

Supplemental Figures and Tables

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TITLE: The role of L- and D-menthol in the glucuronidation and detoxification of the major lung carcinogen, NNAL

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Table S1. Complete screening of menthol and NNAL for individual UGT enzymes.

	L-MG	D-MG	NNAL-N-Gluc ^a	(R)-NNAL-O-Gluc ^b	(S)-NNAL-O-Gluc ^b
UGT1A1	- ^c	-	-	-	-
UGT1A3	-	-	-	-	-
UGT1A4	-	-	+ ^c	-	-
UGT1A5	-	-	-	-	-
UGT1A6	-	-	-	-	-
UGT1A7	+	-	-	-	-
UGT1A8	-	-	-	-	-
UGT1A9	+	+	-	+	+
UGT1A10	+	+	-	+	-
UGT2A1	+	+	-	-	-
UGT2A2	+	+	-	-	-
UGT2A3	+	+	-	-	-
UGT2B4	+	+	-	-	-
UGT2B7	+	+	-	-	+
UGT2B10	-	-	+	-	-
UGT2B11	-	-	-	-	-
UGT2B17	+	+	-	+	-

^a Data adapted from Chen et al, 2008.

^b Data adapted from Kozlovich et al, 2015.

^c -, no formation of the indicated metabolite detected with the corresponding UGT; +, formation of the indicated metabolite detected with the corresponding UGT.

Table S2. Tertile values of each NNK metabolite.^a

	Low	Intermediate	High	P value
Total MG/mg creatinine	0.93 ± 0.14 ^b	6.4 ± 0.43	34 ± 8.1	
NNAL-N-Gluc/free NNAL	2.0 ± 0.18	1.6 ± 0.13	1.7 ± 0.15	ANOVA, p=0.01 Low:Intermediate, p=0.01 Low:High, p=0.01
R-NNAL-O-Gluc/free NNAL	1.3 ± 0.17	1.1 ± 0.13	1.2 ± 0.15	ANOVA, 0.8 Low:Intermediate, 0.9 Low:High, 0.9
S-NNAL-O-Gluc/free NNAL	5.1 ± 0.59	3.8 ± 0.36	4.9 ± 0.74	ANOVA, 0.3 Low:Intermediate, 0.3 Low:High, 0.9
free NNAL/mg creatinine	0.16 ± 0.018	0.28 ± 0.17	0.12 ± 0.020	ANOVA, 0.07 Low:Intermediate, 0.17 Low:High, 0.11 Intermediate:High, 0.9

^a Tertiles shown are subjects stratified by urinary MG levels. Data were used in Figure 3.

^b Data are expressed as the mean ± SE.

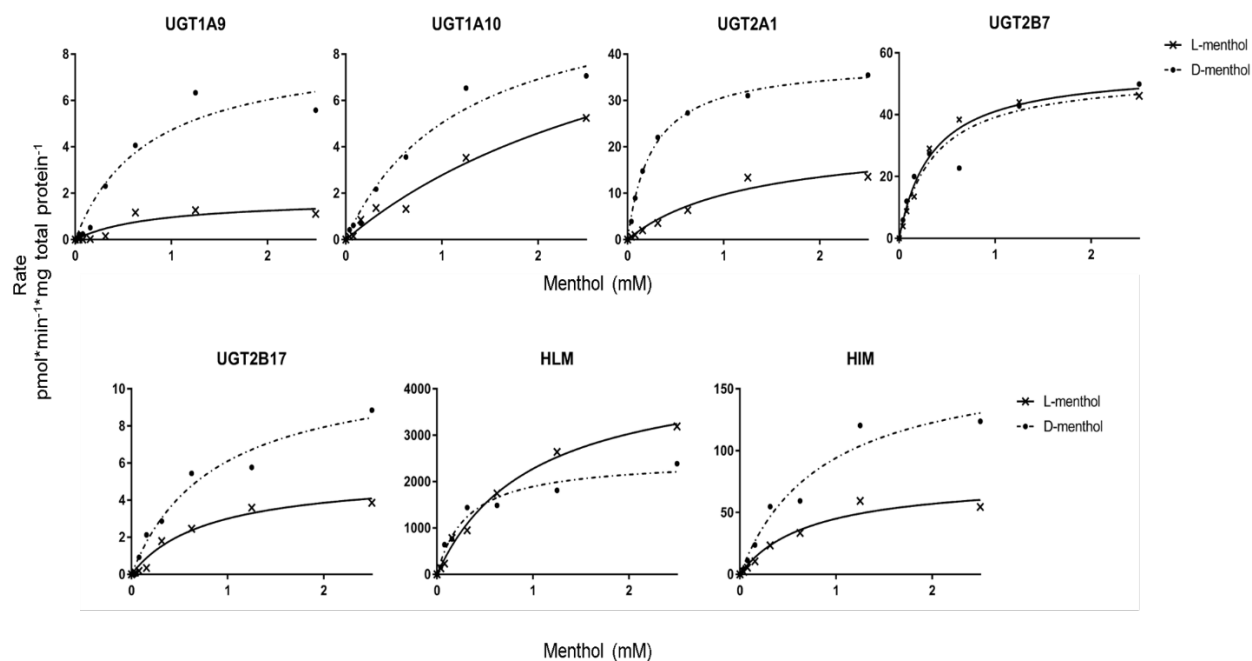


Figure S1. Representative kinetic curves for the formation of L- and D-menthol glucuronides. Kinetic assays for L-menthol (x) and D-menthol (•) were performed by incubating each menthol enantiomer (1.0 μ M – 2.5 mM) with HEK293 UGT-overexpressing cell microsomes. Assays were incubated at 37°C for 30 min. Shown is a representative experiment for each UGT, human liver microsome (HLM) or human intestinal microsomes (HIM); all experiments were performed in triplicate.