Supplemental data

Manuscript title:

Differences of the *in vivo* and *in vitro* metabolism of imrecoxib in humans: formation of rate-limiting aldehyde intermediate

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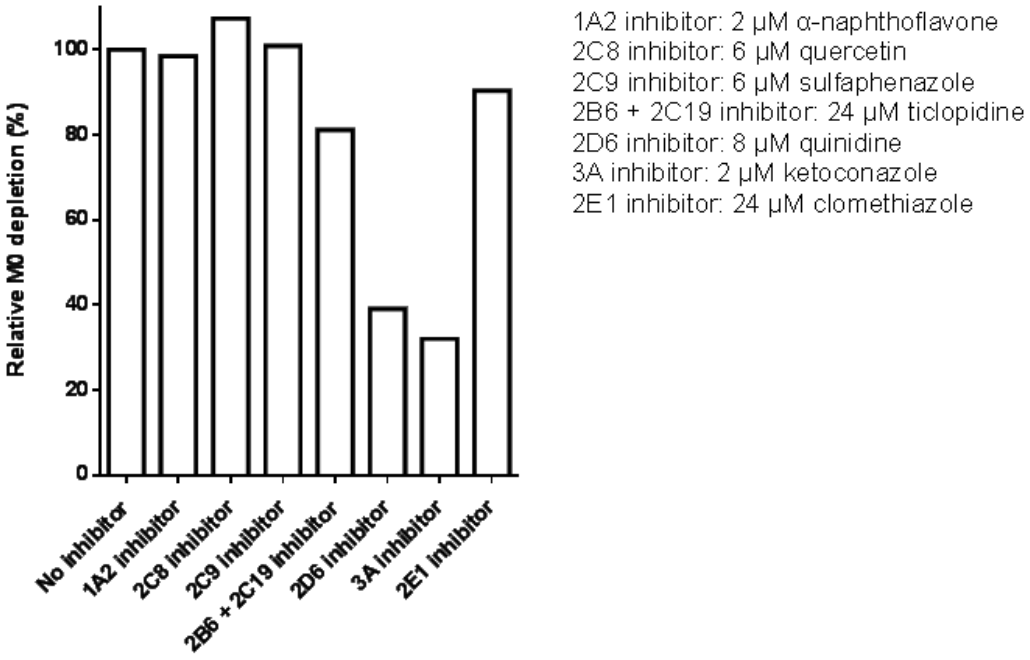
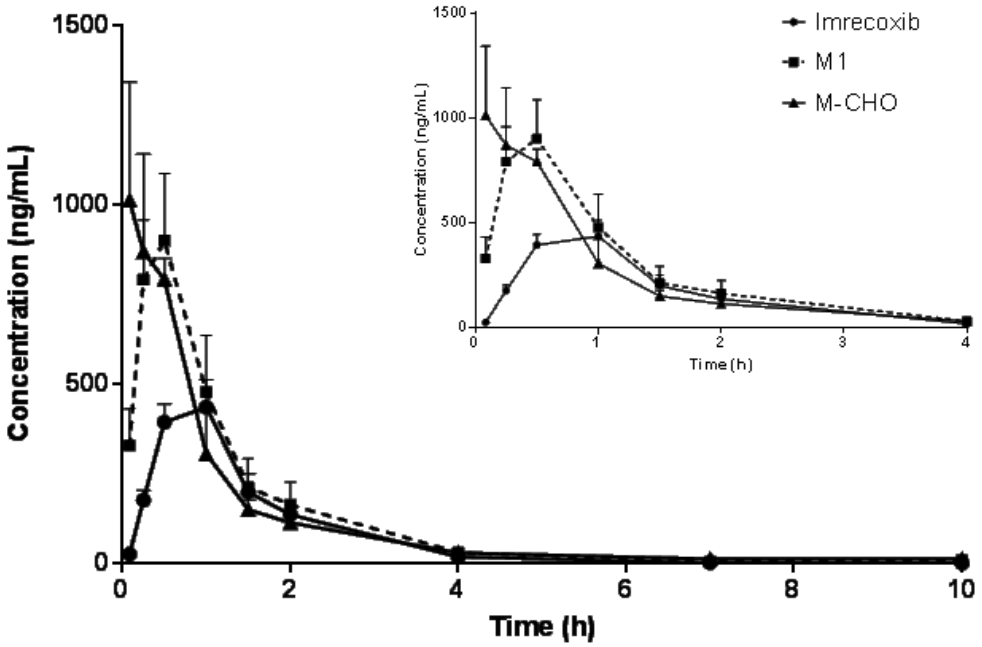
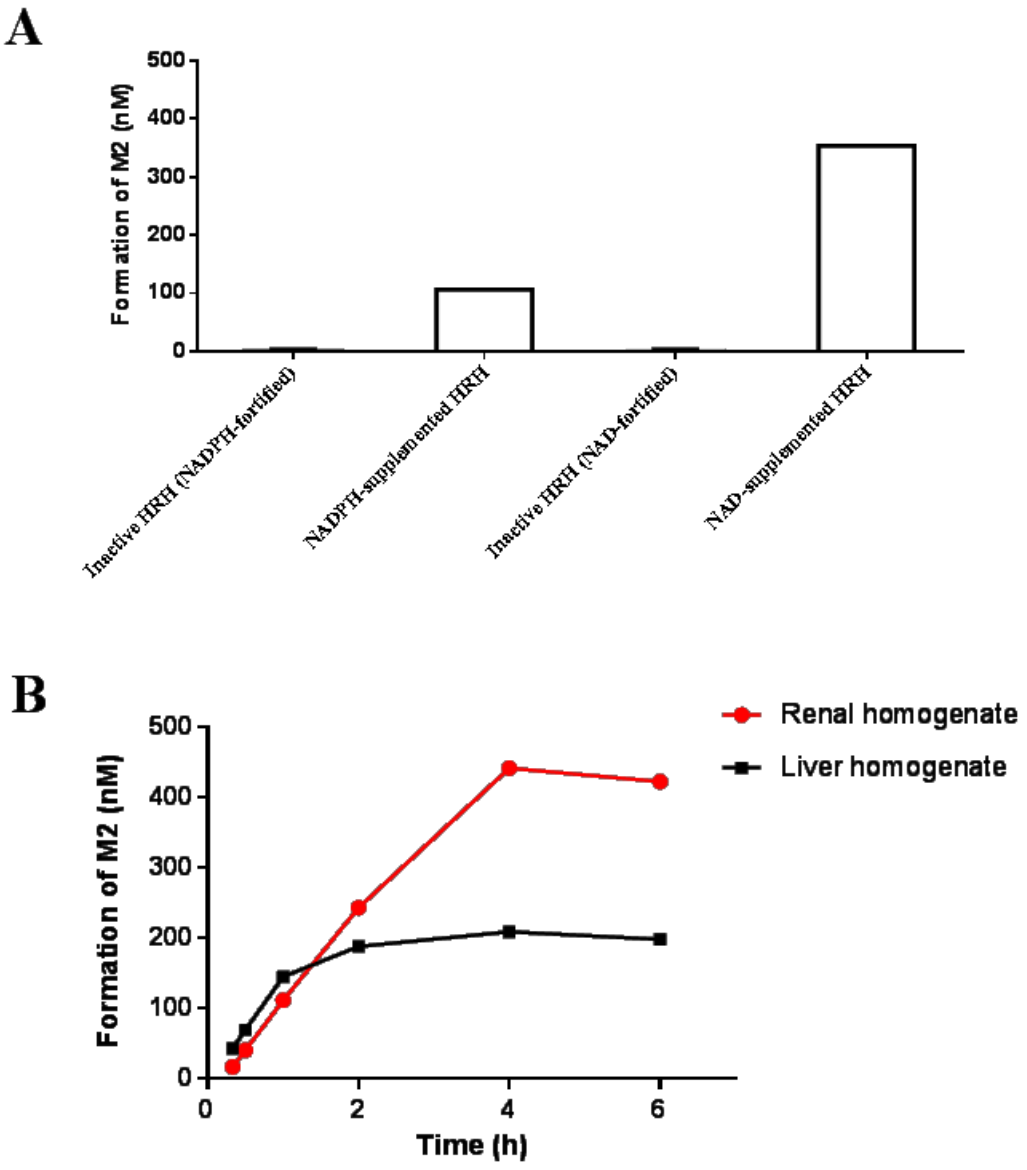
Supplemental Figure 1: Effect of specific CYP inhibitors on M0 depletion in HLM incubations containing imrecoxib, NADPH, and individual inhibitor. The control is normalized to 100%.



Supplemental Figure 2: Mean plasma concentration-time curves for M2 after a single intravenous administration of 5 mg/kg imrecoxib, M1, and M-CHO, respectively to rats (n = 5/group).

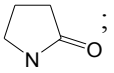


Supplemental Figure 3: Incubations of M1 (10 μM) in NADPH- or NAD-supplemented human renal homogenate (HRH, unknown concentration) (A), and in freshly prepared rat renal and liver homogenate (unknown concentration) for 0.33, 0.5, 1.0, 2.0, 4.0, 6.0 h (B). Each data is displayed as the mean of two separate samples.



Supplemental Table 1: Characterization of imrecoxib metabolites in humans using UPLC/Q-TOF MS.

Metabolite	Proposed Structure			Retention time (min)	Formula	m/z	ppm	Major product ions
	R1	R2	R3					
M0	CH_3	$CH_2CH_2CH_3$	H	11.8	$C_{21}H_{23}NO_3S$	370.1461	-2.9	278.0821, 236.0353, 157.0501, 129.0552
M1[a]	CH_2OH	$CH_2CH_2CH_3$	H	9.97	$C_{21}H_{23}NO_4S$	386.1414	-1.8	386.1405, 356.1303, 326.0836, 278.0837
M2[a]	COOH	$CH_2CH_2CH_3$	H	10.76	$C_{21}H_{21}NO_5S$	400.1206	-1.9	382.1104, 358.0739, 340.0634, 321.1355
M3	CH_2OH	H	O	5.81	$C_{18}H_{15}NO_5S$	358.0742	-0.6	340.0636, 236.0369, 157.0497
M4	CH_2OH	H	H	5.53	$C_{18}H_{17}NO_4S$	344.0946	-1.3	326.0833, 236.0371
M5	CH_3	$CH_2CH_2CH_3$	OH	10.82	$C_{21}H_{23}NO_4S$	386.1401	-5.0	368.1271, 340.1378, 283.0757, 206.1054
M6[b]	CH_3	$CH_2CH_2CH_3$	O	10.56	$C_{21}H_{23}NO_4S$	386.1420	-0.3	368.1460, 326.0905, 283.0790
M7	CH_3	$CH_2CHOHCH_3$	H	10.4	$C_{21}H_{23}NO_4S$	386.1424	0.9	368.1386, 326.0728, 299.0645
M8[c]	CHO	$CH_2CH_2CH_3$	H	N.A.	$C_{21}H_{21}NO_4S$	N.A.	N.A.	N.A.
M9	CH_2OH	$CH_2CH_2CH_3$	O	6.56	$C_{21}H_{21}NO_5S$	400.1215	0.5	370.1125, 343.0928, 354.1150
M10	CH_2OH	$CH_2CH_2CH_3$	O	11.25	$C_{21}H_{21}NO_5S$	400.1210	-0.9	358.0788, 250.0783, 236.0834
M11[b]	CH_2OH	$CH_2CH_2CH_3$	O	7.81	$C_{21}H_{23}NO_5S$	402.1366	-0.8	384.1250, 356.1354, 309.0523
M12	CH_3	$CH_2CHOHCH_3$	OH	6.15	$C_{21}H_{23}NO_5S$	402.1368	-0.4	384.1245, 356.0933
M13	CH_2OH	$CH_2CH_2CH_3$	OH	6.32	$C_{21}H_{23}NO_5S$	402.1368	-0.3	384.1261, 192.0922
M14	CH_2OH	$CH_2CHOHCH_3$	H	8.14	$C_{21}H_{23}NO_5S$	402.1369	-0.1	384.1259, 343.0647, 271.0776, 183.0107
M15	COOH	$CH_2CHOHCH_3$	H	8.95	$C_{21}H_{21}NO_6S$	416.1152	-2.5	398.1051, 357.0433, 183.0089
M16	CH_2OH	$CH_2CHOHCH_3$	O	8.45	$C_{21}H_{21}NO_6S$	416.1155	-1.8	398.1033, 370.1076, 313.0530, 297.0557
M17				6.86	$C_{21}H_{21}NO_6S$	416.1159	-0.7	398.1055, 370.1106, 297.0581, 236.0828
M18	CH_2OH	CH_2COCH_3	OH	6.64	$C_{21}H_{21}NO_6S$	416.1162	-0.1	398.1049, 370.0716
M19[b]	CH_2OH	CH_2COCH_3	O	6.19	$C_{21}H_{21}NO_6S$	416.1163	0.1	356.0952
M20	$CH_2OC_6H_9O_6$	$CH_2CH_2CH_3$	H	9.05	$C_{27}H_{31}NO_{10}S$	562.1727	-2.5	386.1419, 368.1310
M21	CH_3	$CH_2CHOC_6H_9O_6CH_3$	H	10.92	$C_{27}H_{31}NO_{10}S$	562.1735	-1.2	386.2890, 368.1298
M22	$COOC_6H_9O_6$	$CH_2CH_2CH_3$	H	9.57	$C_{27}H_{29}NO_{11}S$	576.1520	-2.5	400.1206

a. Confirmed using references; b. Nuclear structure is  ;

c. Reactive metabolite confirmed by trapping experiment with methoxyamine.

Supplemental Table 2: Pharmacokinetic parameters of imrecoxib and its two active metabolites after an oral administration of 100 mg imrecoxib to healthy volunteers (*n* = 10). Each data presents as mean ± S.D., except for T_{max} [median (range)]. S.D., standard deviation.

Parameter	Unit	Imrecoxib	M1	M2
AUC (0-t)	ng/mL*h	429 ± 387	397 ± 168	1521 ± 634
AUC (0-∞)	ng/mL*h	435 ± 389	411 ± 172	1657 ± 777
C_{max}	ng/mL	48.6 ± 35.4	50.9 ± 16.1	205 ± 83.6
T_{max}	h	1.0 (0.5 - 2.0)	1.0 (0.5 - 3.0)	2.0 (1.0 - 3.0)
$t_{1/2}$	h	10.5 ± 3.71	11.2 ± 7.12	12.5 ± 8.40