第一相反応酵素（CYP 遺伝子）、第二相反応酵素（UGT、GST 遺伝子）、第三相反応酵素（肝関連トランスポーター）の発現を調べたところ、多くの遺伝子はヒト初代培養肝細胞と同程度であった。しかしながら、9 種類の薬剤の代謝プロファイルを調べたところ、分化誘導肝細胞の薬物代謝能は確認されたものの、ヒト初代培養肝細胞よりも劣るものであった。

ナノピラープレートを用いた三次元培養法と FOXA2、HNF1α 遺伝子を導入する技術を併用することで、HepG2 細胞よりも感度良く肝毒性化合物の毒性を検出できる分化誘導肝細胞の作製に成功した。

今後、新たな三次元培養法・共培養法の開発ならびに新規肝成熟化因子の同定とその過剰発現などを行うことにより、分化誘導肝細胞さらなる肝成熟化の促進が期待される。

F. 研究発表

1. 論文発表

1) Takayama K., Inamura M., Kawabata K., Sugawara M., Kikuchi K., Higuchi M., Nagamoto Y., Watanabe H., Tashiro K., Sakurai F., Hayakawa T., Furue MK., Mizuguchi H., Generation of metabolically functioning hepatocytes from human pluripotent stem cells by FOXA2 and HNF1α transduction. ***J. Hepatol.***, 57, 628-636 (2012)

2) Nagamoto Y., Tashiro K., Takayama K., Ohashi K., Kawabata K., Sakurai F., Tachibana M., Hayakawa T., Furue MK., Mizuguchi H., Promotion of hepatic maturation of human pluripotent stem cells in 3D co-culture using type I collagen and Swiss 3T3 cell sheets. ***Biomaterials***, 33, 4526-4534 (2012)

3) 水口裕之、高山和雄、長基康人、川端健二：ヒトiPS細胞から肝細胞への分化誘導の現状と創薬応用、***医薬品レギュラトリーサイエンス***、43、982-987(2012)

4) 早川堯夫、水口裕之：ヒトiPS細胞の再生医療及び創薬研究への応用の現状と展望、***Brain and Nerve***、64, 47-57 (2012)

5) Takayama K., Kawabata K., Nagamoto Y., Kishimoto K., Tashiro K., Sakurai F., Tachibana M., Kanda K., Hayakawae T., Furue MK., Mizuguchi H., 3D spheroid culture of hESC/hiPSC-derived hepatocyte-like cells for drug toxicity testing. ***Biomaterials***, 34, 1781–1789 (2013)

6) Kawabata K., Takayama K., Nagamoto Y., Saldon M.S., Higuchi M., Mizuguchi H., Endodermal and hepatic differentiation from human embryonic stem cells and human induced pluripotent stem cells. ***J. Stem Cell Res. Ther.***, S-10-002 (2012).

7) 高山和雄、川端健二、水口裕之：ヒト多能性幹細胞から肝細胞への分化誘導法の開発とその毒性評価系への応用、*組織培養研究*、印刷中

8) 高山和雄、川端健二、水口裕之：ヒトES/iPS細胞から肝細胞への高効率分化誘導法の開発とその創薬応用、*最新医学*、68、141-144 (2013)

9) 川端健二、高山和雄、水口裕之：ヒト iPS細胞由来分化誘導肝細胞を用いた薬物毒性評価系の開発、*BIO INDUSTRY*、30、19-24 (2013)

10) Takayama K., Inamura M., Kawabata K., Katayama K., Higuchi M., Tashiro K., Nonaka A., Sakurai F., Hayakawa T., Furue MK., Mizuguchi H. Efficient generation of functional hepatocytes from human embryonic stem cells and induced pluripotent stem cells by HNF4 α transduction. ***Mol. Ther.***, 20:127-37 (2012).

11) Takayama K., Inamura M., Kawabata K., Tashiro K., Katayama K., Hayakawa T., Furue MK., Mizuguchi H. Efficient and selective generation of two distinct endoderm lineages from human ES and iPS cells by differentiation stage-specific SOX17 transduction. ***PLoS One***, 6, e21780 (2011).

12) Yoshida T., Takayama K., Kondoh M., Sakurai F., Tani H., Sakamoto N., Matsuura Y., Mizuguchi H., Yagi K. Use of human hepatocyte-like cells derived from induced pluripotent stem cells as a model for hepatocytes in hepatitis C virus infection. ***Biochem Biophys Res Commun.***, 416: 119-24, (2011).

13) Kawabata K., Inamura M., Mizuguchi H. Efficient Hepatic Differentiation of Human iPS Cells by Gene Transfer. ***Methods Mol Biol.***, 826: 115-124 (2012)

2. 学会発表

1) 高山和雄、川端健二、田代克久、神田勝弘、櫻井文教、古江－楠田美保、水口裕之、Nanopillarプレートを用いたヒトES/iPS細胞から肝細胞への分化誘導法の開発とその毒性評価への応用、第 12 回日本再生医療学会総会、神奈川、2013 年 3 月

2) 渡辺仁、高山和雄、稲村充、立花雅史、櫻井文教、川端健二、水口裕之、ヒトES/iPS細胞由来内胚葉から肝幹前駆細胞への分化過程における転写因子HEXの機能解明、第 35 回日本分子生物学会年会、福岡、2012 年 12 月 11-14 日

3) Takayama K., Inamura M., Kawabata K., Sugawara M., Kikuchi K., Tashiro

K., Sakurai F., Furue MK., Mizuguchi H., Generation of mature hepatocytes from human pluripotent stem cells by FOXA2 and HNF1α transduction, 第27回日本薬物動態学会年会、千葉、2012年11月

4) 渡辺仁、高山和雄、稲村充、立花雅史、櫻井文教、古江-楠田美保、川端健二、水口裕之、ヒトES/iPS細胞から肝幹前駆細胞への分化における転写因子HEXの機能解明、第62回日本薬学会近畿支部総会・大会、兵庫、2012年10月

5) 高山和雄、稲村　充、川端健二、菅原道子、菊池きよ美、櫻井文教、古江ー楠田美保、水口裕之、FOXA2およびHNF1α遺伝子導入によるヒトES/iPS細胞から薬剤代謝能を有した肝細胞への分化誘導、第39回日本毒性学会学術年会、宮城、2012年7月

6) 長基康人、　田代克久、高山和雄、大橋一夫、岡野光夫、櫻井文教、立花雅史、古江-楠田美保、川端健二、水口裕之、ヒトES/iPS細胞由来肝細胞のSwiss 3T3 細胞との積層 3 次元共培養下における成熟化・促進機構の解析、第19回肝細胞研究会、北海道、2012年6月

7) 高山和雄、川端健二、稲村　充、櫻井文教、古江ー楠田美保、水口裕之、c/EBPαおよびc/EBPβ遺伝子によるTGFBR2遺伝子発現制御を介した肝幹前駆細胞の運命決定、第19回大会肝細胞研究会、北海道、2012年6月

8) 長基康人、田代克久、高山和雄、　大橋一夫、櫻井文教、立花雅史、古江(楠田)美保、川端健二、水口裕之、Swiss 3T3細胞との積層 3 次元共培養下におけるヒトES/iPS細胞由来肝細胞の肝細胞成熟化・促進機構の解析、第85回大会日本組織培養学会、京都、2012年5月

9) 高山和雄、稲村　充、川端健二、形山和史、櫻井文教、古江ー楠田美保、水口裕之、SOX17、HEX、HNF4α遺伝子導入によるヒト多能性幹細胞から成熟肝細胞の効率良い分化誘導、第85回大会日本組織培養学会、京都、2012年5月

10) Nagamoto Y., Tashiro K., Takayama K., Ohashi K., Kawabata K., Tachibana M., Sakurai F., Furue MK., Mizuguchi H., Type I collagen promotes hepatic maturation from human pluripotent stem cells in 3D co-culture with Swiss 3T3 cell sheet. International Society for Stem Cell Research 10th Annual Meeting, 神奈川, 2012年6月

11) Takayama K., Inamura M., Kawabata

K., Sugawara M., Kikuchi K., Higuchi M., Nagamoto Y., Watanabe H., Tashiro K., Sakurai F., Furue MK., Mizuguchi H., Generation of metabolically functioning hepatocytes from human embryonic stem cells and induced pluripotent stem cells by transduction of FOXA2 and HNF1α, International Society for Stem Cell Research 10th Annual Meeting, 神奈川, 2012年6月

12)渡辺仁、高山和雄、稲村充、立花雅史、櫻井文教、川端健二、水口裕之、転写因子HEXによるヒトES/iPS細胞由来内胚葉から肝幹前駆細胞への分化機構の解明、第12回日本再生医療学会総会、神奈川、2013年3月高山和雄、稲村 充、川端健二、菅原道子、菊池きよ美、櫻井文教、古江－楠田美保、水口裕之：FOXA2・HNF1α遺伝子導入によるヒト多能性幹細胞から薬剤代謝能を有した肝細胞の分化誘導、日本薬学会第 132 年会、札幌、2012 年 3 月 28-31 日

13)長基康人、田代克久、高山和雄、大橋一夫、櫻井文教、立花雅史、古江(楠田)美保、川端健二、水口裕之: 3 次元共培養法によるヒトES・iPS 細胞由来肝細胞の効率的な分化誘導法の開発、日本薬学会第132 年会、札幌、2012年3 月 28-31 日

14)Kazuo Takayama、Mitsuru Inamura、Kenji Kawabata、Kazufumi Katayama、Katsuhisa Tashiro、Fuminori Sakurai、Miho Kusuda Furue 、Hiroyuki Mizuguchi: EFFICIENT GENERATION OF MATURE HEPATOCYTES FROM HUMAN PLURIPOTENT STEM CELLS BY HNF4α TRANSDUCTION, 第 26 回日本薬物動態学会年会, 広島, 2011 年 11 月 16-18 日

15)水口裕之: 創薬応用を目指したヒトES/iPS 細胞から肝細胞への分化誘導技術開発、第 1 回レギュラトリーサイエンス学会学術大会、東京、2011 年 9 月 2-3 日

16)水口裕之: ヒトES/iPS 細胞から肝細胞への分化誘導技術の開発とin vitro毒性評価系への応用、第 38 回日本トキシコロジー学会学術年会、東京、2011 年 7 月 13 日

17)高山和雄、稲村 充、川端健二、田代克久、形山和史、櫻井文教、古江－楠田美保、水口裕之: HNF4α遺伝子導入によるヒトES・iPS 細胞からの成熟肝細胞への高効率分化誘導, 第 18 回肝細胞研究会, 東京, 2011 年 6 月 24-25 日

18) Kazuo Takayama, Mitsuru Inamura, Kenji Kawabata, Kazufumi Katayama, Katsuhisa Tashiro, Fuminori Sakurai, Miho Kusuda Furue, Hiroyuki Mizuguchi: HNF4 α promotes hepatic maturation from human embryonic stem cell-derived hepatoblasts., The 6th Seoul-Kyoto-Osaka Joint Symposium on Pharmaceutical Sciences for Young Scientists, Seoul, Korea, June, 2011

19) Kazuo Takayama, Mitsuru Inamura, Kenji Kawabata, Kazufumi Katayama, Katsuhisa Tashiro, Fuminori Sakurai, Miho Kusuda Furue, Hiroyuki Mizuguchi: EFFICIENT GENERATION OF FUNCTIONAL HEPATOCYTES FROM HUMAN EMBRYONIC STEM CELLS AND INDUCED PLURIPOTENT STEM CELLS BY HNF4 α TRANSDUCTION., International Society for Stem Cell Research, Tronto, June, 2011

3. その他

G. 知的財産権の出願・登録状況

1. 特許取得

1) 水口裕之、川端健二、高山和雄(発明人)、幹細胞から肝細胞への分化誘導方法、特願 2012-128872 号

2. 実用新案登録

Figure 1

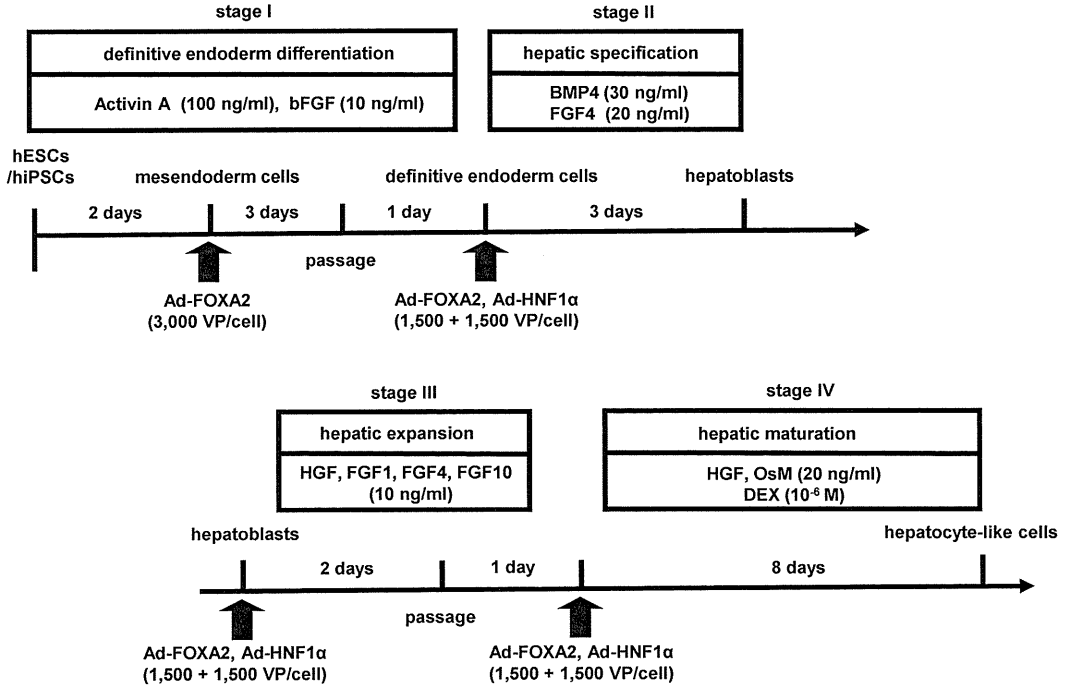

Figure 1 Hepatic differentiation of hESCs and hiPSCs by FOXA2 and HNF1α transduction.
The procedure for differentiation of hESCs and hiPSCs into hepatocyte-like cells via definitive endoderm cells and hepatoblasts is presented schematically. Details of the hepatic differentiation procedure are described in the Materials and Methods section.

Figure 2

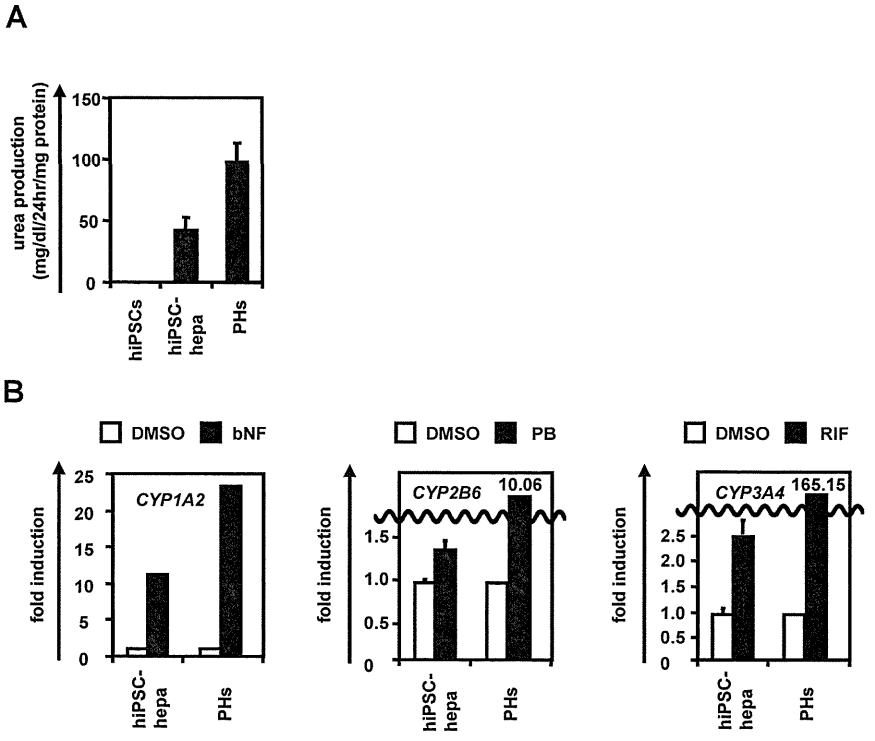

Figure 2 The urea secretion capacity and CYP induction potency of hiPSC-hepa.
hiPSCs (Dotcom) were differentiated into hepatocyte-like cells as described in **Figure 1**. (A) The amount of urea secretion was examined in hiPSCs, hiPSC-hepa (day 20 of the differentiation), and primary human hepatocytes (48hr), which were cultured for 48 hr after plated. (B) Induction of *CYP1A2*, *2B6*, or *3A4* by DMSO (white bar) or inducer (black bar; β-naphthoflavone [bNF], phenobarbital [PB], or rifampicin [RIF]) of hiPSC-hepa and PHs, which were cultured for 48 hr after the cells were plated, was examined. The inducers used in this figure is summarized in **Figure 3**. On the y axis, the gene expression levels of *CYP1A2*, *2B6*, or *3A4* in DMSO-treated cells, which were cultured for 48 hr, were taken as 1.0. All data are represented as means ± SD (n=3).

Figure 3

CYP	Inducer	Conc. (μM)
CYP1A2	β-naphthoflavone	10
CYP2B6	Phenobarbital	750
CYP3A4	Rifampicin	10

Figure 3 List of CYP inducers used in this study

Figure 4

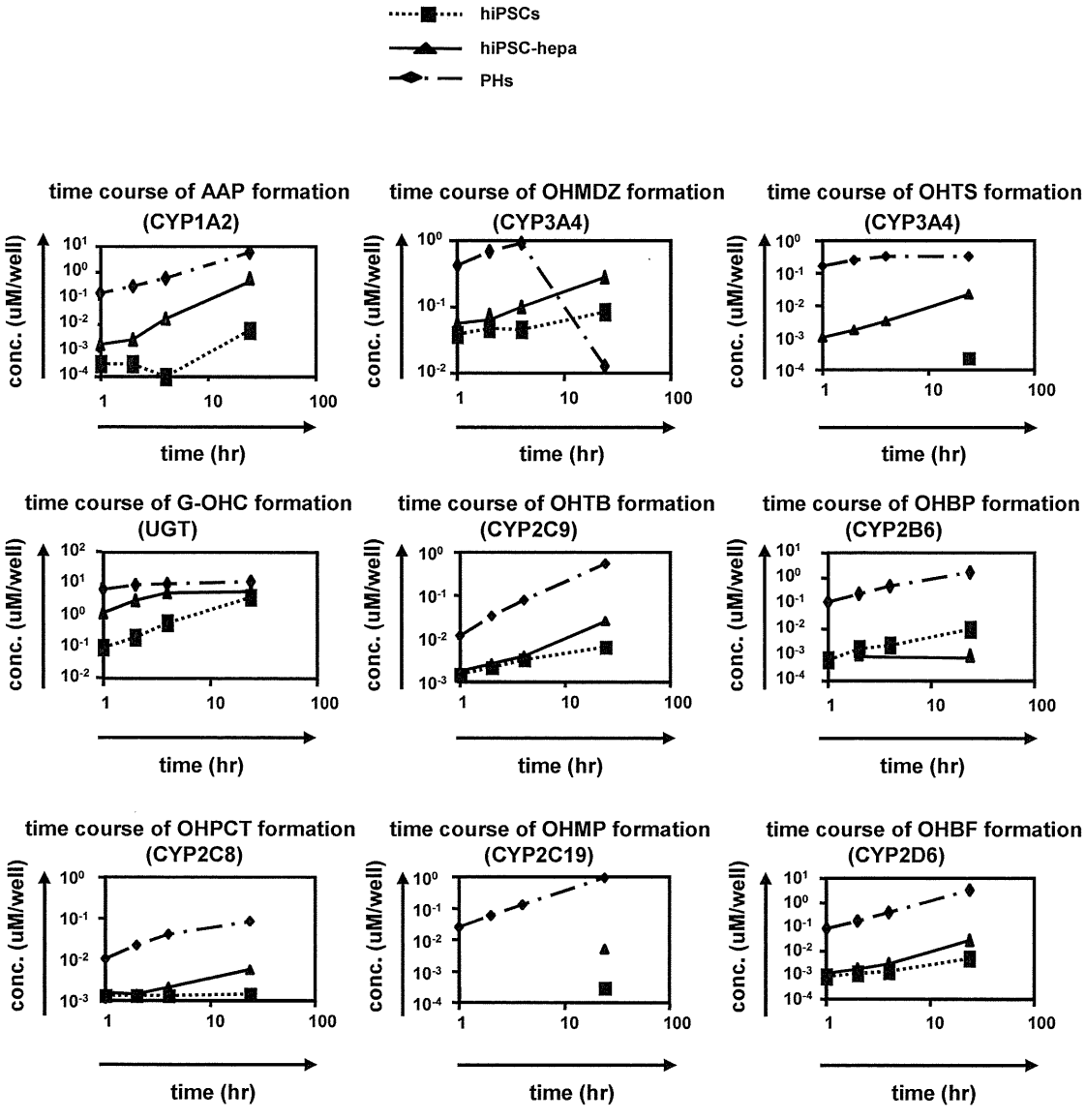

Figure 4 The time course of metabolites formation in hiPSCs, hiPSC-hepa, or PHs.
hiPSCs (Dotcom) were differentiated into hepatocytes as described in Figure 1. Quantitation of metabolites in hiPSCs, hiPSC-hepa, and PHs treated with nine substrates (Phenacetin [PHE], Bupropion [BP], Paclitazel [PCT], Tolbutamide [TB], *S*-mephenytoin [MP], Bufuralol [BF], Midazolam [MDZ], Testosterone [TS], and Hydroxyl coumarin [OHC]) was performed. Supernatants were collected at 1, 2, 4, or 24 hr after incubation with each substrate, which were the probes for CYP1A2, 2B6, 2C8, 2C9, 2C19, 2D6, 3A4, 3A4 and UGT, respectively. The quantity of metabolites (Acetaminophen [AAP], Hydroxybupropion [OHBP], 6α-hydroxypaclitaxel [OHPCT], Hydroxytolbutamide [OHTB], 4'-hydroxymephenytoin [OHMP], 1'-hydroxybufuralol [OHBF], 1'-hydroxymidazolam [OHMDZ], 6β-hydroxytestosterone [OHTS], 7-Hydroxycoumarin glucuronide [G-OHC], respectively) was measured by LC-MS/MS. The substrates and that metabolites used in this study are summarized in Figure 5. All data are represented as means ± SD (*n*=3).

Figure 5

CYP	Substrate	Sub Abbr.	Conc. (μM)	Reaction
CYP1A2	Phenacetin	PHE	10	O-de-ethylation
CYP2B6	Bupropion	BP	150	Hydroxylation
CYP2C8	Paclitaxel	PCT	20	6α-Hydroxylaion
CYP2C9	Tolbtamide	TB	500	Hydroxylation
CYP2C19	S-mephenytoin	MP	200	4'-hydroxylation
CYP2D6	Bufuralol	BF	50	1'-hydroxylation
CYP3A4	Midazolam	MDZ	10	1'-hydroxylation
CYP3A4	Testosterone	TS	100	6β-hydroxylation
UGT	Hydroxy coumarin	OHC	10	Glucuronidation

CYP	Metabolites	Metabolites Abbr.	Detection limit of metabolite (μM)
CYP1A2	Acetaminophen	AAP	0.0031
CYP2B6	Hydroxybupropion	OHBP	0.006
CYP2C8	6α-hydroxypaclitaxel	OHPCT	0.0031
CYP2C9	Hydroxytolbutamide	OHTB	0.001
CYP2C19	4'-hydroxymephenytoin	OHMP	0.003
CYP2D6	1'-hydroxybufuralol	OHBF	0.003
CYP3A4	1'-hydroxymidazolam	OHMDZ	0.003
CYP3A4	6β-hydroxytestosterone	OHTS	0.049
UGT	7-Hydroxycoumarin glucuronide	G-OHC	0.015

Figure 5 List of CYP substrates and that metabolites used in this study.

Figure 6

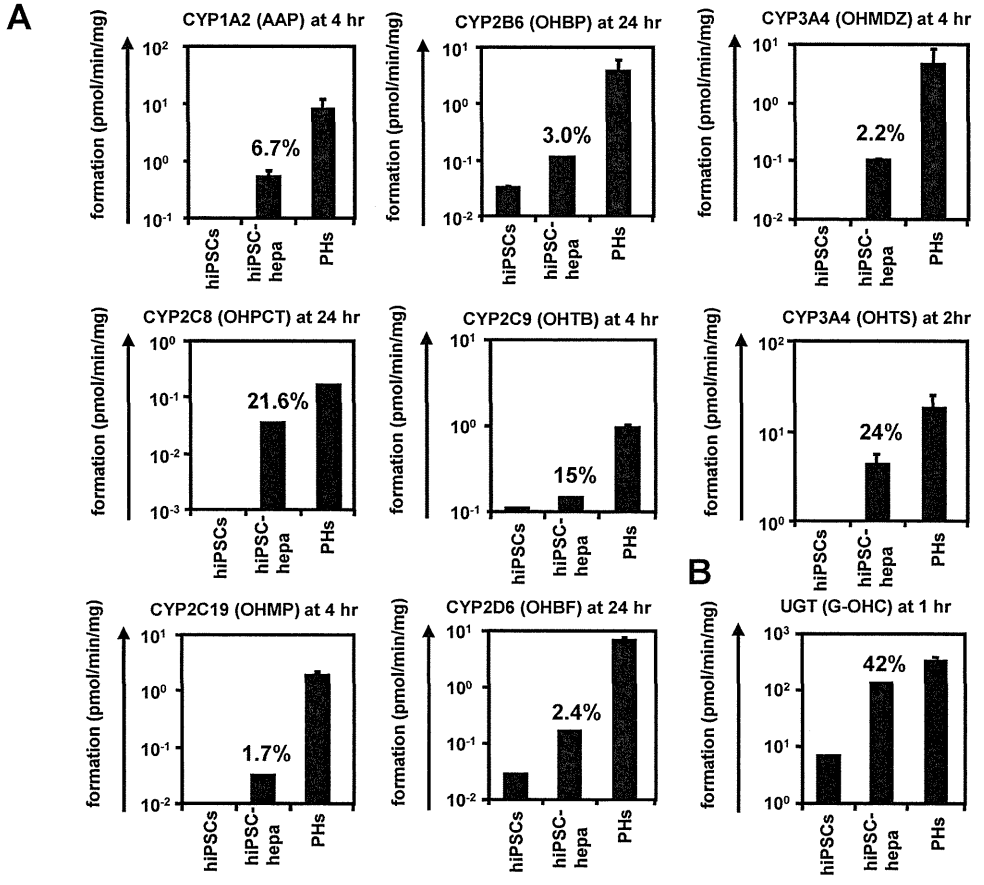

Figure 6 Evaluation of the drug metabolism capacity and hepatic transporter activity of hiPSC-hepa.

The hiPSCs (Dotcom) were differentiated into hepatocytes as described in **Figure 1**. (A and B) Quantitation of metabolites in hiPSCs, hiPSC-hepa, and PHs, which were cultured for 48 hr after the cells were plated, was examined by treating nine substrates (Phenacetin, Bupropion, Paclitazel, Tolbtamide, *S*-mephenytoin, Bufuralol, Midazolam, Testosterone, and Hydroxyl coumarin; these compounds are substrates for CYP1A2, 2B6, 2C8, 2C9, 2C19, 2D6, 3A4, 3A4 (A) and UGT (B), respectively), and then supernatants were collected at the indicated time. The quantity of metabolites (Acetaminophen [AAP], Hydroxybupropion [OHBP], 6a-hydroxypaclitaxel [OHPCT], Hydroxytolbutamide [OHTB], 4'-hydroxymephenytoin [OHMP], 1'-hydroxybufuralol [OHBF], 1'-hydroxymidazolam [OHMDZ], 6b-hydroxytestosterone [OHTS], 7-Hydroxycoumarin glucuronide [G-OHC], respectively) was measured by LC-MS/MS. The substrates and their metabolites are summarized in **Figure 5**. The ratios of the activity levels in hiPSC-hepa to the activity levels in PHs rate are indicated in the graph. All data are represented as means ± SD (n=3).

Figure 7

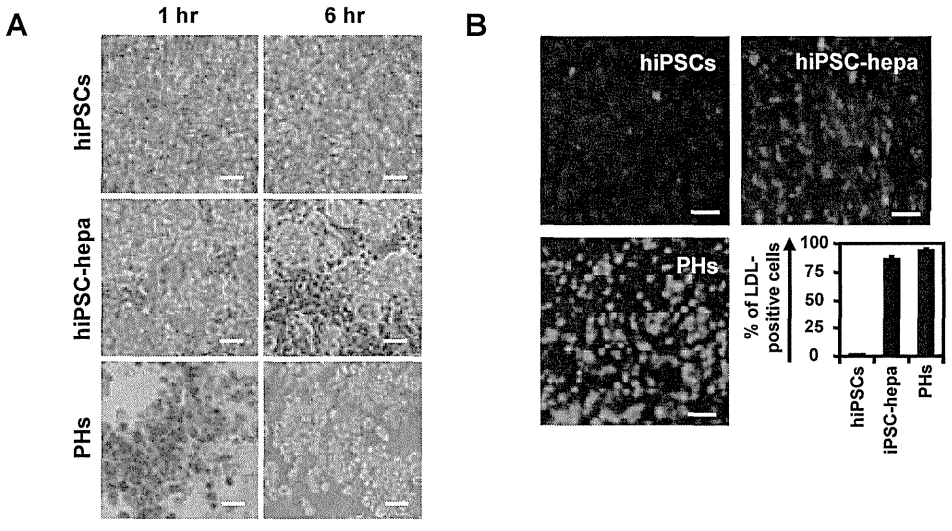

Figure 7 The hepatic characterization of hiPSC-hepa.
The hiPSCs (Dotcom) were differentiated into hepatocytes as described in Figure 1. (A) hiPSCs, hiPSC-hepa, and PHs were examined for their ability to take up ICG (left column) and release it 6 hr thereafter (right column). (B) hiPSCs, hiPSC-hepa, and PHs were cultured with medium containing Alexa-Flour 488-labeled LDL (green) for 1 hr, and immunohistochemistry was performed. Nuclei were counterstained with DAPI (blue). The scale bars represent 50 μm. The percentage of LDL-positive cells was also measured by FACS analysis. All data are represented as means ± SD (n=3).

Figure 8

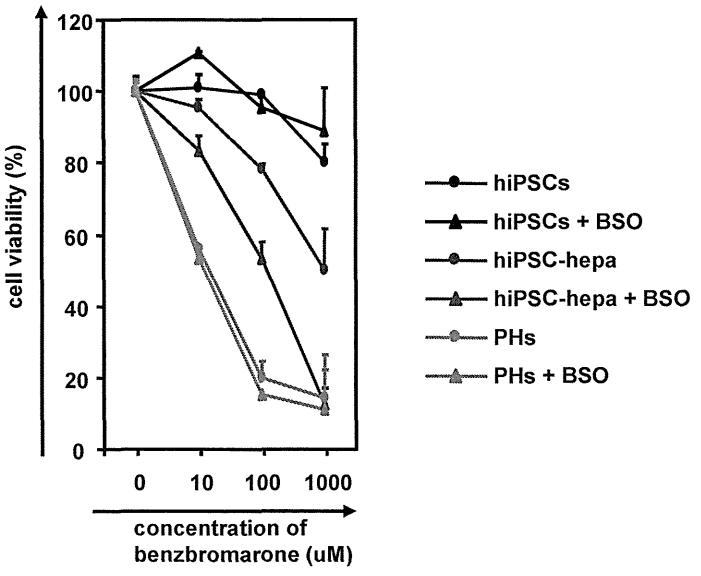

Figure 8 hiPSC-hepa have a potential to be applied in drug screening.
The hiPSCs (Dotcom) were differentiated into hepatocytes as described in Figure 1. The cell viability of hiPSCs (black bar), hiPSC-hepa (blue bar), PHs (red bar), and their BSO-treated cells (0.4 mM BSO was pre-treated for 24 hr) was assessed by Alamar Blue assay after 48 hr exposure to different concentrations of benzbromarone. The cell viability is expressed as a percentage of that in the cells treated only with solvent. All data are represented as means ± SD (*n*=3).

Figure 9

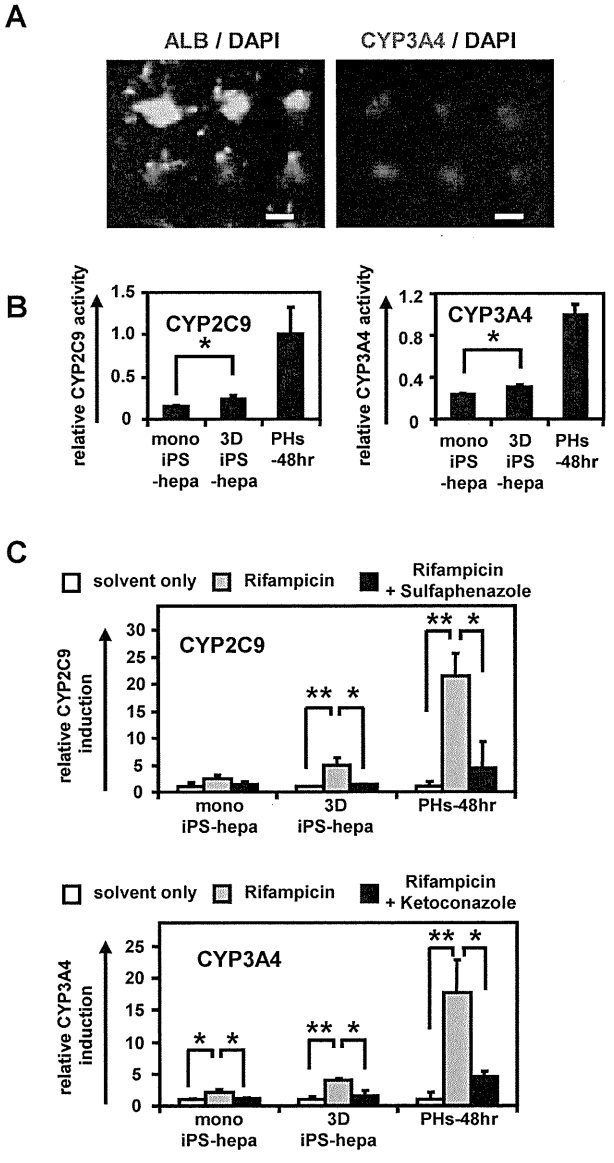

Figure 9 Drug metabolism capacity and CYP induction potency were examined in the 3D iPS-hepa. (A) The 3D iPS-hepa (day 35) were subjected to immunostaining with anti-ALB (green) or CYP3A4 (red) antibodies. Nuclei were counterstained with DAPI (blue). Scale bar represents 100 μm. (B) The CYP activity was measured in the mono iPS-hepa (day 20), the 3D iPS-hepa (day 35), and PHs-48hr. On the y axis, the CYP activity in PHs-48hr was taken as 1.0. (C) Induction of CYP2C9 (left) or CYP3A4 (right) by DMSO (solvent only; white bar), Rifampicin (gray bar), or rifampicin and CYP inhibitor (Sulfaphenazole or Ketoconazole, black bar) in the mono iPS-hepa, the 3D iPS-hepa, and PHs-48hr. On the y axis, the CYP activity of the cells that have been cultured in DMSO-containing medium was taken as 1.0. *P<0.05; **P<0.01.

Figure 10

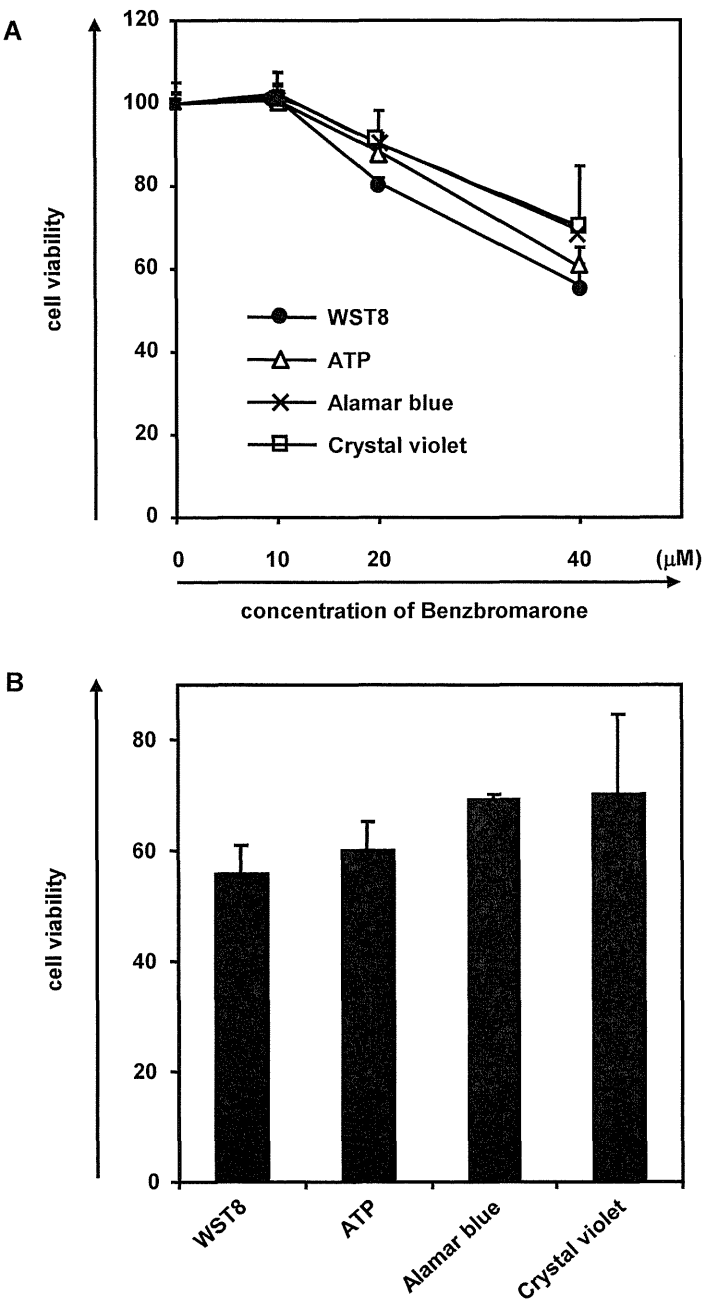

Figure 10 WST-8 assay was the most sensitive method for detecting the drug-induced cytotoxicity.
(A) The cell viability of 3D iPSC (Dotcom)-hepa (day 35) was assessed by WST-8, ATP, Alamar blue, or Crystal violet assay after 24 hr exposure to different concentrations (or 40 μM (B))of Benzbromarone. The cell viability is expressed as the percentage of cells treated with solvent only.

Figure 11

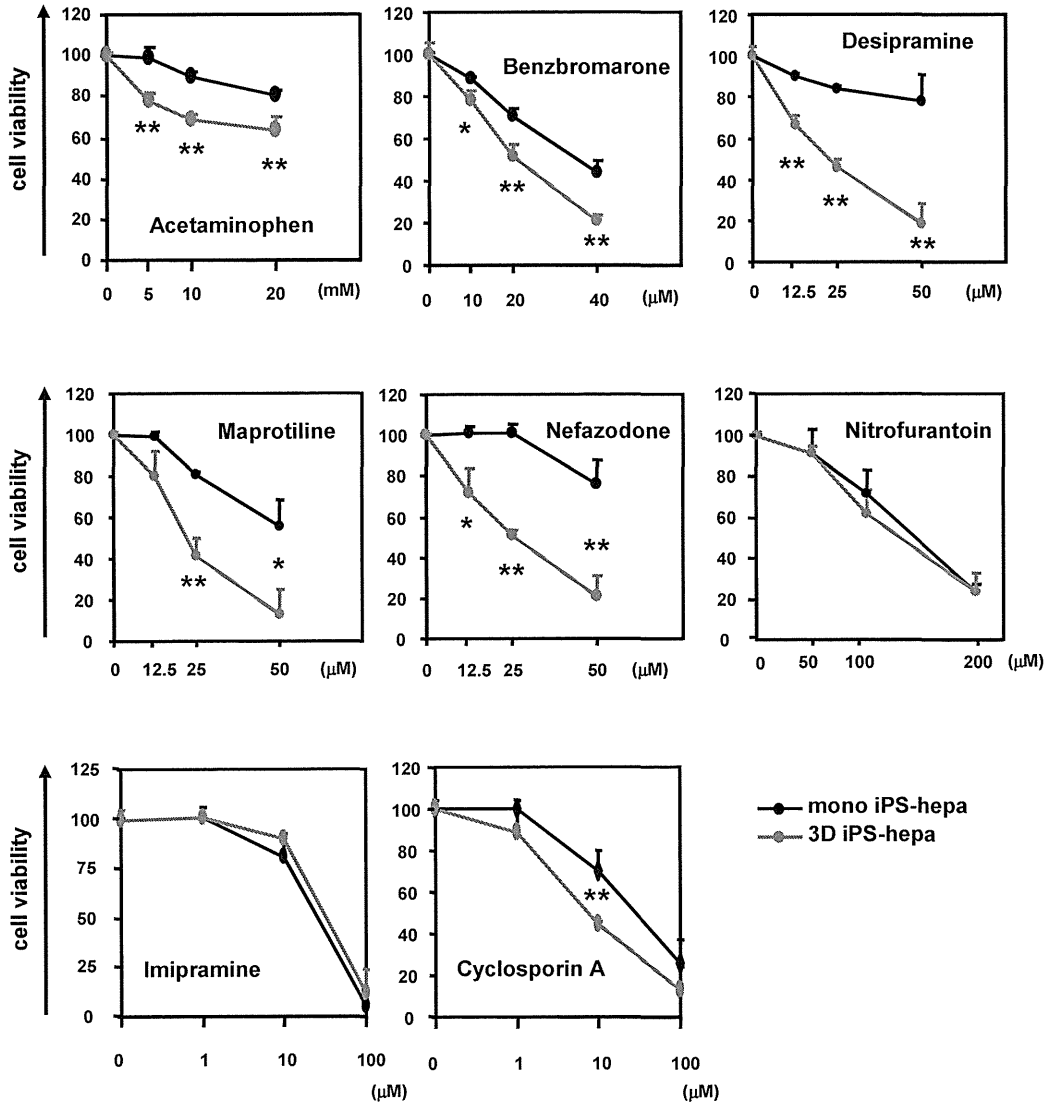

Figure 11 The drug-induced cytotoxicity was more sensitively detected in the 3D iPS-hepa than in the mono iPS-hepa.
On day 11, the hiPSC (Dotcom)-derived cells were plated onto the Nanopillar Plate or the flat plate, and then the cells were cultured until day 35. The cell viability of 3D iPSC-hepa was assessed by WST-8 assay after 24 hr exposure to different concentrations of drugs. Cell viability is expressed as a percentage of cells treated with solvent only.

Figure 12

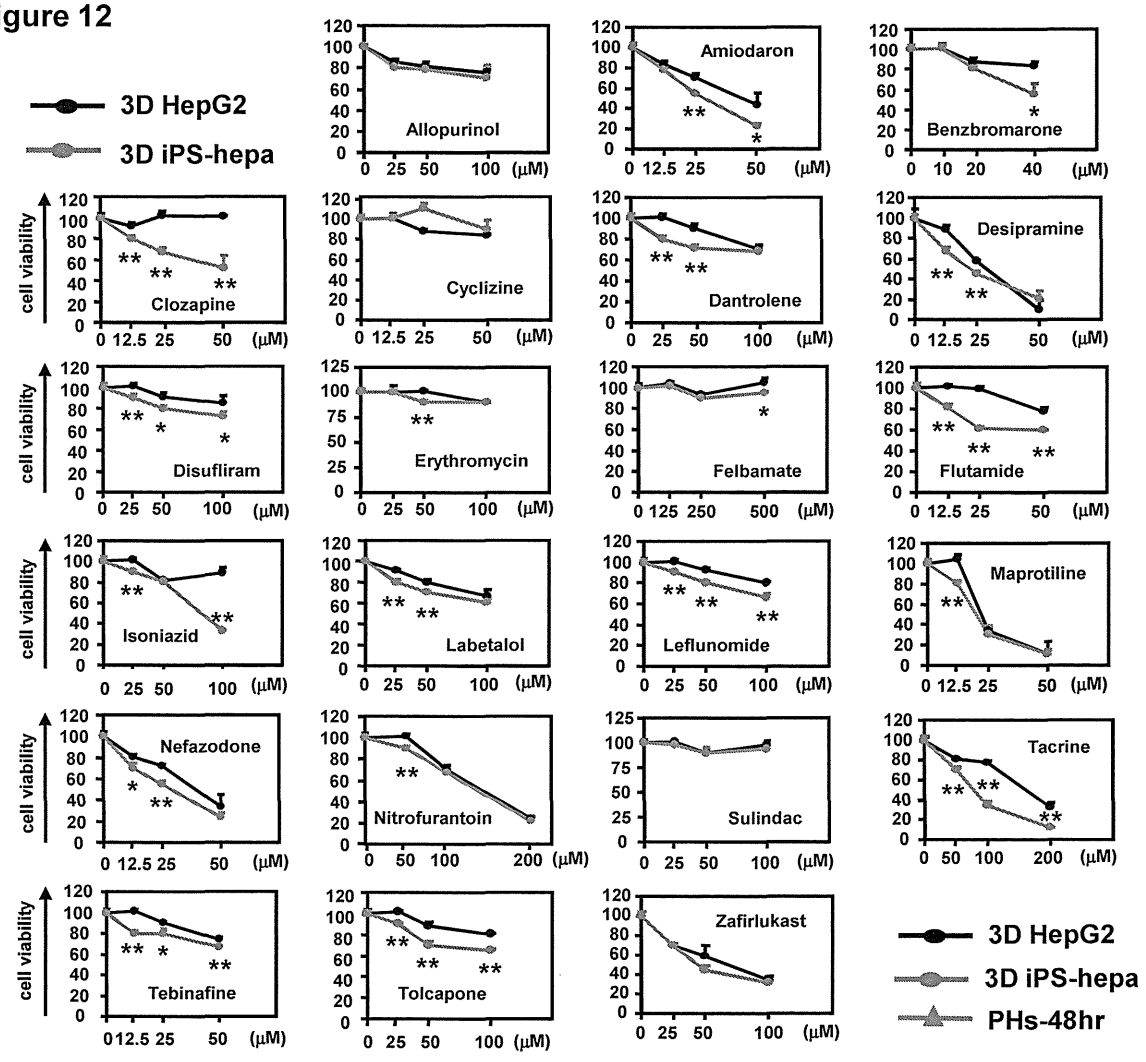

Figure 12 The possibility of applying 3D iPS-hepa to drug testing was examined.
The cell viability of the 3D HepG2 (black) and 3D iPSC-hepa (red) were assessed by WST-8 assay after 24 hr exposure to different concentrations of 22 test compounds. Cell viability is expressed as a percentage of cells treated with solvent only. **P*<0.05; ***P*<0.01 (The data of 3D iPS-hepa was compared with that of 3D HepG2).

Figure 13

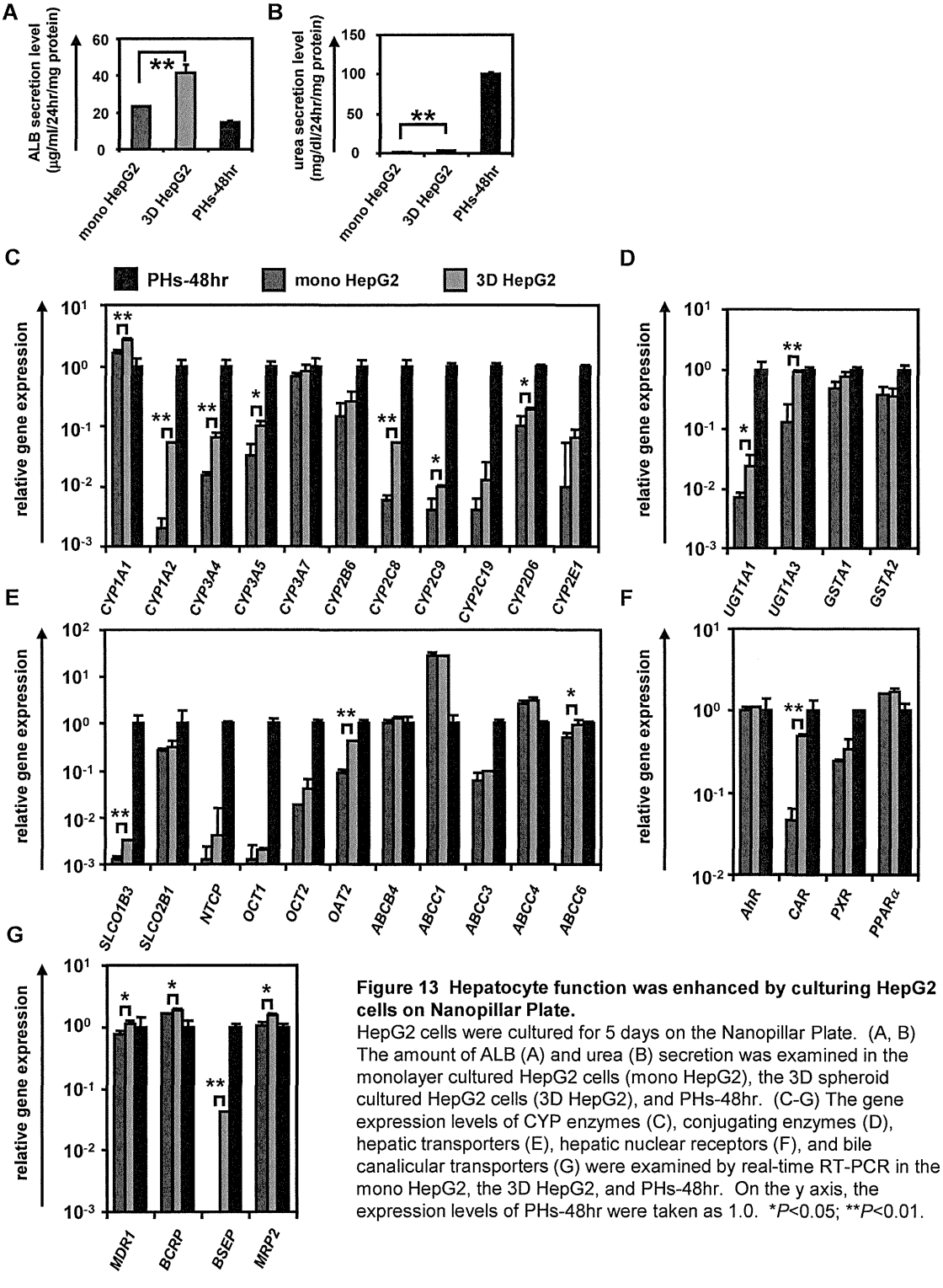

Figure 13 Hepatocyte function was enhanced by culturing HepG2 cells on Nanopillar Plate.

HepG2 cells were cultured for 5 days on the Nanopillar Plate. (A, B) The amount of ALB (A) and urea (B) secretion was examined in the monolayer cultured HepG2 cells (mono HepG2), the 3D spheroid cultured HepG2 cells (3D HepG2), and PHs-48hr. (C-G) The gene expression levels of CYP enzymes (C), conjugating enzymes (D), hepatic transporters (E), hepatic nuclear receptors (F), and bile canalicular transporters (G) were examined by real-time RT-PCR in the mono HepG2, the 3D HepG2, and PHs-48hr. On the y axis, the expression levels of PHs-48hr were taken as 1.0. *P<0.05; **P<0.01.

Figure 14

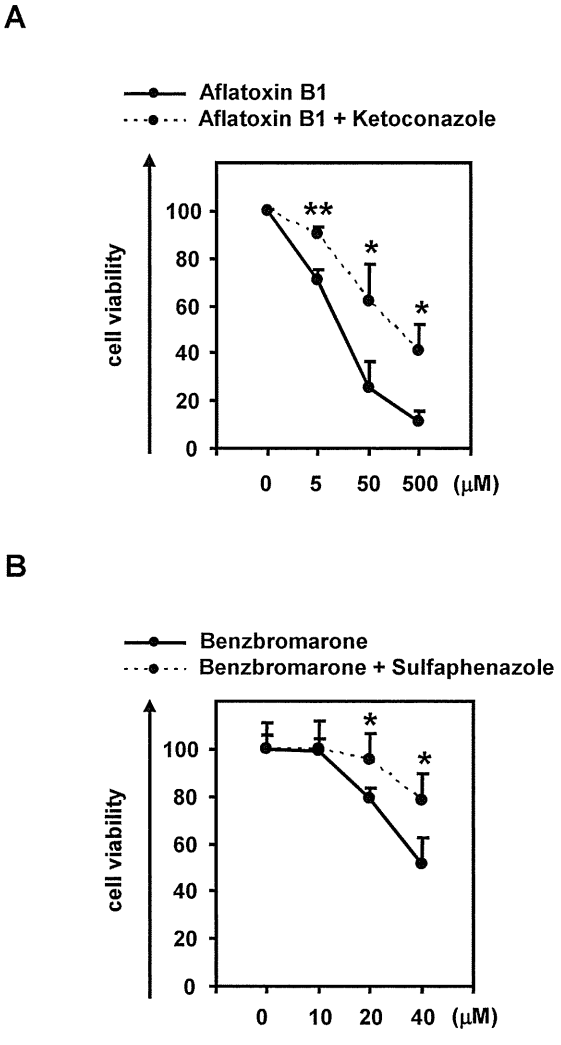

Figure 14 Drug-induced cytotoxicity in the 3D iPS-hepa is mediated by cytochrome P450.
(A, B) The cell viability of the 3D iPSC-hepa was assessed by WST-8 assay after 24 hr exposure to different concentrations of (A) Aflatoxin B1 and (B) Benzbromarone in the presence or absence of the CYP3A4 or 2C9 inhibitor, Ketoconazole or Sulfaphenazole, respectively. Cell viability was expressed as the percentage of cells treated with solvent only. *P<0.05; **P<0.01.

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総合・分担研究報告書

ヒトiPS細胞の肝細胞への高効率分化誘導法の開発

分担研究者　川端　健二
独立行政法人　医薬基盤研究所
創薬基盤研究部　幹細胞制御プロジェクト　プロジェクトリーダー

本研究では、ヒト人工多能性幹（induced pluripotent stem （iPS））細胞から肝細胞への高効率分化誘導法の開発を目的とする。

平成23年度は、非常に高い遺伝子導入効率を示すアデノウイルス(Ad)ベクターを用いて肝分化に必須の7種類の遺伝子（FOXA2、HEX、HNF1α、HNF1ß、HNF4α、HNF6、SOX17）をヒトES/iPS細胞から肝細胞への各分化過程において導入し、さらなる肝成熟化を促進できる遺伝子の同定を試みた。ヒトES/iPS細胞から肝細胞への各分化過程において、7種類の肝関連転写因子を遺伝子導入するスクリーニングを実施した結果、FOXA2とHNF1α遺伝子を組み合わせて導入ことによって、さらなる肝成熟化が確認された。FOXA2とHNF1α遺伝子を導入して作製された分化誘導肝細胞は、ヒト初代培養肝細胞と同程度の肝関連遺伝子の発現が確認された。しかしながら、ヒトES/iPS細胞由来肝細胞（分化誘導肝細胞）のシトクロムP450（CYP）活性がヒト初代培養肝細胞よりも劣っていたため、分化誘導肝細胞のさらなる成熟化が必要であった。

そこで平成24年度は、肝成熟化（肝幹前駆細胞から肝細胞への分化）をこれまで用いていた単層培養条件でなく三次元培養条件で行うことにより、分化誘導肝細胞のさらなる成熟化を目指した。我々は三次元培養を行うために、ナノピラープレート（株式会社　日立ハイテクノロジーより供与）を用いた。その結果、単層培養の肝分化誘導法と比較して、より高い肝機能（各種肝関連遺伝子発現、アルブミン産生能、尿素産生能など）を有した分化誘導肝細胞の作製が可能になった。

以上のことから、ナノピラープレートを用いた三次元培養法および肝関連遺伝子を適切な分化段階で導入する技術を用いることで、効率良く高い肝機能を有した分化誘導肝細胞を作製できることが明らかになった。本成果が創薬における *in vitro* 毒性スクリーニングに応用されることを期待する。

研究協力者

水口裕之　(独)医薬基盤研究所
大阪大学大学院薬学研究科

櫻井文教　大阪大学大学院薬学研究科

高山和雄　(独)医薬基盤研究所
大阪大学大学院薬学研究科