pixlbio

CASE STUDY

Decoding biology.
Pixel by pixel.

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Best in class liver disease modeling with pixHep iPSC-derived hepatocytes

pixlbio's proprietary differentiation protocols enable largescale generation of iPSC-derived hepatocytes (pixHep) with field leading purity and functionality. Importantly, pixHep cells successfully recapitulate key aspects of disease pathophysiology across a wide range of conditions that affect different aspects of liver function.

Advantages

Demonstrate characteristic hepatocyte cobblestone morphology

Express comparable levels of liver maturity markers to primary human hepatocytes

Express higher levels of urea cycle markers and secrete higher levels of urea compared to liver carcinoma cell lines

Demonstrate comparable levels of CYP450 markers and CYP3A4 activity to primary human hepatocytes

Demonstrate functional localization and function of ASGR1 for GalNAc-dependent drug deliveries

Standardized cell product containing iPSC-derived human hepatocytes producing reproducible and biologically relevant data

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pixHep demonstrate the characteristic hepatocyte cobblestone morphology

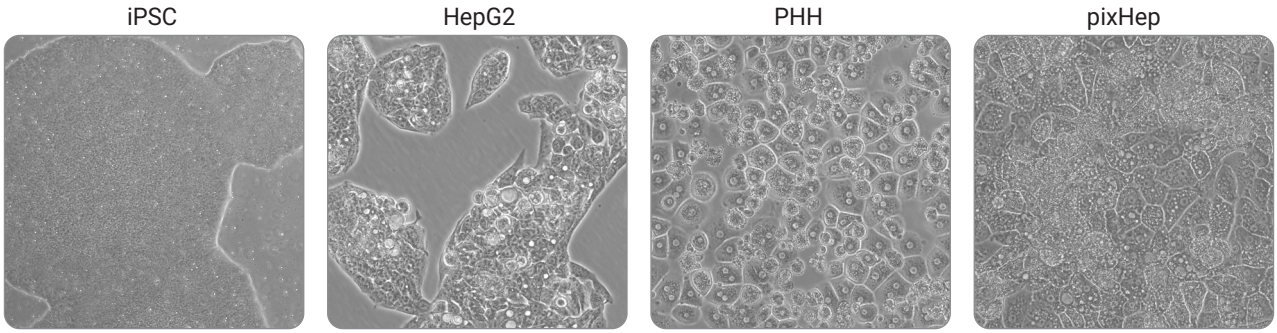

Figure 1: Representative cell morphology images of induced pluripotent stem cells (iPSCs), hepatocellular carcinoma HepG2 cells, primary human hepatocytes (PHH) and pixlbio pixHeps. The pictures reveal the characteristic cobblestone morphology of pixHep, and the presence of a uniform monolayer following >3 weeks of iPSC differentiation. Magnification: 20x.

pixHeps express similar levels of liver maturity markers compared to primary human hepatocytes (PHH)

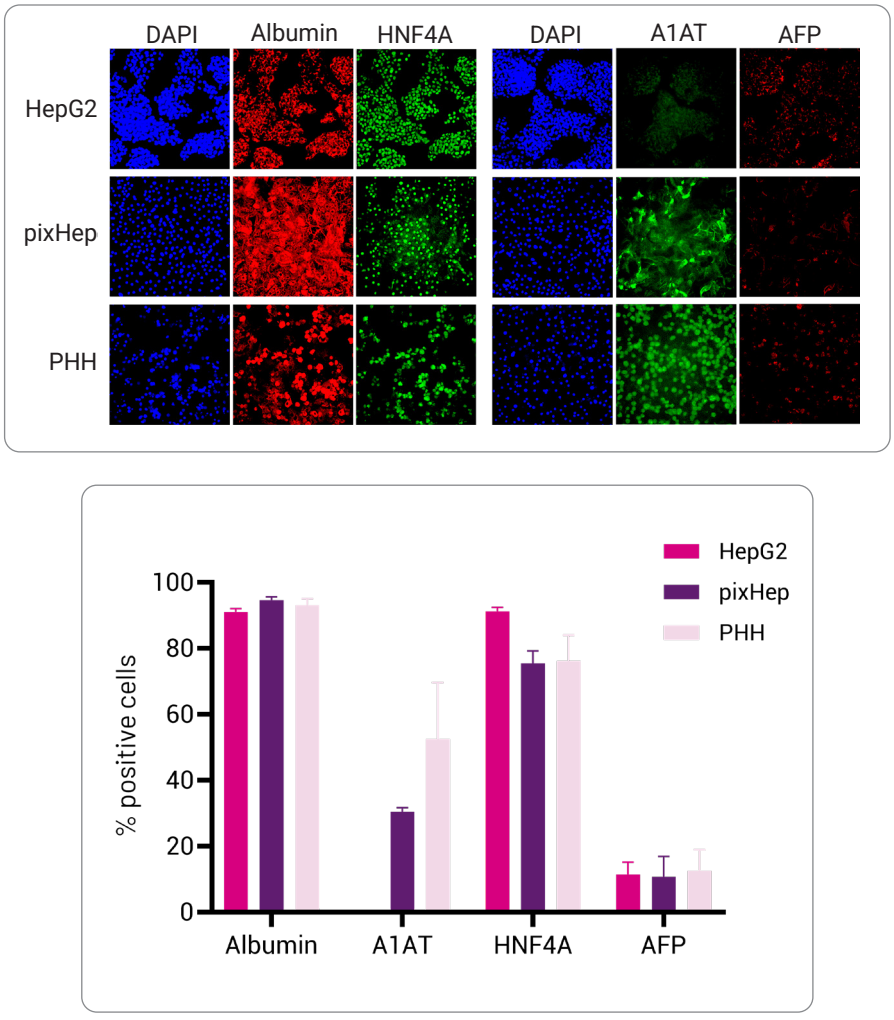

Figure 2: Representative immunocytochemistry images and protein quantification showing expression levels of the hepatocyte maturity markers albumin, alpha-1-antitrypsin (A1AT), hepatocyte nuclear factor 4 (HNF4A), and alpha fetoprotein (AFP) in liver carcinoma HepG2 cells, pixHep, and primary human hepatocytes (PHH; 3 donors). Data are presented as mean±SEM of n=3 independent experiments.

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pixHeps express higher levels of urea cycle markers and secrete higher levels of urea compared to liver carcinoma cells

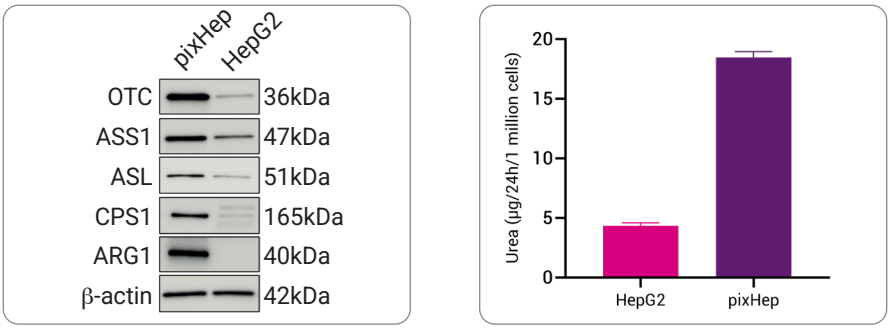

Figure 3: Protein expression levels of the urea cycle enzymes OTC, ASS1, ASL, CPS1, and ARG1 in liver carcinoma HepG2 cells and pixHeps (left). Urea secretion in liver carcinoma HepG2 cells and pixHeps. Data are presented as mean±SEM of n=3 independent experiments (right).

pixHeps demonstrate functional gluconeogenesis pathway and respond to gluconeogenesis inducers

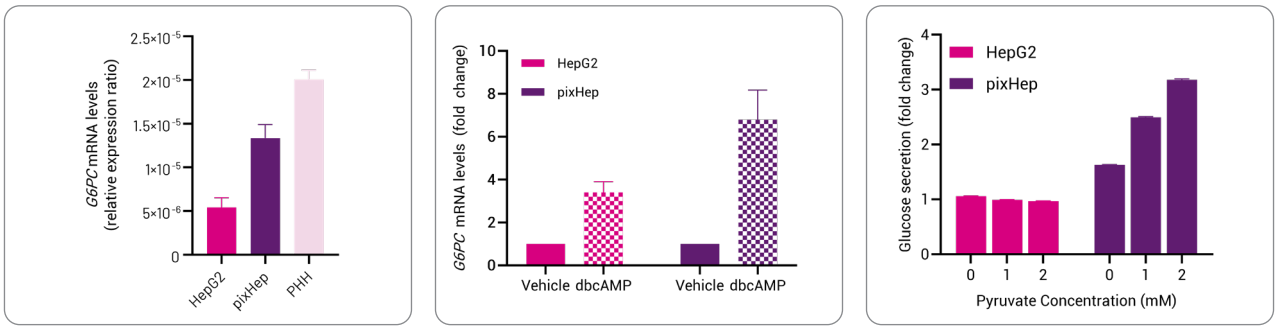

Figure 4: G6PC mRNA levels in liver carcinoma HepG2 cells, pixHeps, and PHHs (left). G6PC mRNA levels in liver carcinoma HepG2 cells and pixHeps treated with 0.1mM dbcAMP (gluconeogenesis inducer)(middle). Glucose secretion in dbcAMP-treated liver carcinoma HepG2 cells and pixHeps upon pyruvate challenge. Data are presented as mean±SEM of n=3 independent experiments. mRNA expression data were normalized to 18S rRNA.

pixHeps demonstrate comparable levels of CYP450 markers and CYP3A4 activity to primary human hepatocytes (PHH)

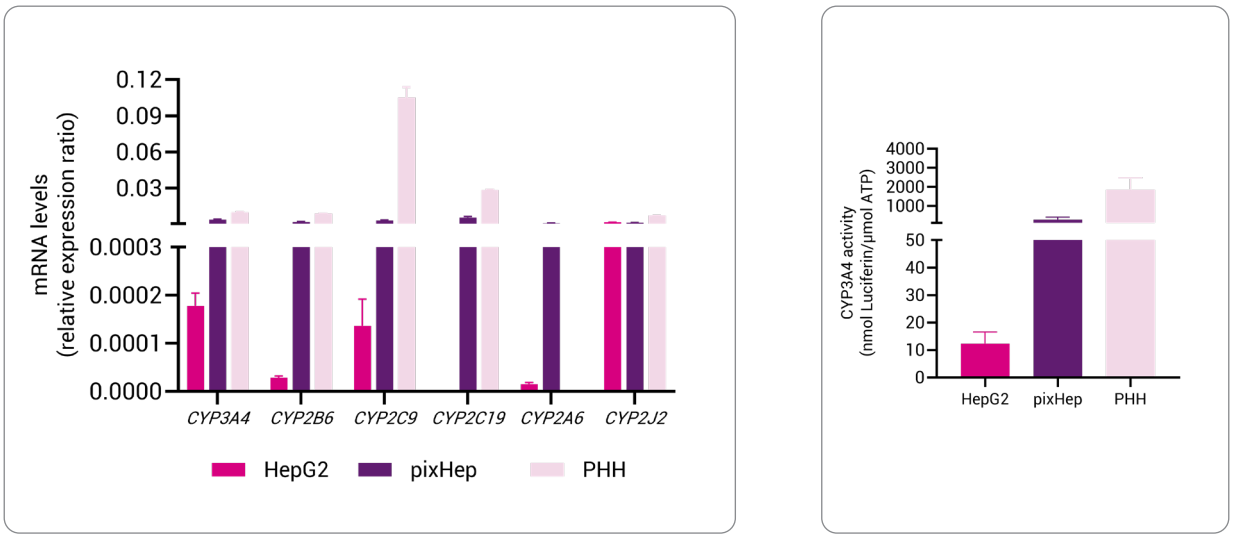

Figure 5: mRNA expression levels of Phase I CYP450 genes in liver carcinoma HepG2 cells, pixHeps, and PHHs (left). Basal CYP3A4 activity in liver carcinoma HepG2 cells, pixHeps, and PHHs (right). mRNA data were normalized to the housekeeping gene 18S rRNA and are presented as mean±SEM of n=3 independent experiments. CYP3A4 activity data were normalized to ATP levels and are presented as mean±SEM of n=3 independent experiments. For PHHs data, cells from 3 independent donors were used.

CASE STUDY

pixHeps demonstrate functional membrane localization and activity of the Asialoglycoprotein receptor 1 (ASGR1)

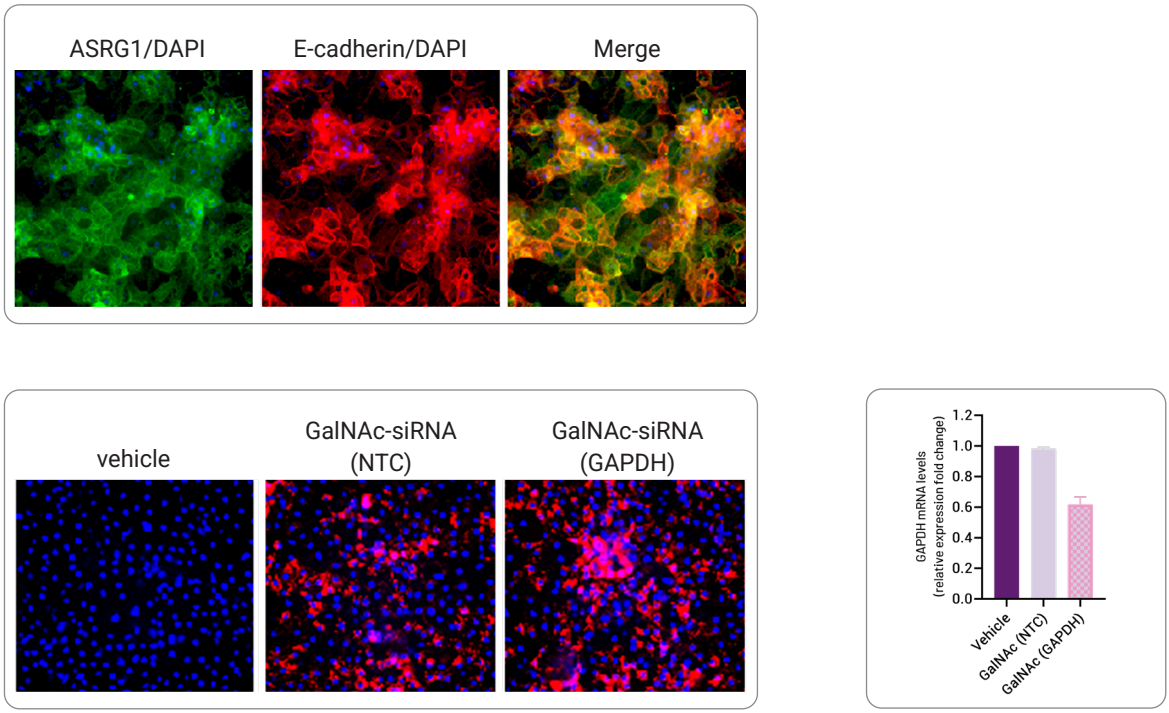

Figure 6: Representative immunocytochemistry images showing the localization of ASGR1 in the pixHep membrane. Cells were counterstained with the membrane marker E-cadherin and DAPI (upper). The effect of ASGR1 in the transport of GalNAc-siRNA conjugate targeting GAPDH in pixHeps using GalNAc-Cy3 staining (bottom left) and qPCR (bottom right). Data are presented as mean±SEM of n=3 independent experiments. mRNA expression data were normalized to 18S rRNA. NTC: non-template control.

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