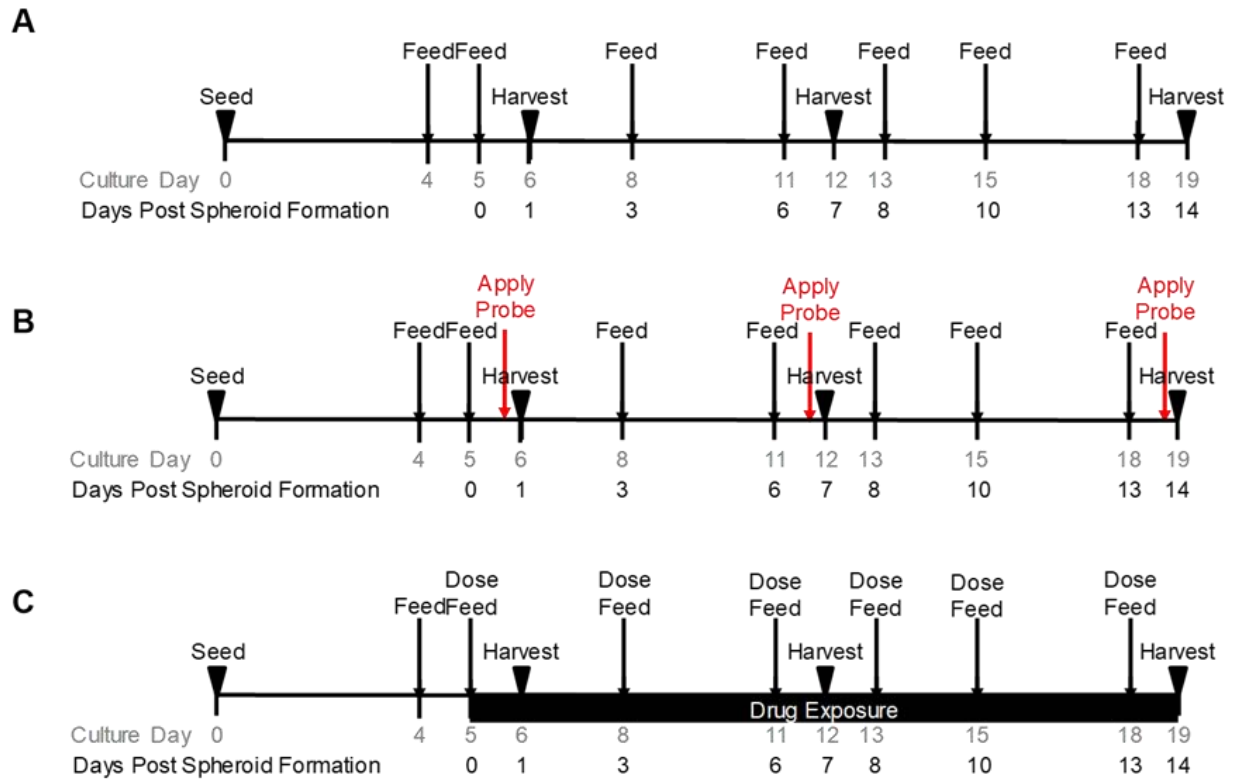


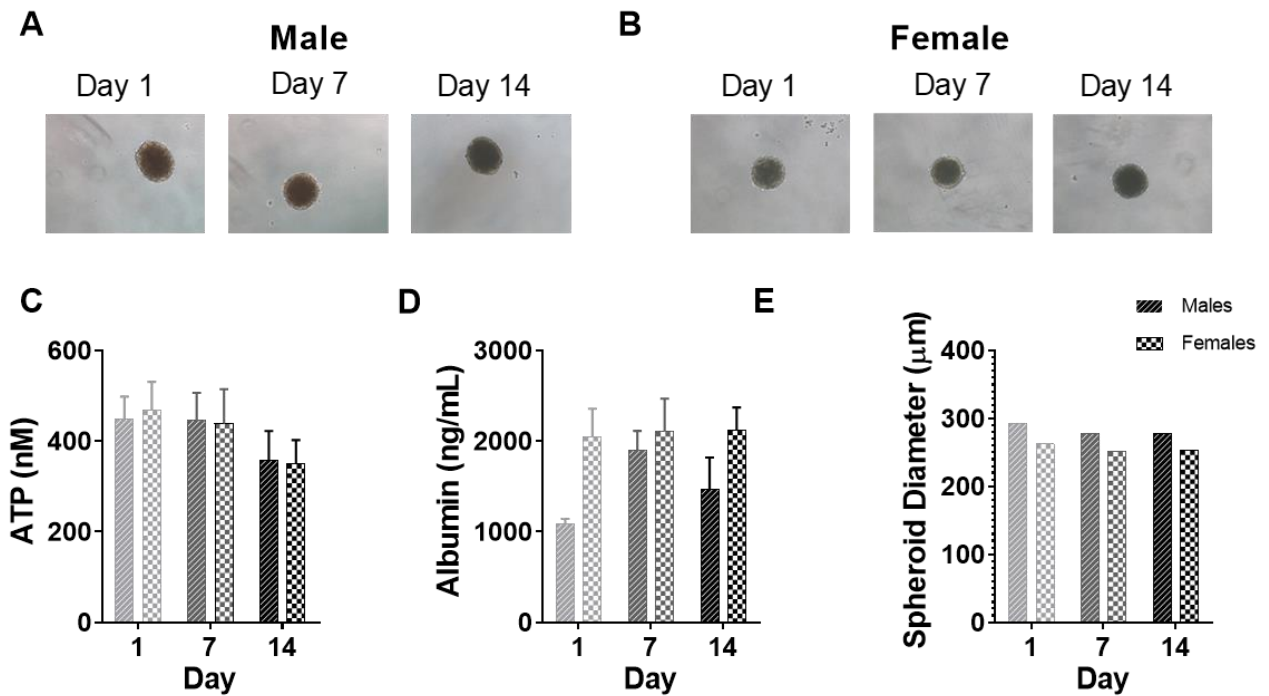
Supplementary Table 1

Key Drug Metabolizing Enzymes and Transporters Measured

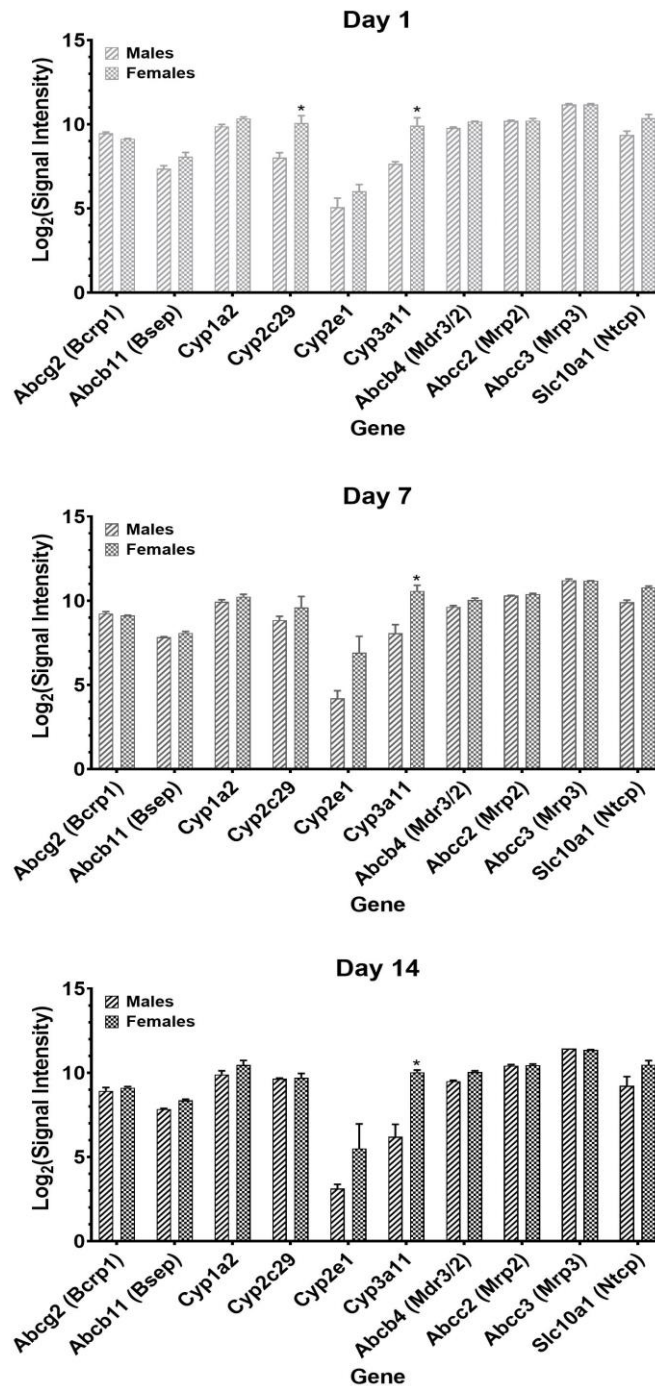
Human Gene Symbol	Mouse mRNA Symbol	Mouse mRNA Name	Mouse RefSeq mRNA ID	Mouse Probe Set ID	Mouse Protein Symbol	Mouse Protein Name	Mouse UniProt ID	Mouse Proteotypic Peptide Sequence
ABCG2	Abcg2	ATP-binding cassette sub-family G member 2	NM_011920	TC0600000711.mm.2	Bcrp1	ATP-binding cassette sub-family G member 2	Q7TMS5	SSLLDVLAARK
ABCB11	Abcb11	ATP-binding cassette, sub-family B (Mdr/Tap), member 11	NM_021022	TC0200003698.mm.2	Bsep	Bile salt export pump	Q9QY30	STALQLIQR
CYP1A2	Cyp1a2	Cytochrome P450 1a2	NM_009993	TC0900002379.mm.2	Cyp1a2	Cytochrome P450 1a2	P00186	NSIQDITSALFK
CYP2C9	Cyp2c29	Cytochrome P450 2c29	NM_007815	TC1900000565.mm.2	Cyp2c29	Cytochrome P450 2c29	Q64458	NISQSFTNFSK
CYP2E1	Cyp2e1	Cytochrome P450 2e1	NM_021282	TC0700002023.mm.2	Cyp2e1	Cytochrome P450 2e1	Q05421	GIIFNNGPTWK
CYP3A4	Cyp3a11	Cytochrome P450 3a11	NM_007818	TC0500003600.mm.2	Cyp3a11	Cytochrome P450 3a11	Q64459	LYPIANR
ABCB4	Abcb4	ATP-binding cassette, sub-family B (Mdr/Tap), member 4	NM_008830	TC0500000054.mm.2	Mdr3/2	Multidrug resistance protein 2	P21440	IATEAIENIR
ABCC2	Abcc2	ATP-binding cassette, sub-family C (Cftr/Mrp), member 2	NM_013806	TC1900000636.mm.2	Mrp2	Multispecific organic anion transporter 1	Q8VI47	EAGIESVNHTEL
ABCC3	Abcc3	ATP-binding cassette, sub-family C (Cftr/Mrp), member 3	NM_029600	TC1100003561.mm.2	Mrp3	Multispecific organic anion transporter 2	B2RX12	GALVAVVGPVGC GK
NTCP	Slc10a1	Solute carrier family 10 (sodium/bile acid cotransporter family), member 1	NM_001177561	TC1200002011.mm.2	Ntcp	Sodium/taurocholate cotransporting polypeptide	O08705	GIYDGDLLK



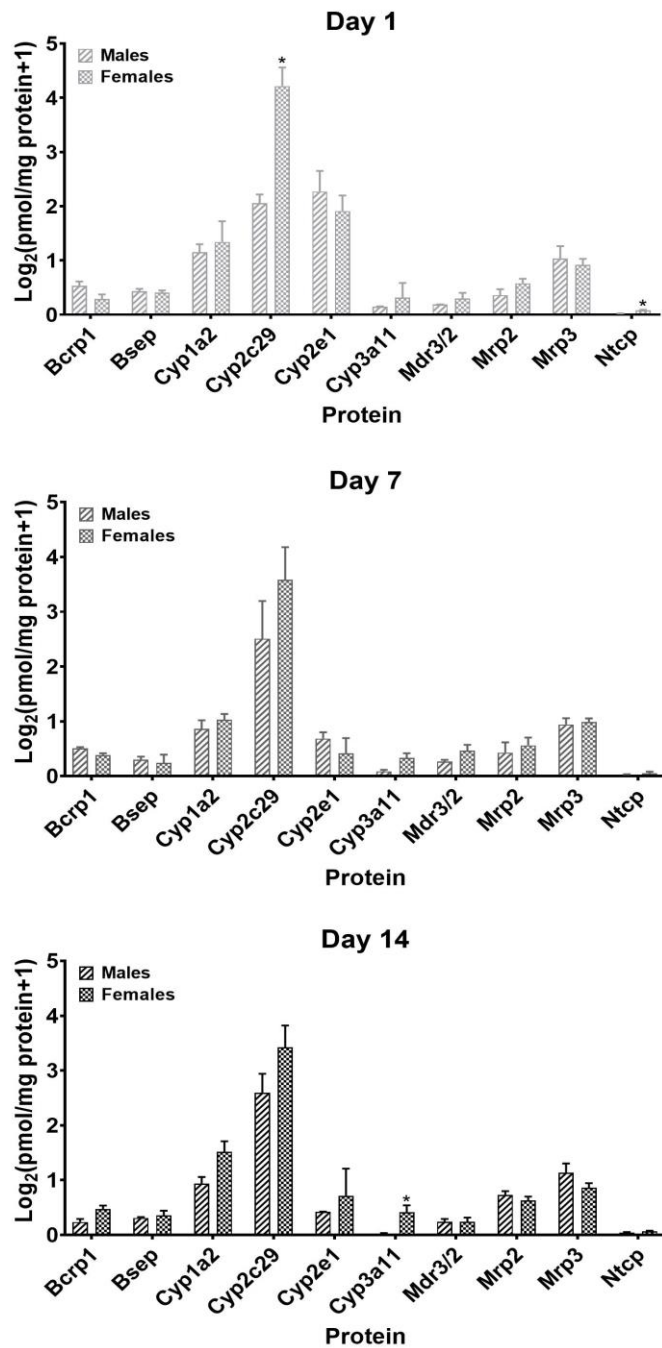
Supplementary Figure 1. Schematic of treatment regimens for measurement of (A) baseline viability, albumin and spheroid diameter, (B) cytochrome P450 activity, and (C) effect of drug exposure over time.



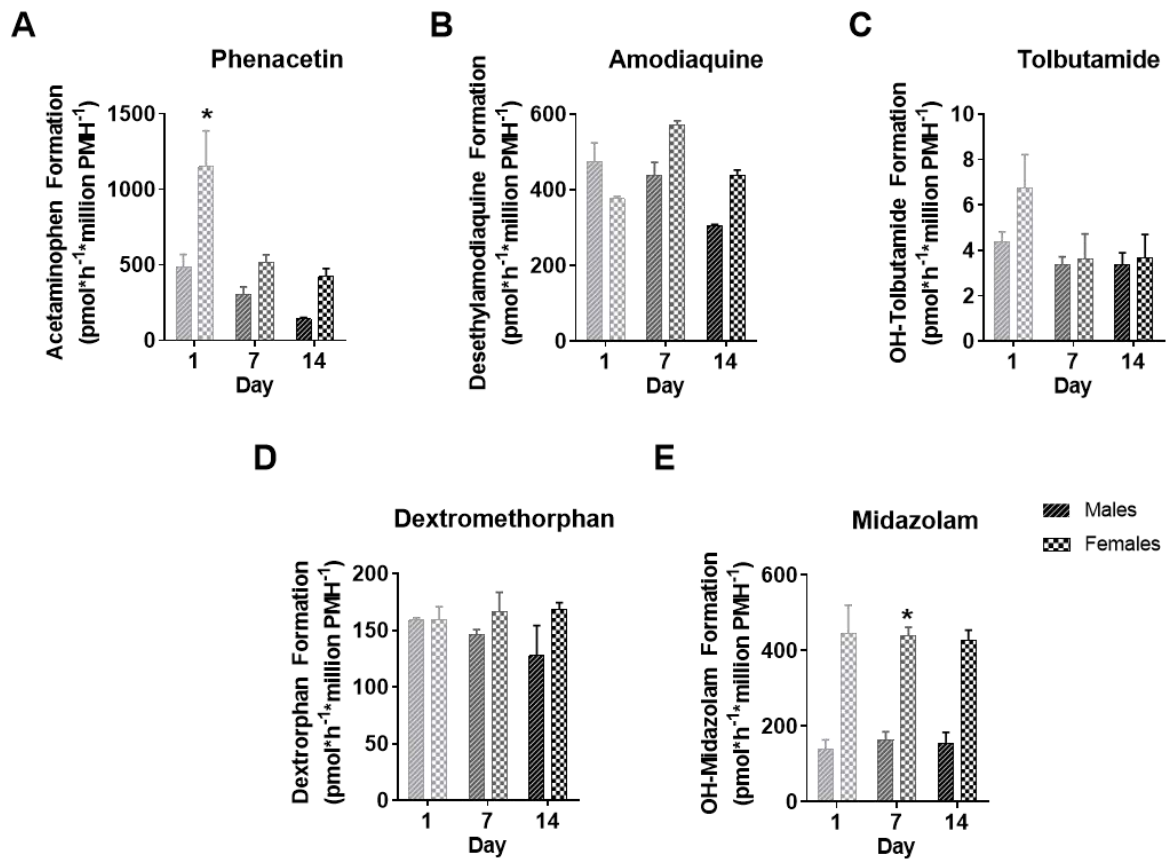
Supplementary Figure 2. Representative phase-contrast photomicrographs showing morphology of (A) male and (B) female spheroids at 10X magnification on Days 1, 7, and 14 post spheroid formation. (C) ATP, (D) albumin, and (E) spheroid diameter measured over time in males and females. Data represent the mean+SEM from N=3 preparations per sex for ATP and albumin, and N=1 preparation per sex for spheroid size. No significant differences were observed between sexes in baseline ATP, albumin, or spheroid diameter at any time point as determined by repeated measures ANOVA with Sidak's multiple comparison test comparing females to males on each day.



Supplementary Figure 3. Log₂(signal intensity) for 10 key drug metabolizing enzymes and transporter mRNAs in spheroid culture over time. Data represent the geometric mean+SD of N=2 male and N=2 female preparations. * indicates FDR $p < 0.05$ and IFCI > 2 as determined by one-way ANOVA with linear contrasts comparing sex within model.



Supplementary Figure 4. Log₂(pmol/mg protein+1) for 10 key drug metabolizing enzymes and transporter proteins in spheroid culture over time. Data represent the geometric mean+SD of N=3 male and N=3 female preparations. * indicates FDR $p < 0.05$ and IFCI > 2 as determined by one-way ANOVA with linear contrasts for sex within each time point.



Supplementary Figure 5. Metabolite levels measured after a 4-hour incubation with cytochrome P450 substrate cocktail. Data are presented as mean+SEM from N=2 preparations per sex. * indicates $p < 0.05$ as determined by repeated measures ANOVA with Sidak's multiple comparison test comparing females to males on each day.