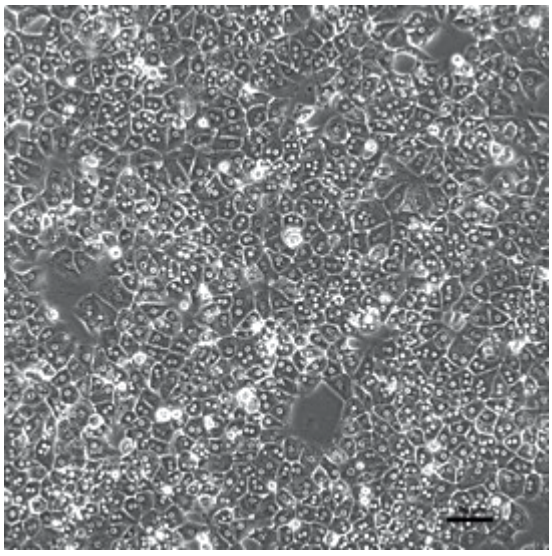
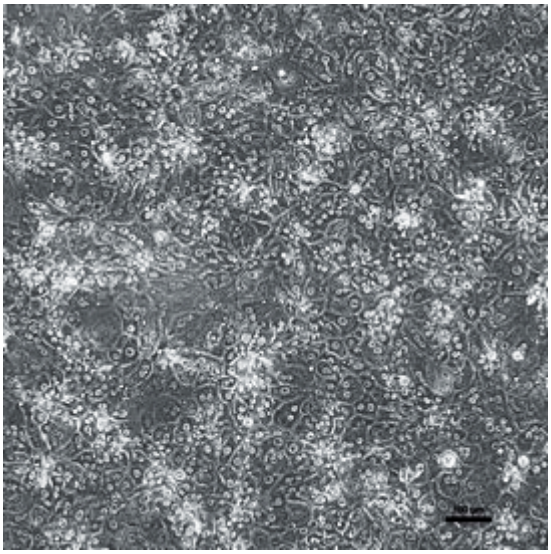
PRIMACYT
Cell Culture Technology

RHCP-I Rat Hepatocytes Cryopreserved Plateable for Induction assays **Cell Specification – Certificate of Analysis (CoA)**	
Lot RH190514	Batch Release: July 25, 2022

Donor data	
Species: Rattus norvegicus forma domestica Strain: Wistar:Han IGS rat // Crl:Wi(Han)	Gender: female Age: 9 months

Cryopreservation and Thawing	
Cryopreservation: Date: May 14, 2019 Amount per vial: 10.0 x 10^6 cells	**Thawing:** n=2 Post-thaw viability: 87.5 ± 2.9 % Post-thaw yield per vial: 4.7 ± 1.5 x 10^6 cells Recovery: 47.0 %

2D culture	
Phase contrast on day 1 after thawing (24well plate)	Phase contrast on day 3 after thawing (24well plate)
	

Recommended seeding density on collagen-coated plates:
24well plate – 300,000 cells/well // 96well plate – 60,000 cells/well
Culture in Human Hepatocyte Maintenance Medium (HHMM).

CYP P450 activity in 2D culture after thawing:	pmol/(mg × min)	X-fold induction
Ethoxyresorufin-O-deethylation:	24well: 88.0 ± 7.8	13.3
Induction with 25 µM ß-Naphthoflavone	96well: 163.3 ± 47.0	15.9

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Suspension culture
Viability test on orbital shaker (Eppendorf Thermomixer C, 1000 rpm at 37 °C with 0.5 x 10^6 cells in 0.5 ml HPM-Cryo):

Time (h)	0	0.5	1	1.5	2	3	4	5
Viability (%)	84.6	74.3	65.5	61.9	63.5	62.1	54.5	58.3

Detailed animal information and husbandry conditions	
Species	Rat (Rattus norvegicus forma domestica)
Strain	Wistar:Han IGS rat // Crl:Wi(Han), IGS (International Genetic Standard)
Vendor	Charles River Germany GmbH & Co. KG, Sulzbach, FRG
Food	Altromin Maintenance diet for rats/mice (ad libitum), Altromin, Lage, FRG
Water	ad libitum
Light/Dark cycle	07:00-19:00 light / 19:00-07:00 dark (12 hours light/dark)
Temperature	20-24 °C
Humidity	40-70 %
Bedding	Hugro hemp bedding, Saerbeck, FRG
Cage	Tecniplast Eurostandard Type III and Type IV incl. behavioural enrichments for the animals
Animals were housed under veterinary control and allowed to acclimate ≥ 7 days before use. Animal housing permits according to §11 Abs. 1 TSchG, dated March 20, 2017 and March 22, 2022 under supervision of Veterinary Office of Landkreis Ludwigslust/Parchim, FRG. Hepatocytes were obtained from a non-infectious, non-contagious, healthy animal. The animal does not originate from a facility conducting work or research with animal pathogens.	

Issued by: M. Reu	Verified by: A. Ullrich