

Sarah Dubaisi, Hailin Fang, Joseph A. Caruso, Roger Gaedigk, Carrie A. Vyhldal, Thomas A. Kocarek, and Melissa Runge-Morris. Developmental Expression of *SULT1C4* Transcript Variants in Human Liver: Implications for Discordance Between *SULT1C4* mRNA and Protein Levels. *Drug Metabolism and Disposition*

**Supplemental Table 1. Primers used in the 5'-RACE and RT-qPCR analysis**

Primer Name	Sequence
RACE Reverse	5'- <b>GAT TAC GCC AAG CTT</b> <sup>1</sup> AGC AGG TCA TCA GGC TTG GCT TGG A-3'
<i>SULT1C4</i> -forward <sup>2</sup>	5'- <b>CTC GCT AGC CTC GAG</b> <sup>1</sup> TAG AGG GCT GGA TAG TGT GGT AGT G-3'
<i>SULT1C4</i> reverse <sup>2</sup>	5'- <b>CTT GAT ATC CTC GAG</b> <sup>1</sup> ATT GAG CTT CCC TCT CTC CAT GTC-3'
<i>SULT1C4</i> -TVs forward <sup>3</sup>	5'-CCT ATC CTA AAG CAG GAA CAA CA-3'
<i>SULT1C4</i> -TV1 reverse <sup>3</sup>	5'-ATG AGC TTG TTC CAA ACC AG-3'
<i>SULT1C4</i> -TV2 reverse <sup>3</sup>	5'-CAG GAG CCC CAG CAC ACA G-3'
<i>SULT1C4</i> -E3DEL reverse <sup>3</sup>	5'-GGA TTT CTT GCT ACA TAG ATT ATC AG-3'
<i>SULT1C4</i> -HindIII-DDK-F <sup>4</sup>	5'-GCG AAG CTT GCC ACC ATG <u>GAT TAC AAG GAT GAC GAC GAT AAG</u> ATG GCC TTA CAC GAC ATG G-3'
<i>SULT1C4</i> -XhoI-R <sup>4</sup>	5'-GCG CTC GAG TCA GAA CTG GAA GTG GAA AGT TAG TCT G-3'

<sup>1</sup> Sequences in bold overlap with the plasmid sequence; used for In-Fusion cloning of the DNA fragments

<sup>2</sup> Primers used to amplify *SULT1C4* TVs by standard RT-PCR; predicted to amplify an approximately 1.2 Kb fragment from the *SULT1C4* TV1 sequence

<sup>3</sup> Primers used for RT-qPCR to measure the individual *SULT1C4* TVs in human liver specimens

<sup>4</sup> Primers used to prepare the DDK-tagged expression plasmids containing the *SULT1C4* TVs. The underscored sequence of the forward primer encodes the DDK tag.

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**Supplemental Table 2. 5'-Positions of 5'-RACE clones relative to SULT1C4 gene**

Sequence	Position of 5'-end on NC_000003.12 (GRCh38.p13)	Position of 5'-end relative to ATG <sup>2</sup>	Inr? <sup>1</sup>
<b>Published Sequences</b>			
SULT1C4 Gene, NM_006588 (TV1), and NM_001321770 (TV2)	108,377,954	-384	
<b>Caco-2 5'-RACE Clones</b>			
1	108,377,958	-380	
2	108,378,021	-317	Y
3	108,377,954	-384	
4	108,377,985	-353	Y
5	108,377,957	-381	Y
6	108,377,954	-384	
7	108,377,938	-400	Y
8	108,377,954	-384	
9	108,378,000	-338	
10	108,377,954	-384	
<b>HepaRG 5'-RACE Clones</b>			
11	108,377,985	-353	Y
12	108,377,648	-690	
13	108,377,970	-368	Y
14	108,377,985	-353	Y
15	108,377,970	-368	Y
16	108,377,966	-372	
17	108,377,970	-368	Y
18	108,377,985	-353	Y
19	108,377,878	-460	
20	108,377,983	-355	
21	108,377,993	-345	
22	108,377,793	-545	
23	108,377,985	-353	Y

<sup>1</sup>Inr?: The 5'-ends of clones marked with "Y" are located within predicted consensus initiator elements. The consensus sequence of the human Inr is BBCA<sub>+1</sub>BW, according to a recent report (Vo ngoc et al, 2017). B=C or G or T; W=A or T

<sup>2</sup>ATG, Translation start site

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**Supplemental Table 3. Median amounts of the three SULT1C4 TVs measured by RT-qPCR**

	SULT1C4 TV1	TV2	E3DEL
Prenatal	7.64	43.6	2.72
Infant	0.379	8.46	0.177
Adult	0.162	4.07	0.109

**Supplemental Table 4. Average of raw transcript per million (TPM) values of the three SULT1C4 TVs that were detected with RNA-seq**

	SULT1C4 TV1 ENST00000272452	SULT1C4 TV2 ENST00000409309	Non-coding SULT1C4 ENST00000494122
Prenatal	1.05	2.29	1.21
Infant	0.311	0.406	0.230
1-2 years old	0.0833	0.0967	0.0733
2-5 years old	0.0727	0.179	0.0964
6-11 years old	0.0592	0.130	0.0985
12-18 years old	0.0927	0.143	0.0536

**Supplemental Table 5. Measurement of SULT1C4 TV1- or TV2-derived protein levels by quantitative proteomics in transfected HEK293 cells**

Sample	Amount of SULT1C4 Protein (fmol/mg)
EV	<LD
TV1	5.31 ± 0.27 <sup>1</sup>
TV2	<LD

<sup>1</sup>Mean ± SD

EV, Empty vector transfected; <LD, below limit of detection