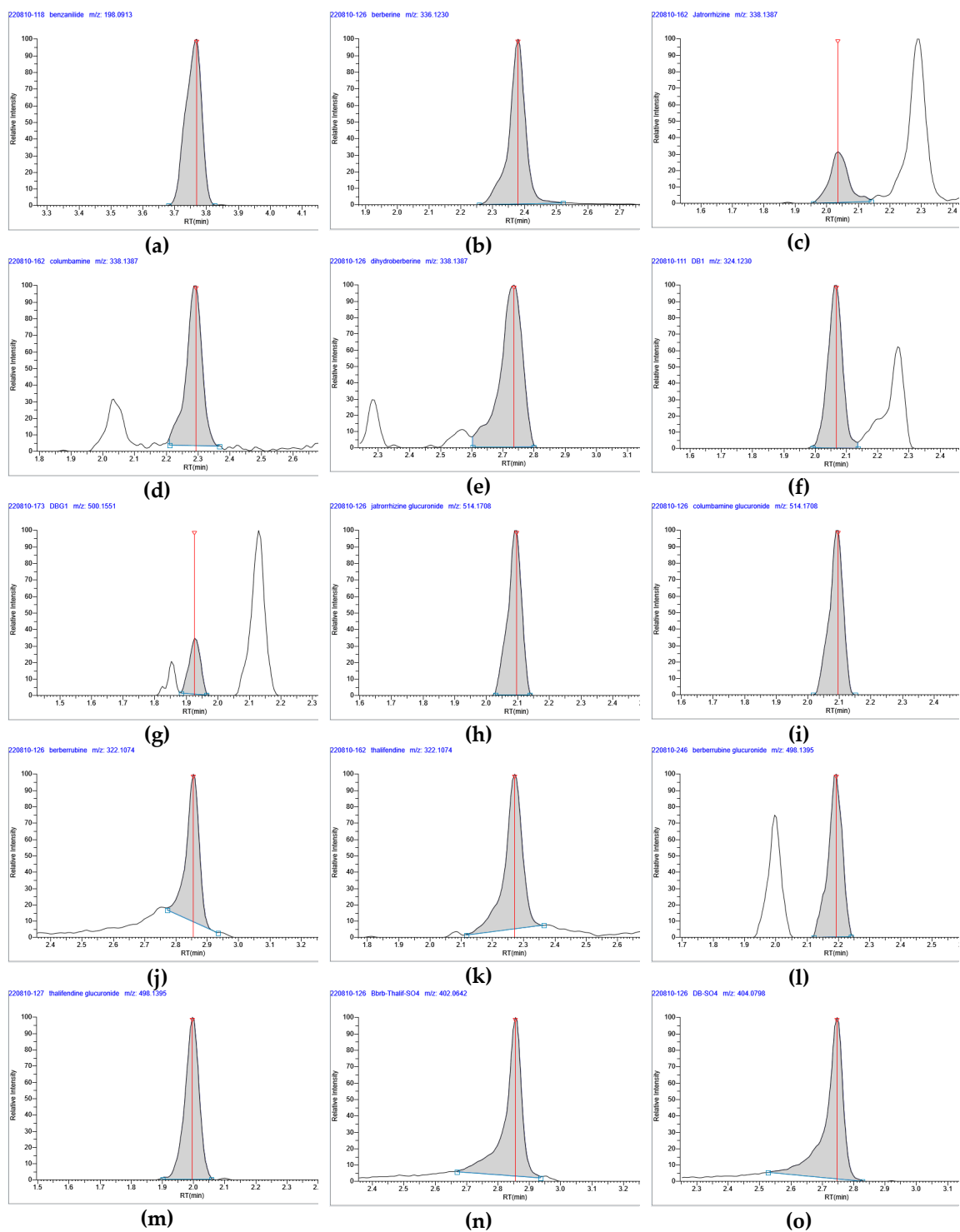


Table S1. HRMS data extraction parameters for berberine and its metabolites

Compound Name	Compound Formula	Adduct	m/z	Retention Time
Benzanilide	C13H11NO	M+H	198.0913	3.78
Berberine	C20H18NO4	M+H	336.1230	2.38
Jatrorrhizine	C20H20NO4	M	338.1387	2.02
Columbamine	C20H20NO5	M	338.1387	2.29
Dihydroberberine	C20H19NO4	M+H	338.1387	2.73
Demethyleneberberine (DB)	C19H18NO4	M+H	324.1230	2.06
Demethyleneberberine Glucuronide (DB-glu)	C25H26NO10	M+H	500.1551	1.93
Jatrorrhizine Glucuronide	C26H28NO10	M+H	514.1708	2.03
Columbamine Glucuronide	C26H28NO10	M+H	514.1708	2.10
Berberrubine	C19H16NO4	M+H	322.1074	2.86
Thalifendine	C19H16NO4	M+H	322.1074	2.25
Berberrubine Glucuronide	C25H24NO10	M+H	498.1395	2.19
Thalifendine Glucuronide	C25H24NO10	M+H	498.1395	1.99
Berberrubine or Thalifendine Sulfate (Bbrb/Thalif SO4)	C19H16NO7S	M+H	402.0642	2.85
Demethyleneberberine Sulfate (DB SO4)	C19H16NO7S	M+H	404.0798	2.75
Jatrorrhizine 3-O-Sulfate (Jatrorrhizine SO4)	C20H19NO7S	M+H	418.0955	2.75
Dihydroxy- Berberrubine or Thalifendine (dihydroxy-Bbrb/Thalif)	C19H16NO6	M+H	354.0972	2.76
Dihydroxyberberine	C20H18NO6	M	368.1129	2.07



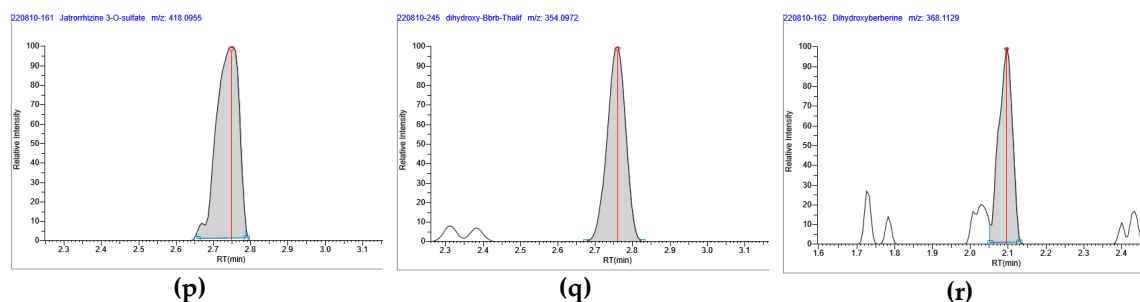
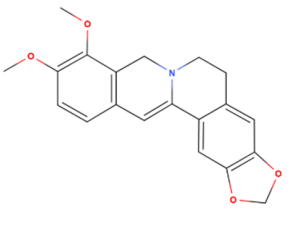
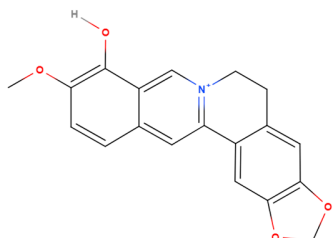
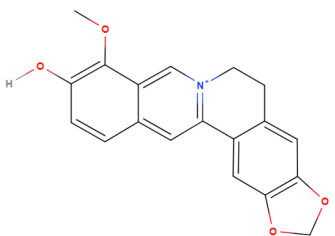
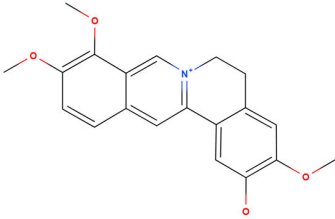
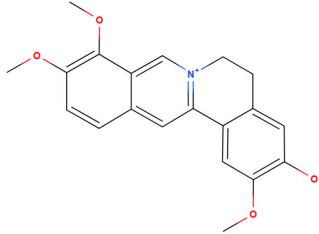
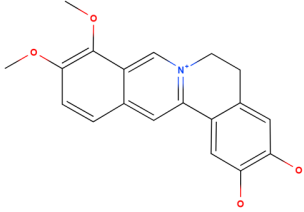
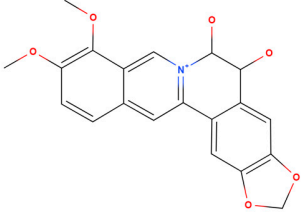
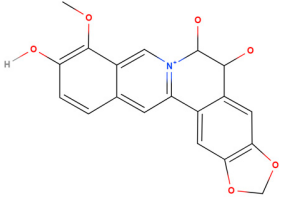
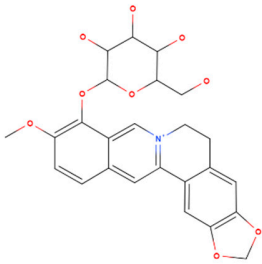
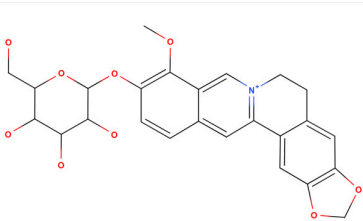
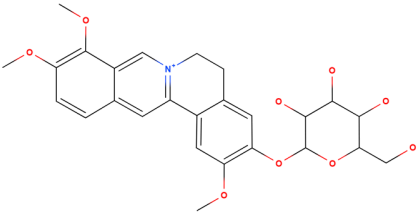
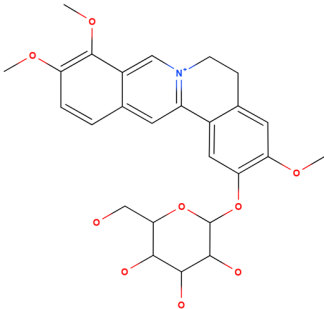
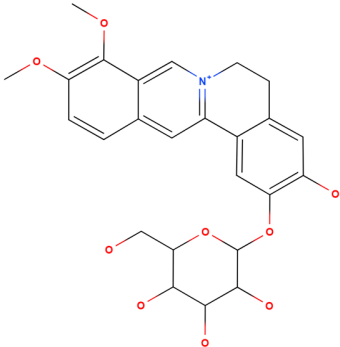


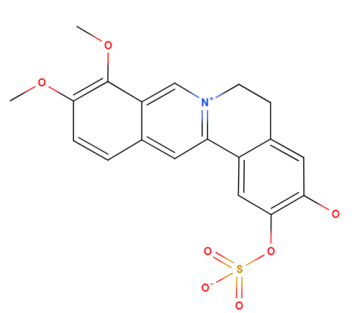
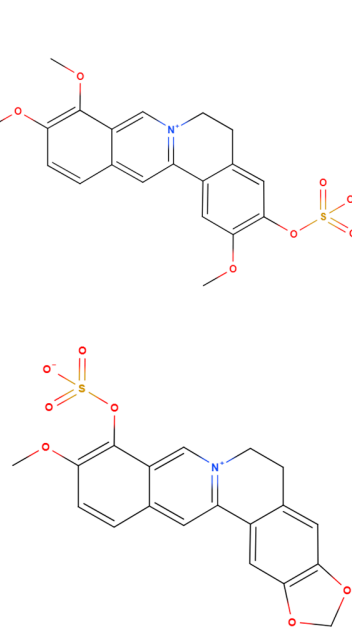
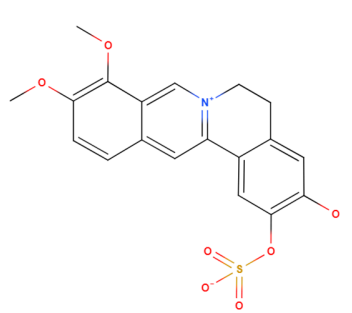
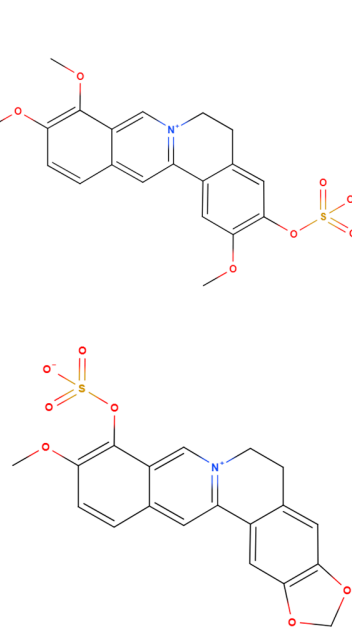
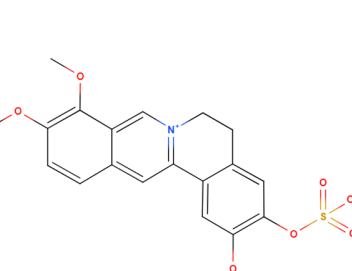
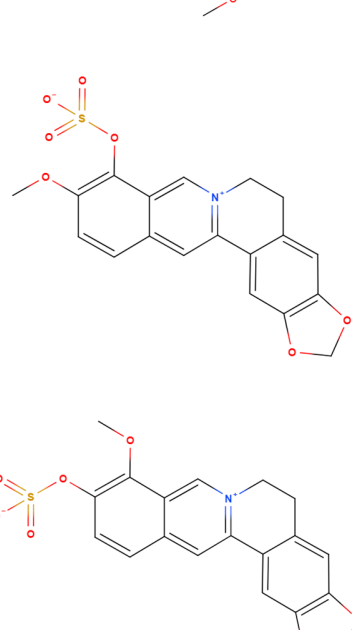
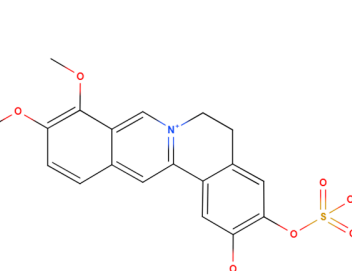
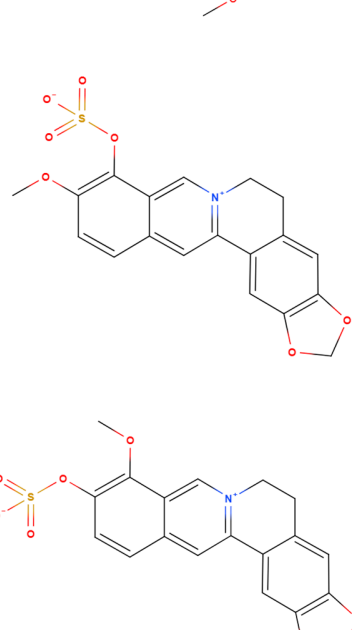
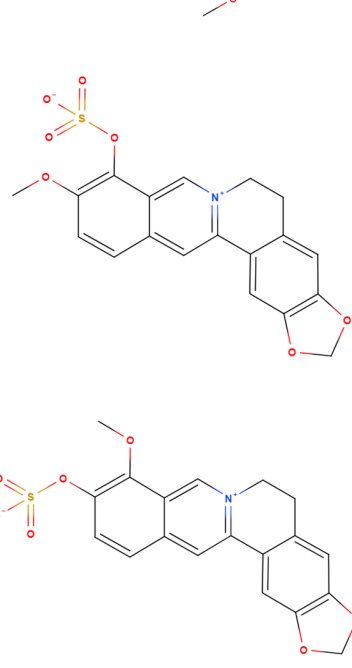

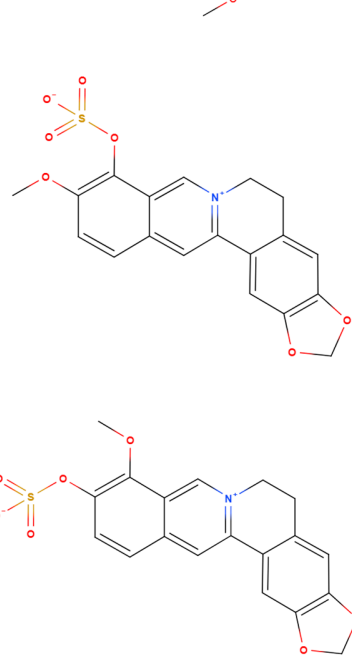

Figure S1: LC-HRMS chromatograms of berberine and its metabolites. (a) benzanilide (internal standard), (b) berberine, (c) jatrorrhizine, (d) columbamine, (e) dihydroberberine, (f) demethyleneberberine, (g) demethyleneberberine glucuronide, (h) jatrorrhizine glucuronide, (i) columbamine glucuronide, (j) berberrubine, (k) thalifendine, (l) berberrubine glucuronide, (m) thalifendine glucuronide, (n) berberrubine or thalifendine sulfate, (o) demethyleneberberine sulfate, (p) jatrorrhizine 3-o-sulfate, (q) dihydroxy-berberrubine or thalifendine, and (r) dihydroxyberberine

Table S2. Summary of observed differences in DHB and LMB treatments in Caco-2 metabolites and Human blood metabolites.

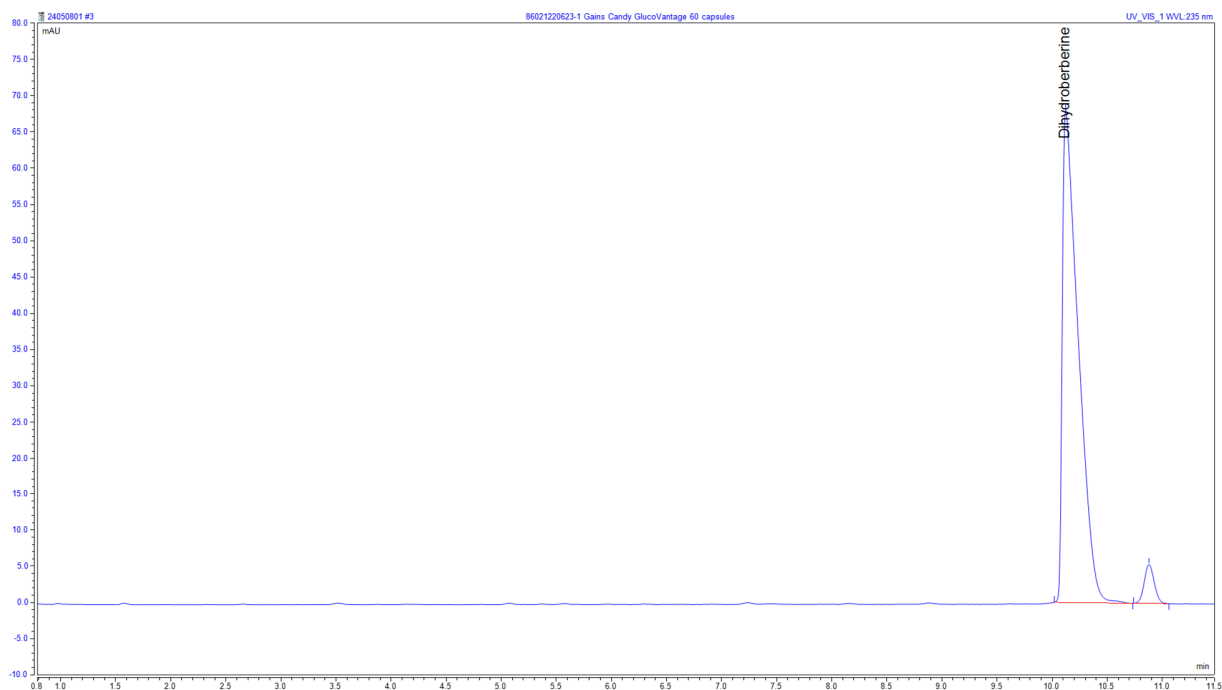
Compound Name	Chemical Structure	Concentrations in Caco-2 cell (ng/mL)		Concentrations in Human Blood (ng/mL)	
		DHB	LMB	DHB	LMB
Dihydroberberine		ND	22 ± 2	0.85 ± 0.45	0.012 ± 0.007
Berberrubine		0.75 ± 0.10	43 ± 7	1.3 ± 0.4	0.085 ± 0.03
Thalifendine		0.0083 ± 0.0008	ND	3.6 ± 1.4	0.73 ± 0.12
Columbamine		1360 ± 110	56 ± 19	1.4 ± 0.8	0.32 ± 0.13

Jatrorrhizine		0.032 ± 0.014	0.004 ± 0.004	1.07 ± 0.25	0.59 ± 0.16
Demethyleneberberine		0.23 ± 0.04	0.98 ± 0.14	7.9 ± 2.7	0.12 ± 0.05
Dihydroxyberberine		ND	ND	0.16 ± 0.06	0.0023 ± 0.0013
Dihydroxy-berberrubine (top) and Dihydroxy-thalifendine (bottom)		ND	0.14 ± 0.03 [‡]	0.25 ± 0.10 [‡]	0.25 ± 0.10 [‡]
					
Berberrubine glucuronide		ND	ND	ND	0.055 ± 0.013

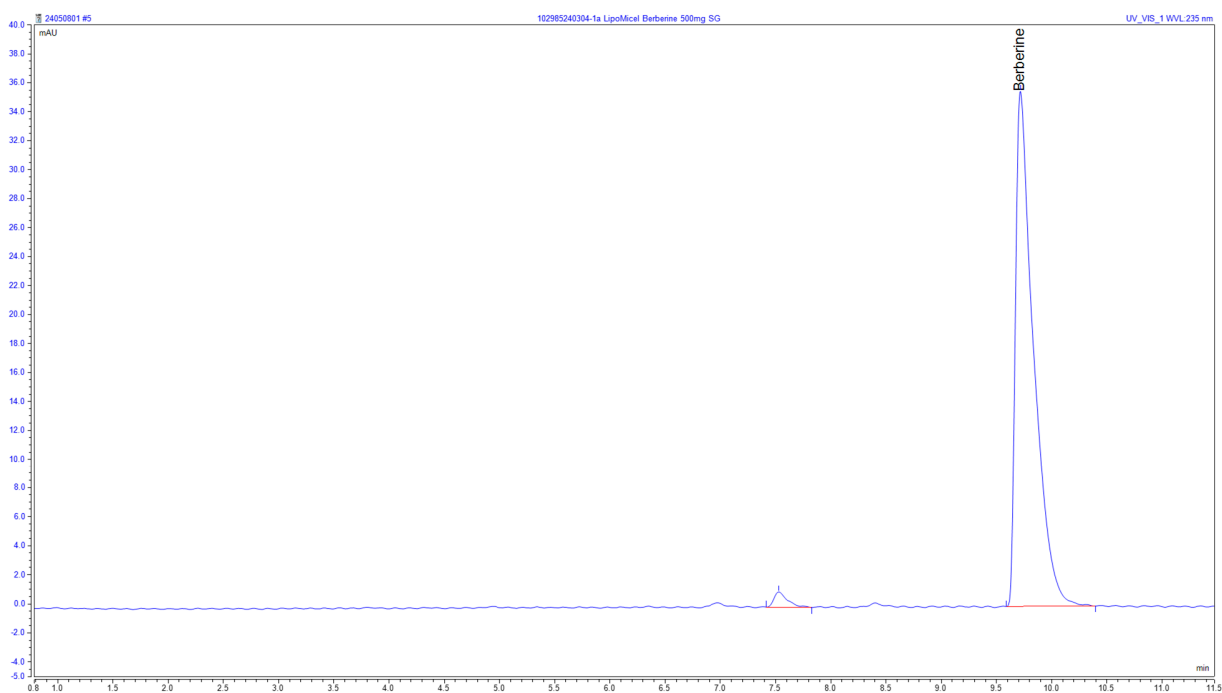
					
Thalifendine glucuronide		ND	ND	0.21 ± 0.12	0.06 ± 0.01
Jatrorrhizine glucuronide		0.0023 ± 0.002	ND	0.050 ± 0.043	0.019 ± 0.007
Columbamine glucuronide		ND	ND	0.07 ± 0.04	0.007 ± 0.003
Demethyleneberberine glucuronide (DBG)		ND	ND	0.098 ± 0.044	0.0098 ± 0.0036
Demethyleneberberine sulfate (DB SO ₄)		0.27 ± 0.04	0.78 ± 0.10	2.4 ± 0.8	0.35 ± 0.05

 <p>Jatrorrhizine sulfate</p>	 <p>Berberrubine sulfate (top) and thalifendine sulfate (bottom) (Bbrb/Thalif SO4)</p>		
 <p>Jatrorrhizine sulfate</p>	 <p>Berberrubine sulfate (top) and thalifendine sulfate (bottom) (Bbrb/Thalif SO4)</p>		
 <p>Jatrorrhizine sulfate</p>	 <p>Berberrubine sulfate (top) and thalifendine sulfate (bottom) (Bbrb/Thalif SO4)</p>		

ND = Not detected. Mean \pm SEM reported. # Results reported are the sums of both compounds.



(a)



(b)

Figure S2: UHPLC chromatograms of study treatments. (a) DHB, chromatogram confirmed the identity and purity of dihydroberberine at its characteristic retention time of 10.12 minute with no detectable amount of berberine. The smaller peak at 10.9 minute likely resulted from the black pepper extract. (b) LMB, chromatogram confirmed the identity and purity of berberine at its characteristic retention time of 9.72 minute with no detectable amount of dihydroberberine. The smaller peak at 7.5 minute likely resulted from one of the excipients used to manufacture the capsule.