±0	0.4758608±0.01321485	0.777628±0.1514861	0.9487422±0.04338546
7	1±0	1.350917±0.1409964	1.404503±0.8161396	1.892837±0.7494608
13	1±0**	1.06113±0.0433289**	4.342758±0.1206	3.068679±0.865601
Time (day)	*DPPIV* Relative expression			
	P	S	M	H
1	1±0	0.5002428±0.06276498*	0.6416527±0.2306971	1.116004±0.005470071
7	1±0***	1.016999±0.2435388***	1.12588±0.2998816**	2.107581±0.3597825
13	1±0**	0.4835809±0.01026999***	1.218084±0.1651177*	1.810666±0.02251712

Time (day)	*NTCP* Relative expression			
	P	S	M	H
1	1±0	0.2586869±0.03748237	0.4481162±0.01244436	0.763702±0.1229874
7	1±0**	0.6115074±0.08563742***	1.646629±0.3286855*	3.400477±1.46865
13	1±0*	0.4683639±0.02065358**	1.187968±0.02522944*	2.553051±0.2070147
Time (day)	*CD147* Relative expression			
	P	S	M	H
1	1±0***	0.8093291±0.1329447***	1.385459±0.1400982**	1.826723±0.09247769
7	1±0*	0.5580937±0.0555275***	0.7571806±0.137782**	1.315044±0.1740706
13	1±0	0.4717139±0.05153196*	0.5242555±0.002569525*	0.8466558±0.09799007

H, hepatic plate-mimetic group; M, mixed 3D group; S, sandwich group; P, mixed 2D/plate group. Comparison between H and other groups: $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$

Table S10. Significantly enriched KEGG pathways associated with differentially upregulated genes with a fold change of > 1.5 at different time points in the hepatic plate-mimetic 3D culture group (H) and the mixed 3D culture group (M)

Comparison	Pathway ID	Pathway Term	*P*-value
H1 Vs M1	path:hsa04978	Mineral absorption	0.000157778
	path:hsa00051	Fructose and mannose metabolism	0.000169043
	path:hsa00220	Arginine biosynthesis	0.000259352
	path:hsa01230	Biosynthesis of amino acids	0.00120009
	path:hsa04066	HIF-1 signaling pathway	0.001342566
	path:hsa04710	Circadian rhythm	0.001468243
	path:hsa01210	2-Oxocarboxylic acid metabolism	0.002387932
	path:hsa04150	mTOR signaling pathway	0.002749162
	path:hsa00750	Vitamin B6 metabolism	0.003741977
	path:hsa00260	Glycine, serine and threonine metabolism	0.003813379
	path:hsa00524	Butirosin and neomycin biosynthesis	0.005183426
	path:hsa04950	Maturity onset diabetes of the young	0.008213594

path:hsa00330	Arginine and proline metabolism	0.008491769
path:hsa00310	Lysine degradation	0.0097392
path:hsa05130	Pathogenic Escherichia coli infection	0.011824311
path:hsa04370	VEGF signaling pathway	0.016806951
path:hsa04713	Circadian entrainment	0.01868368
path:hsa05211	Renal cell carcinoma	0.021832632
path:hsa00562	Inositol phosphate metabolism	0.027689487
path:hsa00910	Nitrogen metabolism	0.030209023
path:hsa05220	Chronic myeloid leukemia	0.030270637
path:hsa04931	Insulin resistance	0.031576834
path:hsa00565	Ether lipid metabolism	0.035939195
path:hsa01200	Carbon metabolism	0.036079
path:hsa04670	Leukocyte transendothelial migration	0.042239228
path:hsa00520	Amino sugar and nucleotide sugar metabolism	0.042316647
path:hsa05144	Malaria	0.044560597
path:hsa04510	Focal adhesion	0.045254979

	path:hsa04110	Cell cycle	1.86E-05
	path:hsa03320	PPAR signaling pathway	0.000286215
	path:hsa04610	Complement and coagulation cascades	0.000658424
	path:hsa04975	Fat digestion and absorption	0.000939358
	path:hsa04977	Vitamin digestion and absorption	0.000940273
	path:hsa04976	**Bile secretion**	0.002054999
	path:hsa00051	Fructose and mannose metabolism	0.003178358
	path:hsa00532	Glycosaminoglycan biosynthesis-chondroitin sulfate / dermatan sulfate	0.005827585
H7 Vs M7	path:hsa04964	Proximal tubule bicarbonate reclamation	0.008690179
	path:hsa04914	Progesterone-mediated oocyte maturation	0.010024493
	path:hsa04918	Thyroid hormone synthesis	0.010439708
	path:hsa00062	Fatty acid elongation	0.010982863
	path:hsa03010	Ribosome	0.013769553
	path:hsa00330	Arginine and proline metabolism	0.014009647
	path:hsa00630	Glyoxylate and dicarboxylate	0.01501891

	metabolism	
path:hsa00130	Ubiquinone and other terpenoid-quinone biosynthesis	0.017201333
path:hsa05204	Chemical carcinogenesis	0.018598831
path:hsa00561	Glycerolipid metabolism	0.024351538
path:hsa00790	Folate biosynthesis	0.027436293
path:hsa04114	Oocyte meiosis	0.027777323
path:hsa00590	Arachidonic acid metabolism	0.028602292
path:hsa00040	Pentose and glucuronate interconversions	0.029379993
path:hsa00380	Tryptophan metabolism	0.038527762
path:hsa00260	Glycine, serine and threonine metabolism	0.038527762
path:hsa00190	Oxidative phosphorylation	0.038593421
path:hsa00360	Phenylalanine metabolism	0.039534921
path:hsa05219	Bladder cancer	0.041016071
path:hsa00511	Other glycan degradation	0.043940917
path:hsa00980	Metabolism of xenobiotics by	0.047768876

		cytochrome P450	
	path:hsa04120	Ubiquitin mediated proteolysis	0.003058741
	path:hsa05020	Prion diseases	0.005744056
	path:hsa04115	p53 signaling pathway	0.005823348
	path:hsa04724	Glutamatergic synapse	0.006781086
	path:hsa05200	Pathways in cancer	0.008219621
	path:hsa03013	RNA transport	0.009188028
	path:hsa03050	Proteasome	0.01086583
H13 Vs M13	path:hsa05202	Transcriptional dysregulation in cancers	0.011347171
	path:hsa03040	Spliceosome	0.013123
	path:hsa00330	Arginine and proline metabolism	0.015376417
	path:hsa04550	Signaling pathways regulating pluripotency of stem cells	0.016514069
	path:hsa04072	Phospholipase D signaling pathway	0.017445152
	path:hsa03460	Fanconi anemia pathway	0.017971936
	path:hsa05217	Basal cell carcinoma	0.019829741

	path:hsa00500	Starch and sucrose metabolism	0.020797018
	path:hsa04150	mTOR signaling pathway	0.024922433
	path:hsa00563	Glycosylphosphatidylinositol (GPI)-anchor biosynthesis	0.028014266
	path:hsa04919	Thyroid hormone signaling pathway	0.035243312
	path:hsa05216	Thyroid cancer	0.036907338
	path:hsa00650	Butanoate metabolism	0.036907338
	path:hsa04920	Adipocytokine signaling pathway	0.037023613
	path:hsa04071	Sphingolipid signaling pathway	0.037145999
	path:hsa00052	Galactose metabolism	0.039279326
	path:hsa04110	Cell cycle	0.041126985
	path:hsa04710	Circadian rhythm	0.041707886
	path:hsa04215	Apoptosis - multiple species	0.044191601

H1/M1, Day 1; H7/M7, Day 7; H13/M13, Day 13

Table S11. Differentially expressed genes associated with bile secretion selected were analyzed by qPCR (mean±standard deviation, n=3)

Time (day)	*AQP8* Relative expression		Time (day)	*MRP2* Relative expression	
	M	H		M	H
1	1±0*	1.990887±0.05900449	1	1±0	0.8312579±0.008148334
7	1±0**	2.983497±0.4612111	7	1±0**	1.529937±0.02999271
13	1±0	0.6265533±0.02353821	13	1±0***	2.956692±0.1255563
Time (day)	*MDR1* Relative expression		Time (day)	*MDR3* Relative expression	
	M	H		M	H
1	1±0	0.7204214±0.01882982	1	1±0	1.189309±0.1415074
7	1±0*	1.545329±0.09083691	7	1±0*	1.384652±0.0142716
13	1±0**	2.710086±0.2419823	13	1±0**	2.221313±0.08183114

Time (day)	*FXR* Relative expression	
	M	H
1	1±0	1.305289±0.1128833
7	1±0**	2.036736±0.08866728
13	1±0***	2.892278±0.1280152

Time (day)	*RXRα* Relative expression	
	M	H
1	1±0*	1.427805±0.05130831
7	1±0**	2.231346±0.131162
13	1±0*	1.481471±0.1498071

Time (day)	*SHP* Relative expression	
	M	H
1	1±0**	1.578386±0.02836432
7	1±0**	1.681224±0.09608293
13	1±0	0.9976249±0.094392

Time (day)	*NTCP* Relative expression	
	M	H
1	1±0	1.370812±0.09398874
7	1±0*	1.491654±0.03898769
13	1±0**	2.071188±0.158884

Time (day)	*OST-α* Relative expression	
	M	H
1	1±0**	1.836304±0.03599872
7	1±0***	2.290733±0.1084924
13	1±0	1.116624±0.05288485

H, hepatic plate-mimetic group; M, mixed 3D group. Comparison between H and M: $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$