Supplementary Materials: I*n Vitro* Model for Hepatotoxicity Studies Based on Primary Human Hepatocyte Cultivation in a Perfused 3D Bioreactor System

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Table S1. Overview of the genes differentially expressed between hepatocyte donors in control bioreactors on Day 10 (LogFC: Log_2 fold change in expression).

Entrez-ID	Symbol	Gene Name	logFC	Adjusted *p*-Value
397	*ARHGDIB*	Rho GDP dissociation inhibitor (GDI) β	1.35	0.046
1551	*CYP3A7*	Cytochrome P450, family 3, subfamily A, polypeptide 7	3.94	0.046
4072	*EPCAM*	Epithelial cell adhesion molecule	2.40	0.046
4495	*MT1G*	Metallothionein 1G	−4.92	0.046
4496	*MT1H*	Metallothionein 1H	−5.85	0.0076
30001	*ERO1L*	ERO1-like (S. cerevisiae)	1.05	0.0092
51302	*CYP39A1*	Cytochrome P450, family 39, subfamily A, polypeptide 1	1.075	0.0060
57600	*FNIP2*	Folliculin interacting protein 2	−0.74	0.039
64097	*EPB41L4A*	Erythrocyte membrane protein band 4.1 like 4A	1.73	0.021
79668	*PARP8*	Poly (ADP-ribose) polymerase family, member 8	0.79	0.032
118663	*BTBD16*	BTB (POZ) domain containing 16	1.17	0.046

Table S2. Overview of the genes most significantly affected upon treatment with 300 μM diclofenac (LogFC: Log_2 fold change in expression as compared with control BR).

Entrez-ID	Symbol	Gene Name	logFC	Adjusted *p*-Value
		Top 10 Downregulated Genes		
3108	*HLA-DMA*	Major histocompatibility complex, class II, DM α	−2.01	0.00025
3240	*HP*	Haptoglobin	−1.76	0.00025
7805	*LAPTM5*	Lysosomal protein transmembrane 5	−1.82	0.00025
11010	*GLIPR1*	GLI pathogenesis-related 1	−2.13	0.00052
2207	*FCER1G*	Fc fragment of IgE, high affinity I, receptor for; gamma polypeptide	−2.22	0.00055
51156	*SERPINA10*	Serpin peptidase inhibitor, clade A (α-1 antiproteinase, antitrypsin), member 10	−2.19	0.00055
6659	*SOX4*	SRY (sex determining region Y)-box 4	−1.97	0.00055
6696	*SPP1*	Secreted phospho protein 1	−2.86	0.00055
713	*C1QB*	Complement component 1, q subcomponent, B chain	−1.97	0.00059
6347	*CCL2*	Chemokine (C–C motif) ligand 2	−4.20	0.00065
		Top 10 Upregulated Genes		
337	*APOA4*	Apolipoprotein A-IV	4.075	0.00025
771	*CA12*	Carbonic anhydrase XII	1.47	0.00055
1491	*CTH*	Cystathionine gamma-lyase	2.55	0.00085
1649	*DDIT3*	DNA-damage-inducibletranscript 3	2.27	0.0017
3934	*LCN2*	Lipocalin 2	3.050	0.0022
2673	*GFPT1*	Glutamine-fructose-6-phosphate transaminase 1	1.74	0.0024
4953	*ODC1*	Ornithine decarboxylase 1	1.76	0.0033
6520	*SLC3A2*	Solute carrier family 3 (amino acid transporter heavy chain), member 2	2.41	0.0033
3376	*IARS*	Isoleucyl-tRNA synthetase	1.25	0.0035
84513	*PPAPDC1B*	Phosphatidic acid phosphatase type 2 domain containing 1B	1.11	0.0036

Table S3. Overview of the genes most significantly affected upon treatment with 1000 μM diclofenac (LogFC: Log_2 fold change in expression as compared with control BR).

Entrez-ID	Symbol	Gene Name	logFC	Adjusted *p*-Value
Top 10 Downregulated Genes				
7805	*LAPTM5*	Lysosomal protein transmembrane 5	−6.036	1.13×10^{-11}
1974	*EIF4A2*	Eukaryotic translation initiation factor 4A2	−6.028	2.09×10^{-10}
3015	*H2AFZ*	H2A histone family, member Z	−5.44	2.31×10^{-10}
4069	*LYZ*	Lysozyme	−5.30	8.70×10^{-11}
5516	*PPP2CB*	Protein phosphatase 2, catalytic subunit, β isozyme	−5.71	2.38×10^{-10}
7941	*PLA2G7*	Phospholipase A2, group VII (platelet-activating factor acetylhydrolase, plasma)	−4.72	1.32×10^{-10}
11342	*RNF13*	Ring fingerprotein 13	−4.89	2.75×10^{-10}
6166	*RPL36AL*	Ribosomal protein L36a-like	−5.56	4.64×10^{-10}
9516	*LITAF*	Lipopolysaccharide-induced TNF factor	−4.41	4.48×10^{-10}
5431	*POLR2B*	Polymerase (RNA) II (DNA directed) polypeptide B, 140 kDa	−4.24	5.60×10^{-10}
Top 10 Upregulated Genes				
1178	*CLC*	Charcot-Leydencrystalgalectin	2.61	9.02×10^{-6}
644809	*C15orf56*	Chromosome 15 open readingframe 56	1.85	1.07×10^{-5}
203076	*C8orf74*	Chromosome 8 open readingframe 74	1.38	1.62×10^{-5}
3456	*IFNB1*	Interferon, β 1, fibroblast	1.66	1.83×10^{-5}
91544	*UBXN11*	UBX domainprotein 11	1.41	2.19×10^{-5}
56253	*CRTAM*	Cytotoxic and regulatory T cell molecule	1.18	2.28×10^{-5}
1133	*CHRM5*	Cholinergicreceptor, muscarinic 5	1.224	2.67×10^{-5}
5319	*PLA2G1B*	Phospholipase A2, group IB (pancreas)	1.27	3.14×10^{-5}
9153	*SLC28A2*	Solute carrier family 28 (concentrative nucleoside transporter), member 2	1.33	3.14×10^{-5}
84560	*MT4*	Metallothionein 4	1.60	3.26×10^{-5}

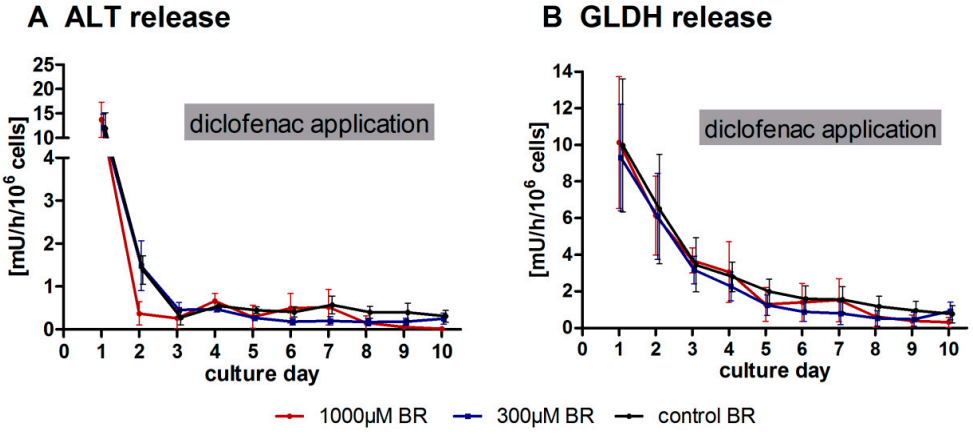

Figure S1. Release rates of alanine transaminase (ALT) and glutamate dehydrogenase (GLDH) in bioreactor with primary human liver cells treated or non-treated with diclofenac. Enzyme release rates in 300 μM bioreactors (300 μM BR) (**blue**) or 1000 μM bioreactors (1000 μM BR) (**red**) or in the non-treated control bioreactors (control BR) (**black**) were determined before (from Day 0 to 3) and during diclofenac application (from Day 3 to 10). ALT (**A**) and GLDH (**B**) were measured as indicators of potential cell damage. Values were normalized to 10^6 inoculated cells. Values were normalized to 10^6 inoculated cells. The influence of the drug dose (Day 3–10) on the metabolic activity of the cells in comparison to the control was analyzed by means of one-way analysis of variance (ANOVA) with Dunnett's multiple comparison test .(n = 3, mean ± SEM).