Activation of Aryl Hydrocarbon Receptor Signaling in Human Trophoblasts Alters Markers of Growth and Differentiation

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Tables legends

Table 1: RNA-seq expression of multiple markers of the placenta and human trophoblast stem cells.

Table 2: IPA analysis of overlapping differentially altered transcripts between 25µg/ml and 40µg/ml CSC treatment.

Figure legends

Figure 1. Venn diagram displays the total number of differentially altered transcripts following CSC treatment.

Figure 2. IPA heatmap differentially regulated transcripts of AHR battery/target genes following CSC treatment

Figure 3. R Studio heatmap representation of CYP1A1 & CYP1B1 communities following CSC treatment.

Table 1

	Human Trophoblasts (25 µg/ml)		Human Trophoblasts (40 µg/ml)	
Gene	RNAseq mean count	RNAseq mean TPM	RNAseq mean count	RNAseq mean TPM
CDX1	9	6	1073	7
CDX2	0	0	6	0
EOMES	0	0	2	0
ESRRB	0	0	0	0
ELF5	1,091	7	1272	7
HAND1	184	1	183	1
ID2	587	8	544	8
HLA-G	2,079	17	2071	17
CGB	1,862.00	25	2120	30
GCM1	896	3	842	3
GATA3	4,519.00	19	4585	21
TFAP2C	5200	23	5517	25
KRT7	24,698.0	191	25,441.0	209
FOS	5266	50	5536	51
CDKN1C	9087	70	7529	63
PAPP-A	17	0	39	0
CTSC	5891	36	6542	43
FAP	197	1	234	2
SPARC	7514	76	7453	81
CAV1	967	6	1034	7
TIMP3	44739	98	47737	110
COL4A1	35730	55	35434	57
HIF2A (EPAS1)	12148	44	11238	45
FN1	7967	12	8521	13
GPC3	1202	6	1185	6
IGF2	3915	9	3946	10
TIMP2	10144	30	11055	34
BPGM	1148	7	1246	8
IGF1R	2958	9	2729	9
IL6ST	7100	22	7274	22
HBB	0	0	0	0
MUC1	28	0	32	1

Table 2

Treatment	25 µg/ml		40 µg/ml		Function
Gene	**TPM**	**log2FC**	**TPM**	**log2FC**	
CYP1B1	9475.80	2.894	9,319.52	3.07	Moxygenases which catalyze many reactions involved in drug metabolism and synthesis of cholesterol, steroids. Upregulated in Pre-Eclampsia (UPE)
CYP1A1	126076.1	2.73	135,133.6	2.82	
AMER2	159.298	2.327	162.834	2.432	Negative regulator of the canonical Wnt signaling. Canonical Wingless (Wnt) signaling play a crucial role in the invasive differentiation of trophoblasts
SERPINB2	73.6694	1.068	66.429	1.731	A TGF-β-responsive gene that plays a role in differentiation and regulated by ZFP36. UPE
POU2F3	245.387	1.465	298.320	1.587	A TF that regulates cell type-specific differentiation pathways (keratinocyte functions)
VIPR1	381.929	1.924	384.518	1.553	VIP contributes to smooth muscle relaxation favoring uterus quiescence. VIP act as a homeostatic mechanism during the early stages at the maternal-placental interface to control exacerbated inflammatory responses such as the ones observed in intrauterine infections
STRA6	1359.58	1.304	1464.334	1.469	Functions as a surface signaling receptor by which retinol regulates insulin responses
MT1H	351.317	0.793	416.506	1.366	Encodes a metal-binding protein that protects cells from oxidative stress. UPE
ALOX15B	110.126	0.771	119.411	1.285	Encodes enzyme oxidizes polyunsaturated fatty acids. UPE
TIPARP	2790.88	1.243	2842.666	1.275	A negative regulator of AHR
IFIT3	110.126	-1.749	80.523	-3.209	Acts as an inhibitor of cellular as well as viral processes, cell migration, proliferation, signaling, and viral replication. Associated with Preeclampsia (APE)
ACTA1	658.753	-2.003	840.1584	-2.695	Plays a role in cell motility, structure, and integrity
IFIT1	110.589	-1.692	103.1941	-2.687	Increased levels in pregnancy implicated a role in the endometrial pathways critical for uterine support of peri-implantation conceptus survival, growth
RSAD2	75.2674	-1.906	67.8999	-2.144	Plays a role in the implantation stage of pregnancy by preventing viral infection
TAGLN	10065.5	-1.593	12735.062	-2.088	Participate in the process of cell migration through regulation of MMP9 expression and MLC phosphorylation
NPPB	2121.29	-1.501	2713.429	-1.949	May play a protective role in counterbalancing the effects of postpartum cardiac hypertrophy
SPAG17	142.149	-1.102	121.210	-1.74	Gene Knockout in mice is associated with male infertility
PRICKLE1	132.415	-1.542	150.763	-1.73	Encodes a nuclear receptor is a negative regulator of the Wnt/beta-catenin signaling
FGF1	633.509	-1.183	691.796	-1.56	Play a role in luteal cell proliferation during early pregnancy and may thereby contribute to the maintenance of luteal function
UNC13C	104.501	-1.715	89.623	-1.501	May play a role in vesicle maturation during exocytosis as a target of the diacylglycerol second messenger pathway
TGFB2	86.8603	-1.825	72.387	-1.428	Involved in cell proliferation. Disruption of the TGF-beta/SMAD pathway has been implicated in a variety of human cancers. Regulated by TF: ATF1, 2 and USF2

Figure 1.

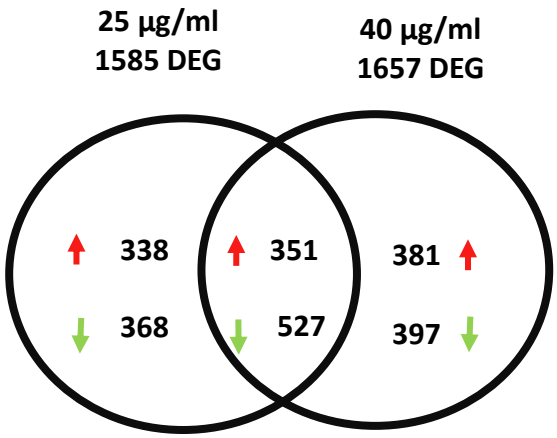


Figure 2.

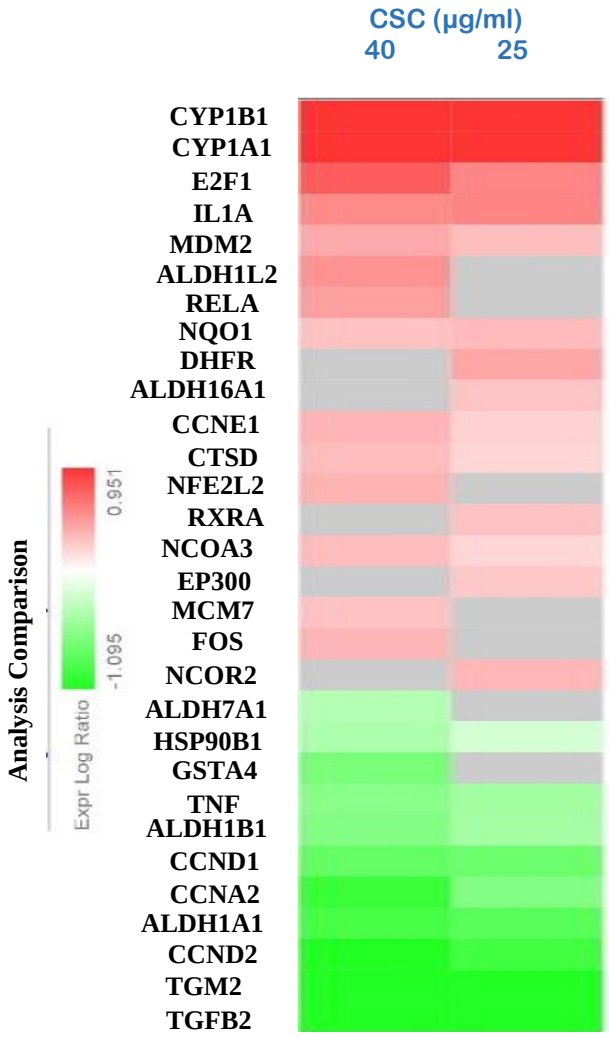
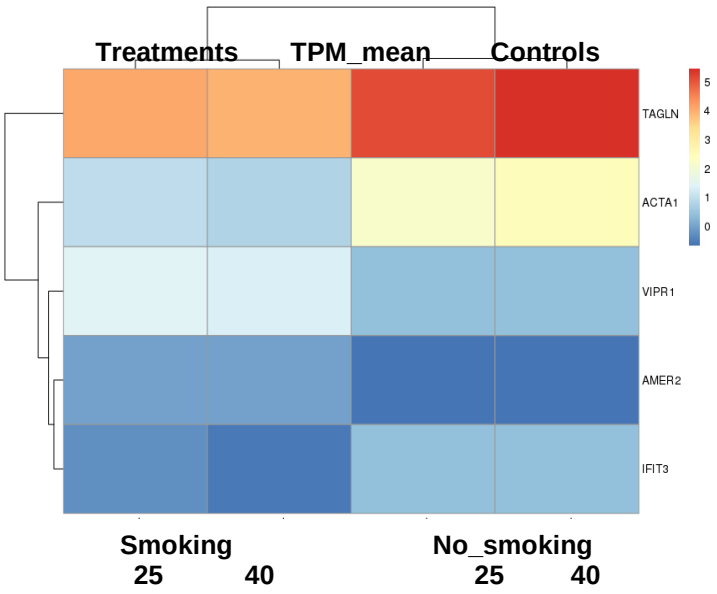
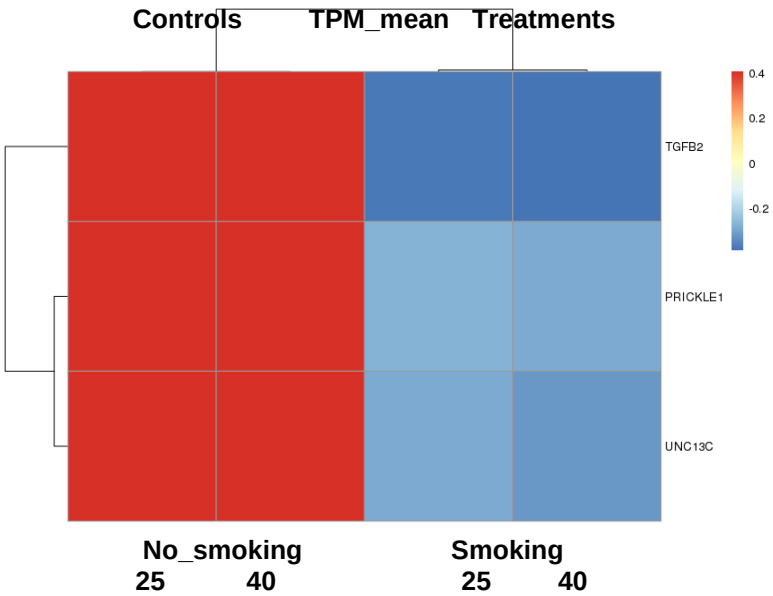

CSC (µg/ml)
40
25
CYP1B1
CYP1A1
E2F1
IL1A
MDM2
ALDH1L2
RELA
NQO1
DHFR
ALDH16A1
CCNE1
CTSD
NFE2L2
RXRA
NCOA3
EP300
MCM7
FOS
NCOR2
ALDH7A1
HSP90B1
GSTA4
TNF
ALDH1B1
CCND1
CCNA2
ALDH1A1
CCND2
TGM2
TGFB2
Analysis Comparison
0.951
-1.095
Expr Log Ratio

Figure 3.

A.


Treatments
TPM_mean
Controls
TAGLN
ACTA1
VIPR1
AMER2
IFIT3
5
4
3
2
1
0
Smoking
25
40
No_smoking
25
40

B.


Controls
TPM_mean
Treatments
TGFB2
PRICKLE1
UNC13C
0.4
0.2
0
-0.2
No_smoking
25
40
Smoking
25
40