

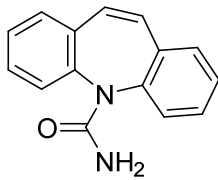
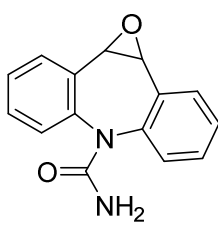
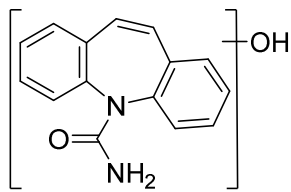
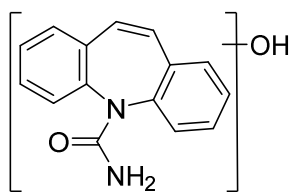
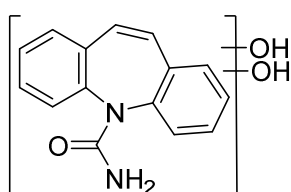
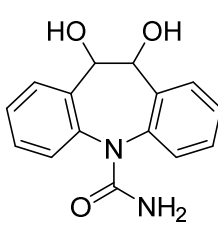
Analytical and Bioanalytical Chemistry

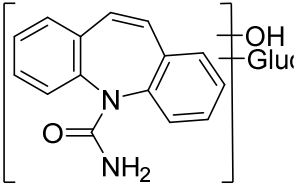
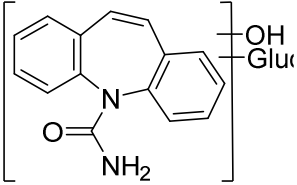
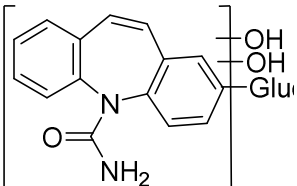
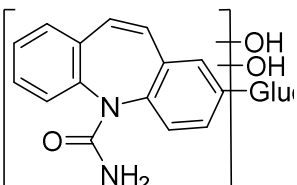
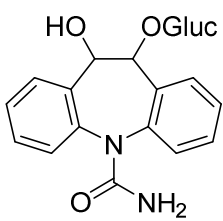
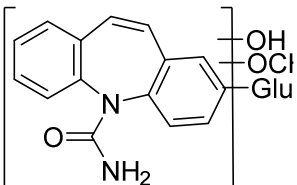
Electronic Supplementary Material

Metabolite profiling of carbamazepine and ibuprofen in *Solea senegalensis* bile using high-resolution mass spectrometry

Jaume Aceña, Sandra Pérez, Peter Eichhorn, Montse Solé, Damià Barceló

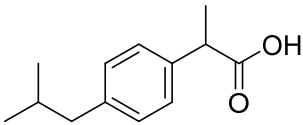
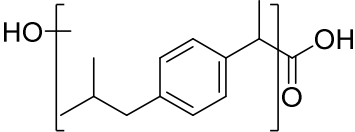
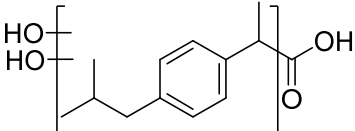
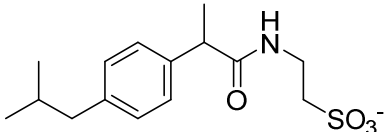
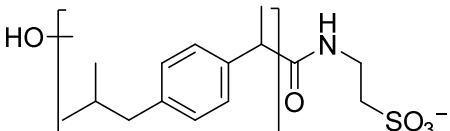
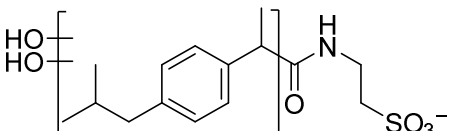
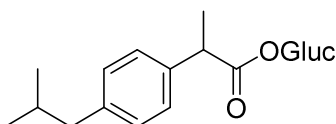
Table S1 Detection and relative quantification of CBZ and its metabolites in bile

Compound	t _R (min)	m/z of [M-H] ⁻	Theor. m/z	Rel. error (ppm)	Formula [M-H] ⁻	Rel. area at 15°C	Rel. area at 20°C
Carbamazepine	4.42	237.1022	237.1022	0.0	C ₁₅ H ₁₃ N ₂ O	68.32	74.81
							
CBZ-M252-A	3.25	253.0972	253.0972	0.0	C ₁₅ H ₁₃ N ₂ O ₂	0.87	0.97
							
CBZ-M252-B	3.60**	253.0973	253.0972	0.4	C ₁₅ H ₁₃ N ₂ O ₂	0.18	0.21
							
CBZ-M252-C	3.90**	253.0974	253.0972	0.8	C ₁₅ H ₁₃ N ₂ O ₂	2.09	1.72
							
CBZ-M268	3.35	269.0921	269.0921	0.0	C ₁₅ H ₁₃ N ₂ O ₃	0.20	0.11
							
CBZ-M270	3.17	271.1075	271.1077	-0.7	C ₁₅ H ₁₅ N ₂ O ₃	0.09	0.10
							
CBZ-M428-A	2.79	429.1291	429.1292	-0.2	C ₂₁ H ₂₁ N ₂ O	1.72	1.42

	8						
CBZ-M428-B	3.03	429.1293	429.1292	0.2	C ₂₁ H ₂₁ N ₂ O	25.90	20.27
	8						
CBZ-M444-A	2.73	445.1245	445.1242	0.7	C ₂₁ H ₂₁ N ₂ O	0.38	0.22
	9						
CBZ-M444-B	3.00	445.1243	445.1242	0.2	C ₂₁ H ₂₁ N ₂ O	0.12	0.09
	9						
CBZ-M446	2.60	447.1400	447.1398	0.4	C ₂₁ H ₂₃ N ₂ O	0.06	0.03
	9						
CBZ-M458	2.80	459.1400	459.1398	0.4	C ₂₂ H ₂₃ N ₂ O	0.09	0.05
	9						

*First detection in fish bile; **confirmed by authentic standard

Table S2 Detection and relative quantification of IBU and their metabolites in bile

Compound	t _R (min)	m/z of [M-H] ⁻	Theor. m/z	Rel. error (ppm)	Formula [M-H] ⁻	Rel. area at 15°C	Rel. area at 20°C
IBU 	6.71	205.1230	205.1234	-2.0	C ₁₃ H ₁₇ O ₂	0.72	0.68
IBU-M222 	3.81***	221.1181	221.1183	-0.9	C ₁₃ H ₁₇ O ₃	1.25	1.23
IBU-M238* 	2.81	237.1130	237.1132	-0.8	C ₁₃ H ₁₇ O ₄	0.04	0.04
IBU-M313 	6.64	312.1279	312.1275	1.3	C ₁₅ H ₂₂ NO ₄ S	2.84	3.23
IBU-M329 	4.28	328.1228	328.1224	1.2	C ₁₅ H ₂₂ NO ₅ S	41.13	40.55
IBU-M345* 	3.28	344.1174	344.1173	0.3	C ₁₅ H ₂₂ NO ₆ S	3.19	4.09
IBU-M382 	6.31- 6.95***	381.1558	381.1555	0.8	C ₁₉ H ₂₅ O ₈	5.38	5.32
IBU-M398-A (e)	2.37	397.1504	397.1504	0.0	C ₁₉ H ₂₅ O ₉	0.54	0.67

	IBU-M398-B (e)	2.71	397.1506	397.1504	0.5	C ₁₉ H ₂₅ O ₉	0.43	0.65	
	IBU-M398-C (a)	4.29-	397.1506	397.1504	0.5	C ₁₉ H ₂₅ O ₉	41.13	40.55	
	IBU-M414-A (e)*	4.52**	1.92	413.1455	413.1453	0.5	C ₁₉ H ₂₅ O ₁₀	0.43	0.36
	IBU-M414-B (a)*	3.29-	413.1456	413.1453	0.7	C ₁₉ H ₂₅ O ₁₀	2.64	2.29	
	IBU-M505-A	3.38**	2.74	504.1544	504.1545	-0.2	C ₂₁ H ₃₀ NO ₁₁ S	0.05	0.04
	IBU-M505-B	3.05	504.1545	504.1545	0.0	C ₂₁ H ₃₀ NO ₁₁ S	0.23	0.28	

*First detection in fish bile; **unresolved peak cluster; ***confirmed by authentic standard

Table S3 Fish samples characteristics

	Temperature	Code	Total Weight (g)	Total Length (cm)	Sex	Liver Weight (g)	Gonad Weight (g)
Carrier	15°C	100	239	25.5	Female	2.175	-
	15°C	101	292	25.5	Female	2.725	3.292
	15°C	102	302	27.5	Female	2.640	2.870
	15°C	103	231	24.5	Female	2.253	2.447
	15°C	104	211	25	Female	2.356	2.735
	15°C	105	241	24	Male	2.380	0.148
	20°C	106	301	25.5	Male	2.563	0.172
	20°C	107	276	25	Female	1.984	3.314
	20°C	108	226	24.5	Female	1.834	2.922
	20°C	109	222	24	Male	1.774	0.183
	20°C	110	279	27	Male	2.133	0.117
	20°C	111	225	23.5	Male	2.049	0.145
Ibuprofen	15°C	200	238	25.5	Male	2.332	0.140
	15°C	201	255	26	Male	2.137	0.221
	15°C	202	328	26.5	Female	3.034	3.785
	15°C	203	290	27	Female	2.681	2.850
	15°C	204	259	26.5	Female	2.193	2.828
	15°C	205	272	26	Female	2.236	2.805
	20°C	206	234	26.5	Male	1.513	0.070
	20°C	207	290	28.5	Male	2.546	0.076
	20°C	208	223	24.5	Male	1.415	0.061
	20°C	209	271	27.5	Male	2.756	0.111
	20°C	210	234	23.5	Female	1.645	2.573
	20°C	211	266	25.5	Female	1.940	2.768
Carbamazepine	15°C	300	309	28.5	Female	2.710	3.303
	15°C	301	258	24.5	Male	2.111	0.152
	15°C	302	228	25.5	Female	2.390	2.597
	15°C	303	319	27	Female	2.775	3.588
	15°C	304	284	25	Female	2.601	0.670
	15°C	305	320	26.5	Male	2.753	0.185
	20°C	306	293	28.5	Female	1.957	3.161
	20°C	307	230	25.5	Male	1.641	0.027
	20°C	308	289	28	Male	1.536	0.132
	20°C	309	291	28.5	Male	1.753	0.198
	20°C	310	207	24.5	Male	1.554	0.117
	20°C	311	279	28	Male	1.333	0.094

Table S4 Accurate mass measurements of CBZ and its metabolites obtained by UPLC–(+)ESI-Orbitrap-MS in MS or MS/MS mode

Compound/ Photoproduct	Measured ion mass [m/z]	Elemental composition	Theoretical ion mass [m/z]	Rel. error [ppm]
CARBAMAZEPINE	237.1021	C ₁₅ H ₁₃ N ₂ O	237.1022	-0.5
	220.0757	C ₁₅ H ₁₀ NO	220.0757	-0.2
	194.0964	C ₁₄ H ₁₂ N	194.0964	0.1
CBZ-M252	253.0972	C ₁₅ H ₁₃ N ₂ O ₂	253.0972	0.1
	236.0710	C ₁₅ H ₁₀ NO ₂	236.0706	1.8
	210.0915	C ₁₄ H ₁₂ NO	210.0913	0.9
	182.0962	C ₁₃ H ₁₂ N	182.0964	-1.2
CBZ-M268	269.0920	C ₁₅ H ₁₃ N ₂ O ₃	269.0921	-0.1
	226.0864	C ₁₄ H ₁₂ NO ₂	226.0863	0.7
	208.0759	C ₁₄ H ₁₀ NO	208.0757	0.8
	180.0809	C ₁₃ H ₁₀ N	180.0808	0.5
CBZ-M270	271.1078	C ₁₅ H ₁₅ N ₂ O ₃	271.1077	0.3
	254.0813	C ₁₅ H ₁₂ NO ₃	254.0812	0.8
	210.0916	C ₁₄ H ₁₂ NO	210.0913	1.2
	180.0810	C ₁₃ H ₁₀ N	180.0808	1.2
CBZ-M428 (A,B)	429.1291	C ₂₁ H ₂₁ N ₂ O ₈	429.1292	-0.3
	253.0973	C ₁₅ H ₁₃ N ₂ O ₂	253.0972	0.5
	236.0706	C ₁₅ H ₁₀ NO ₂	236.0706	0.1
	210.0915	C ₁₄ H ₁₂ NO	210.0913	0.7
	182.0970	C ₁₃ H ₁₂ N	182.0964	3.3
	141.0185	C ₆ H ₅ O ₄	141.0182	2.1
	113.0238	C ₅ H ₅ O ₃	113.0233	4.2
CBZ-M444 (A,B)	445.1244	C ₂₁ H ₂₁ N ₂ O ₉	445.1242	0.5
	269.0923	C ₁₅ H ₁₃ N ₂ O ₃	269.0921	0.8
	253.0971	C ₁₅ H ₁₃ N ₂ O ₂	253.0972	-0.2
	226.0864	C ₁₄ H ₁₂ NO ₂	226.0863	0.6
	208.0759	C ₁₄ H ₁₀ NO	208.0757	1.0
	180.0807	C ₁₃ H ₁₀ N	180.0808	-0.4
	141.0185	C ₆ H ₅ O ₄	141.0182	1.7
	113.0239	C ₅ H ₅ O ₃	113.0233	4.9
CBZ-M446	447.1400	C ₂₁ H ₂₃ N ₂ O ₉	447.1398	0.4
	272.1650	C ₁₇ H ₂₂ NO ₂	272.1645	1.8
	253.0974	C ₁₅ H ₁₃ N ₂ O ₂	253.0972	0.9
	210.0916	C ₁₄ H ₁₂ NO	210.0913	1.2
	196.0761	C ₁₃ H ₁₀ NO	196.0757	1.9
CBZ-M458	459.1400	C ₂₂ H ₂₃ N ₂ O ₉	459.1398	0.4
	240.1019	C ₁₅ H ₁₄ NO ₂	240.1019	-0.1
	225.0783	C ₁₄ H ₁₁ NO ₂	225.0784	-0.5
	180.0806	C ₁₃ H ₁₀ N	180.0808	-0.7

Table S5 Accurate mass measurements of IBU and its metabolites obtained by UPLC–(+)ESI-Orbitrap-MS in MS or MS/MS mode

Compound/ Photoproduct	Measured ion mass [m/z]	Elemental composition	Theoretical ion mass [m/z]	Rel. error [ppm]
IBUPROFEN	205.1230	C ₁₃ H ₁₇ O ₂	205.1234	-2.2
IBU-M222	221.1181	C ₁₃ H ₁₇ O ₃	221.1183	-1.0
IBU-M238	237.1130	C ₁₃ H ₁₇ O ₄	237.1132	-0.9
IBU-M313	312.1279	C ₁₅ H ₂₂ NO ₄ S	312.1275	1.1
	269.0725	C ₁₂ H ₁₅ O ₄ NS	269.0727	-1.0
	149.9855	C ₃ H ₄ O ₄ NS	149.9867	-7.3
	124.0061	C ₂ H ₆ O ₃ NS	124.0074	-10.1
	106.9795	C ₂ H ₃ O ₃ S	106.9808	-12.5
	79.9560	O ₃ S	79.9574	-17.0
IBU-M329	328.1228	C ₁₅ H ₂₂ NO ₅ S	328.1224	1.1
	310.1140	C ₁₅ H ₂₀ NO ₄ S	310.1119	6.9
	269.0727	C ₁₂ H ₁₅ O ₄ NS	269.0727	-0.2
	149.9853	C ₃ H ₄ O ₄ NS	149.9867	-8.8
	124.0060	C ₂ H ₆ O ₃ NS	124.0074	-10.6
	106.9794	C ₂ H ₃ O ₃ S	106.9808	-13.2
	79.9558	O ₃ S	79.9574	-19.5
IBU-M345	344.1174	C ₁₅ H ₂₂ NO ₆ S	344.1173	0.3
	326.1060	C ₁₅ H ₂₀ O ₅ NS	326.1068	-2.3
	269.0723	C ₁₂ H ₁₅ O ₄ NS	269.0727	-1.6
	150.9932	C ₃ H ₅ O ₄ NS	150.9945	-8.4
	124.0062	C ₂ H ₆ O ₃ NS	124.0074	-9.5
	106.9803	C ₂ H ₃ O ₃ S	106.9808	-5.0
	79.9559	O ₃ S	79.9574	-18.3
IBU-M382	381.1558	C ₁₉ H ₂₅ O ₈	381.1555	0.9
	193.0347	C ₆ H ₉ O ₇	193.0354	-3.5
	175.0243	C ₆ H ₇ O ₆	175.0248	-2.9
	159.1175	C ₁₂ H ₁₅	159.1179	-2.7
	129.0180	C ₅ H ₅ O ₄	129.0193	-10.3
	117.01780	C ₄ H ₅ O ₄	117.0193	-13.1
	113.0232	C ₅ H ₅ O ₃	113.0244	-10.7
	99.0077	C ₄ H ₃ O ₃	99.0088	-10.8
	85.0281	C ₄ H ₅ O ₂	85.0295	-16.5
IBU-M398 (A,B,C)	397.1506	C ₁₉ H ₂₅ O ₉	397.1504	0.5
	221.1177	C ₁₃ H ₁₇ O ₃	221.1183	-2.8
	193.0348	C ₆ H ₉ O ₇	193.0354	-3.0
	177.1278	C ₁₂ H ₁₇ O	177.1285	-3.9
	157.0133	C ₆ H ₅ O ₅	157.0142	-6.0
	113.0230	C ₅ H ₅ O ₃	113.0244	-12.5
	85.0280	C ₄ H ₅ O ₂	85.0295	-17.6
IBU-M414 (A,B)	413.1456	C ₁₉ H ₂₅ O ₁₀	413.1453	0.8
	237.1135	C ₁₃ H ₁₇ O ₄	237.1132	1.1
	193.0347	C ₆ H ₉ O ₇	193.0354	-3.5

	175.0236	C ₆ H ₇ O ₆	175.0248	-6.9
	157.0128	C ₆ H ₅ O ₅	157.0142	-9.2
	113.0229	C ₅ H ₅ O ₃	113.0244	-13.4
	85.0279	C ₄ H ₅ O ₂	85.0295	-18.8
IBU-M505	504.1545	C ₂₁ H ₃₀ NO ₁₁ S	504.1545	0.1
(A, B)	328.1223	C ₁₅ H ₂₂ NO ₅ S	328.1224	-0.4
	124.0059	C ₂ H ₆ O ₃ NS	124.0074	-11.9
	106.9794	C ₂ H ₃ O ₃ S	106.9808	-13.5
	79.9557	O ₃ S	79.9574	-20.4

Fig. S1 Extracted ion chromatograms (XIC) of A) treated sample of bile with CBZ and B) 2-hydroxycarbamazepine and 3-hydroxycarbamazepine standards

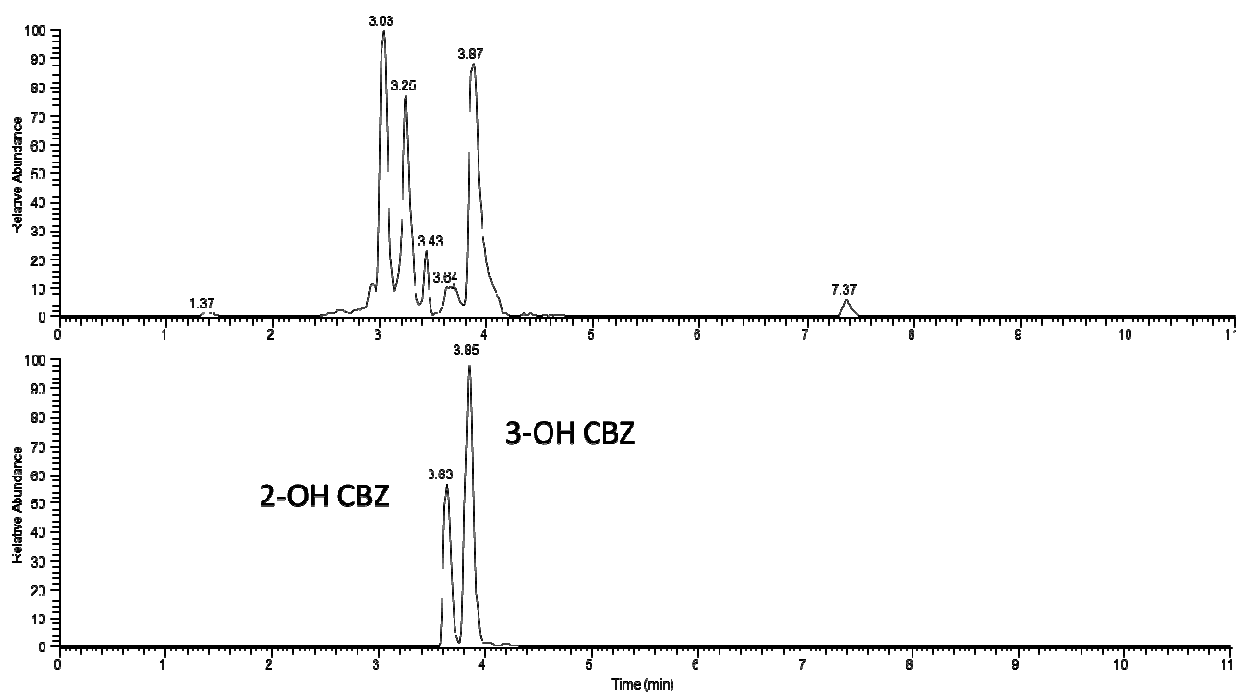


Fig. S2 Extracted ion chromatograms (XIC) of IBU detected metabolites in bile and corresponding confirmation with authentic standards

