

Supplementary Materials

Table S1. Information on chemicals and reagents used for *in vitro* and *in vivo* experiments.

Category	Chemicals and reagents	Company
Cell line	HaCaT	CLS Cell Lines Service GmbH (Eppelheim, Baden-Württemberg, Germany)
Cell culture	DMEM FBS Penicillin and streptomycin	Thermo Fisher Scientific (Waltham, MA, USA)
Reagent	Acetonitrile (LC–MS grade) Methanol (LC–MS grade) Water (LC–MS grade)	Fisher Scientific Co., LLC (San Jose, CA, USA)
	Ammonium formate	Kanto Chemical Co., Inc. (Tokyo, Japan)
	Prednisolone Silymarin	Sigma-Aldrich (St. Louis, MO, USA)
	Biostir-AD [®]	Biostir Inc. (Osaka, Japan)
	Entobar (pentobarbital sodium)	Hanlim Pharm. Co., Ltd. (Seoul, Republic of Korea)
	Formic acid	Fujifilm Wako Pure Chemical Co., Ltd. (Osaka, Japan)
	IFN- γ TNF- α	R&D Systems Inc. (Minneapolis, MN, USA)
	10% Neutral-buffered formalin	BBC Biochemical (WA, USA)

Assay kit	CCK-8	Dojindo (Kumamoto, Japan)
	Corticosterone Cortisol	MyBioSource, Inc. (San Diego, CA, USA)
	Histamine	Oxford Biomedical Research Inc. (MI, USA)
	IgE	Abcam (Cambridge, UK & Cambridge, Massachusetts, USA)
	MDC RANTES TARC TNF- α	R&D Systems Inc. (Minneapolis, MN, USA)
Antibody	CD4 ⁺ T cell TSLP	Abcam (Cambridge, UK & Cambridge, Massachusetts, USA)
	ICAM-1 IL-4	Biorbyt Ltd. (Cowley Road, Cambridge, United Kingdom)

Table S2. UPLC-MS/MS MRM analysis conditions for simultaneous quantification of compounds in CTE.

UPLC conditions		MS conditions	
System	Acquity UPLC I-Class	System	Xevo TQ-XS
Column	Acquity UPLC BEH C18 column (2.1 mm × 100 mm, 1.7 μm)	Software	MassLynx v4.2
Column temp.	45°C	Ion source	ESI positive (+)
Sample temp.	5°C	Acquisition mode	MRM
Injection volume	2.0 μL	Capillary voltage	1.2 kV
Flow rate	0.3 mL/min	Cone gas flow	50 L/h
Mobile phase A	0.1% (v/v) aqueous formic acid with 5 mM ammonium formate	Desolvation gas flow	700 L/h
Mobile phase B	Acetonitrile	Desolvation temperature	500°C
	Time (min) A (%) B (%)	Source temperature	150°C
	Initial 80 20		
	0.1 80 20		
Gradient	14.0 5 95		
	15.0 0 100		
	15.1 80 20		
	18.0 80 20		

CTE, Corydalis Tuber 70% ethanol extract; ESI, electrospray ionization; MRM, multiple reaction monitoring; UPLC-MS/MS, ultra-performance liquid chromatography-tandem mass spectrometry.

Table S3. UPLC-MS/MS MRM conditions for simultaneous analysis of compounds in CTE.

Code No.	Name	Ion mode	Molecular weight	MRM transition		Cone voltage (V)	Collision energy (eV)	Retention time (min)
				Precursor ion	Product ion			
1	Tetrahydrocolumbamine	+	341.4	342.3	178.0	30	25	2.07
2	Protopine	+	353.4	354.3	188.0	30	30	2.39
3	Columbamine	+	338.4	338.3	322.0	30	30	2.54
4	Glaucine	+	355.4	356.3	293.9	30	20	2.64
5	Coptisine Cl	+	355.8	320.3	292.0	45	25	2.68
6	Tetrahydropalmatine	+	355.4	356.4	192.1	30	25	2.70
7	Tetrahydrocoptisine	+	323.3	324.3	176.0	30	30	2.85
8	Berberrubine Cl	+	357.8	322.2	307.1	30	30	2.90
9	Canadine	+	339.4	340.5	176.0	30	30	3.02
10	Corydaline	+	369.5	370.3	192.1	30	30	3.18
11	Palmatine Cl	+	387.9	352.3	336.0	40	30	3.25
12	Berberine Cl	+	371.8	336.3	320.0	35	30	3.34
13	Dehydrocorydaline	+	366.4	366.6	350.0	30	30	3.61

CTE, Corydalis Tuber 70% ethanol extract; MRM, multiple reaction monitoring; UPLC-MS/MS, ultra-performance liquid chromatography-tandem mass spectrometry.

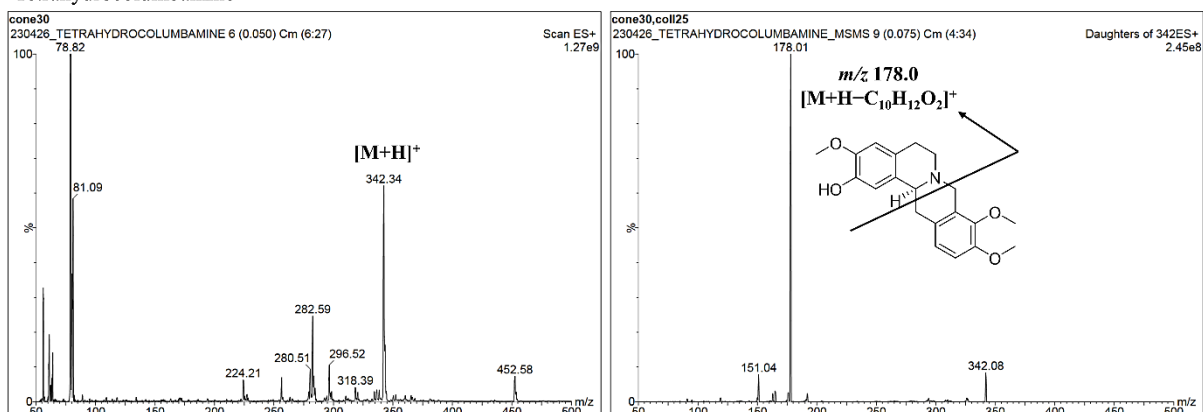
Table S4. Parameters for simultaneous quantification of compounds in CTE using the UPLC-MS/MS MRM method.

Code No.	Name	Linear range ($\mu\text{g/L}$)	Regression equation $y = ax + b$	r^2	LOD ($\mu\text{g/L}$)	LOQ ($\mu\text{g/L}$)
1	Tetrahydrocolumbamine	5 – 500	$y = 127390x - 120495$	0.9981	0.01	0.03
2	Protopine	5 – 500	$y = 29438x - 58058.8$	0.9971	0.03	0.08
3	Columbamine	10 – 1,000	$y = 43294x - 295391$	0.9964	0.19	0.58
4	Glaucine	5 – 500	$y = 80863x - 198295$	0.9985	0.02	0.06
5	Coptisine Cl	50 – 5,000	$y = 6147x - 119463$	0.9959	0.05	0.15
6	Tetrahydropalmatine	5 – 500	$y = 159174x - 505675$	0.9969	0.01	0.03
7	Tetrahydrocoptisine	5 – 500	$y = 133471x - 303105$	0.9995	0.01	0.04
8	Berberrubine Cl	5 – 500	$y = 122839x - 491686$	0.9962	0.04	0.12
9	Canadine	10 – 1,000	$y = 72227x - 691221$	0.9927	0.03	0.08
10	Corydaline	10 – 1,000	$y = 63976x - 236447$	0.9989	0.18	0.54
11	Palmatine Cl	10 – 1,000	$y = 92806x - 450835$	0.9965	0.01	0.02
12	Berberine Cl	50 – 5,000	$y = 19982x - 682596$	0.9991	0.03	0.08
13	Dehydrocorydaline	10 – 1,000	$y = 39203x - 183104$	0.9987	0.06	0.18

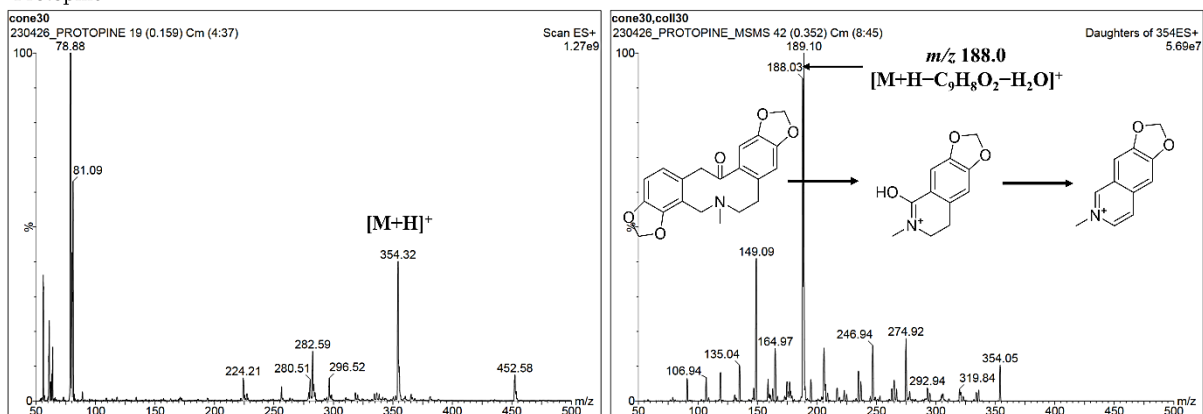
CTE, Corydalis Tuber 70% ethanol extract; LOD, limit of detection; LOQ, limit of quantitation; MRM, multiple reaction monitoring; UPLC-MS/MS, ultra-performance liquid chromatography-tandem mass spectrometry; x , concentration ($\mu\text{g/L}$) of each reference compound; y , peak area of each reference compound.

Figure S1. Precursor ion (Q1, left) and product ion (Q3, right) chromatograms of each reference standard compound by UPLC-MS/MS MRM mode.

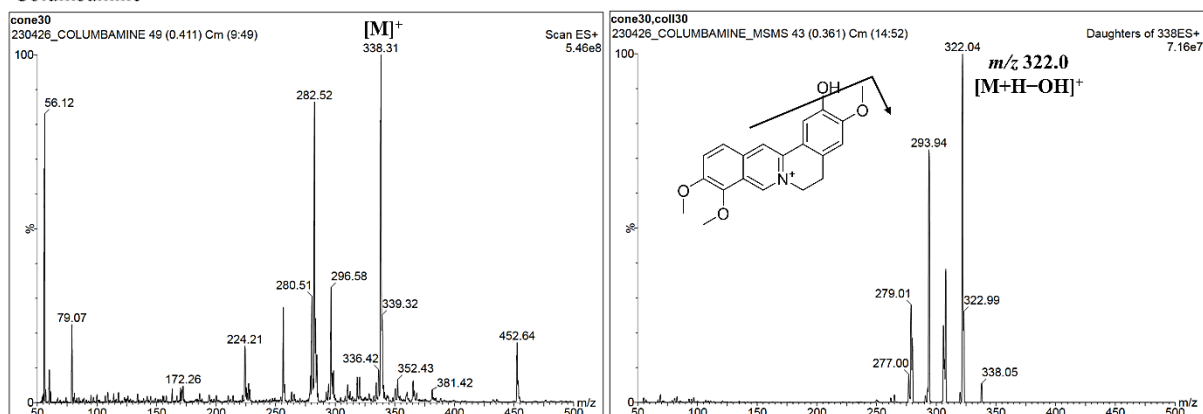
Tetrahydrocolumbamine



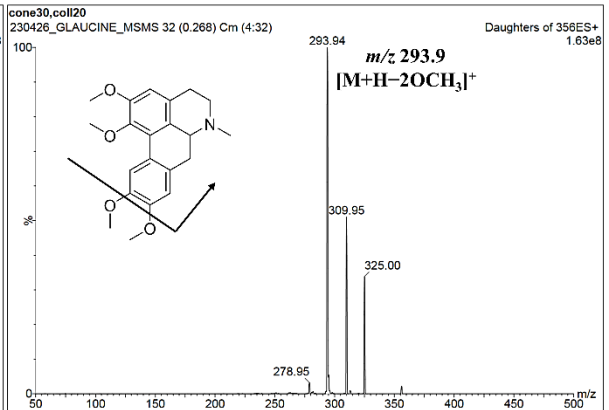
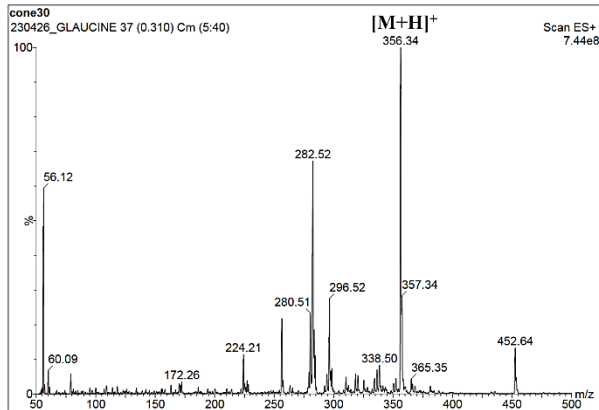
Protopine



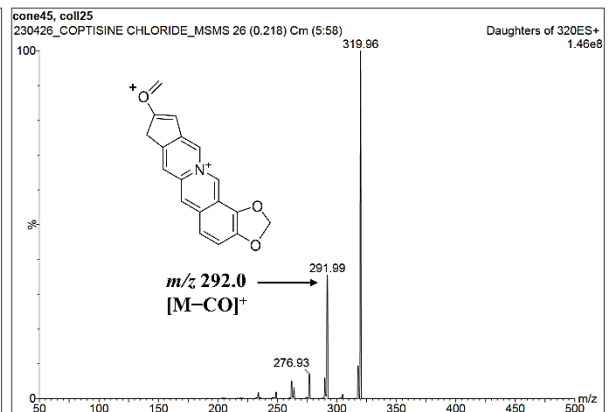
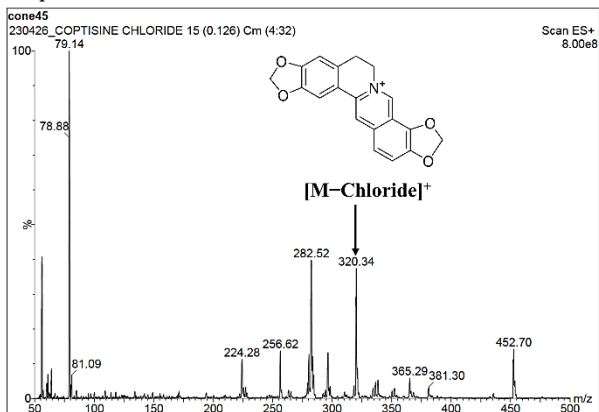
Columbamine



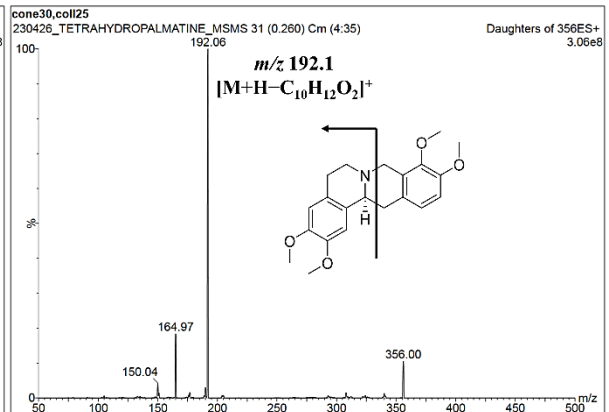
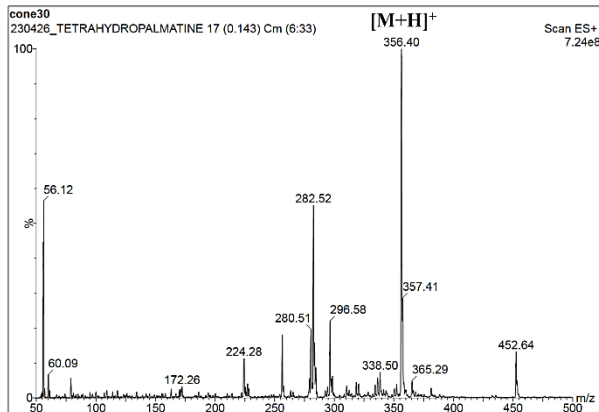
Glaucine



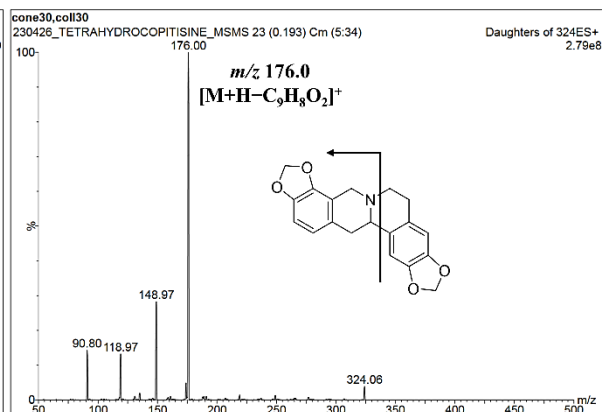
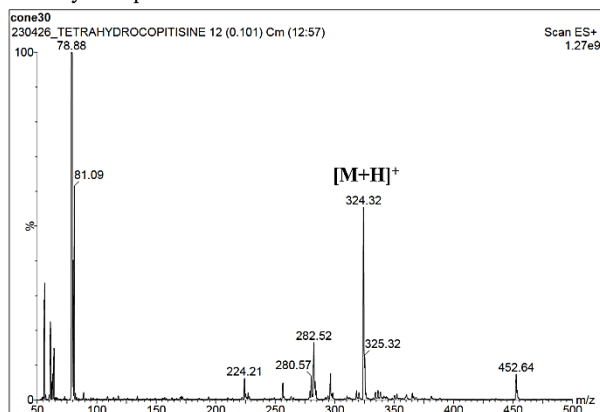
Coptisine Cl



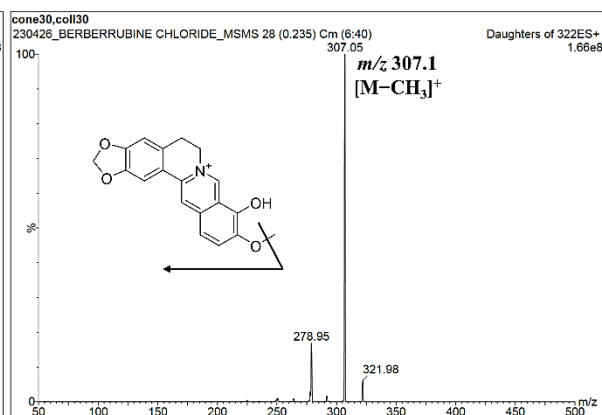
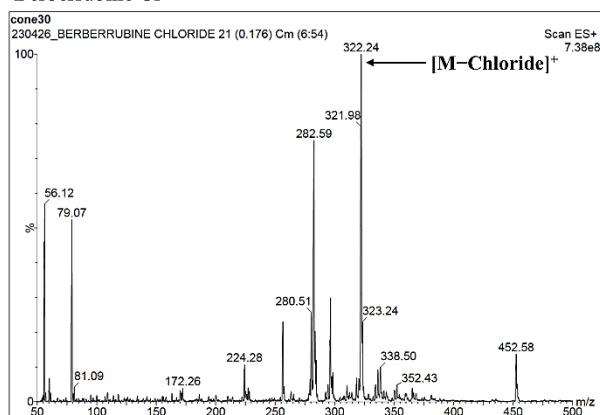
Tetrahydropalmatine



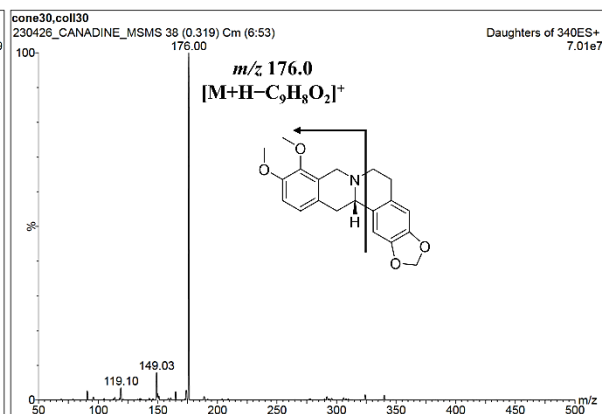
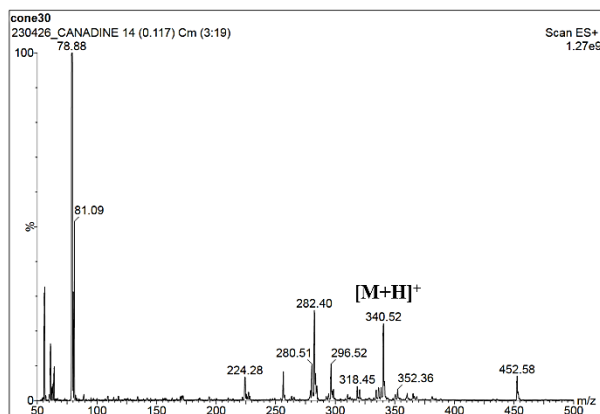
Tetrahydrocoptisine



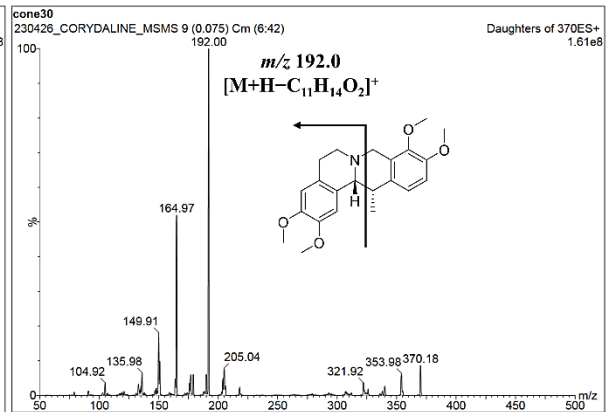
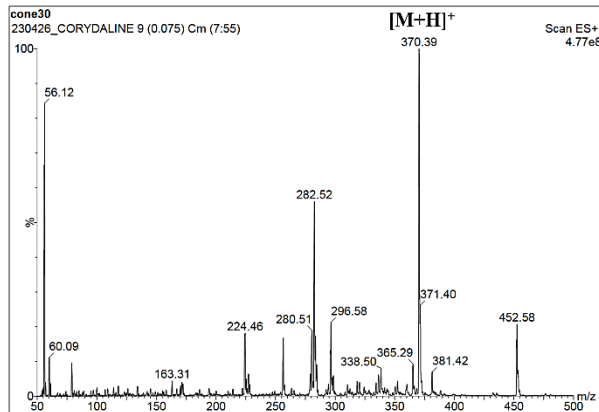
Berberrubine Cl



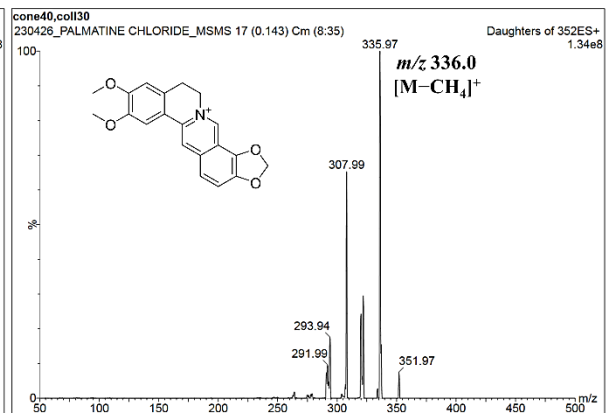
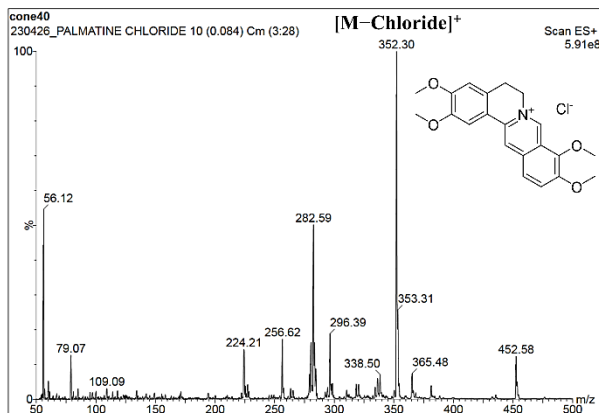
Canadine



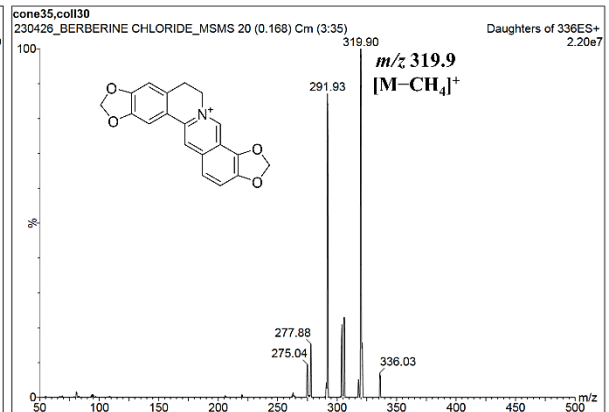
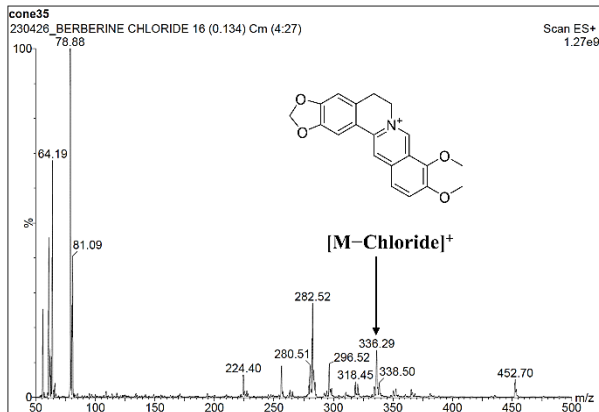
Corydaline



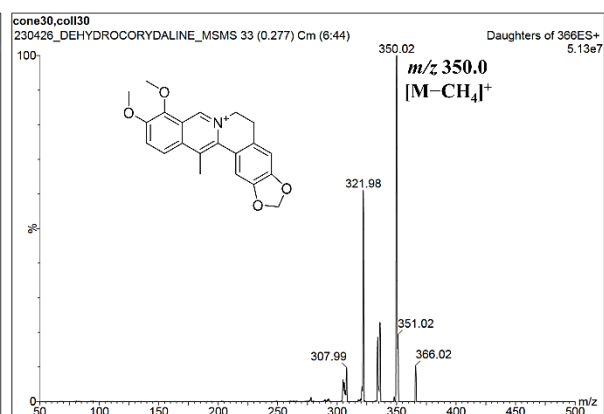
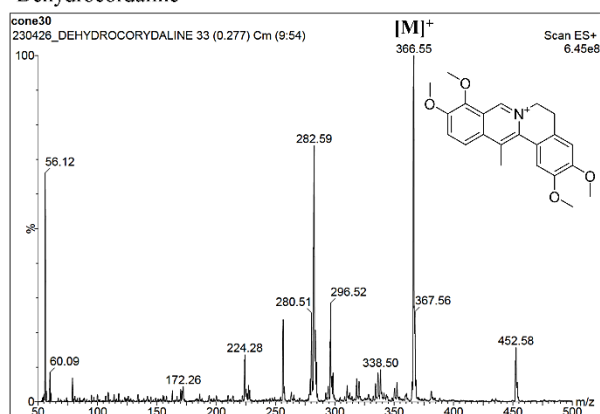
Palmatine Cl



Berberine Cl

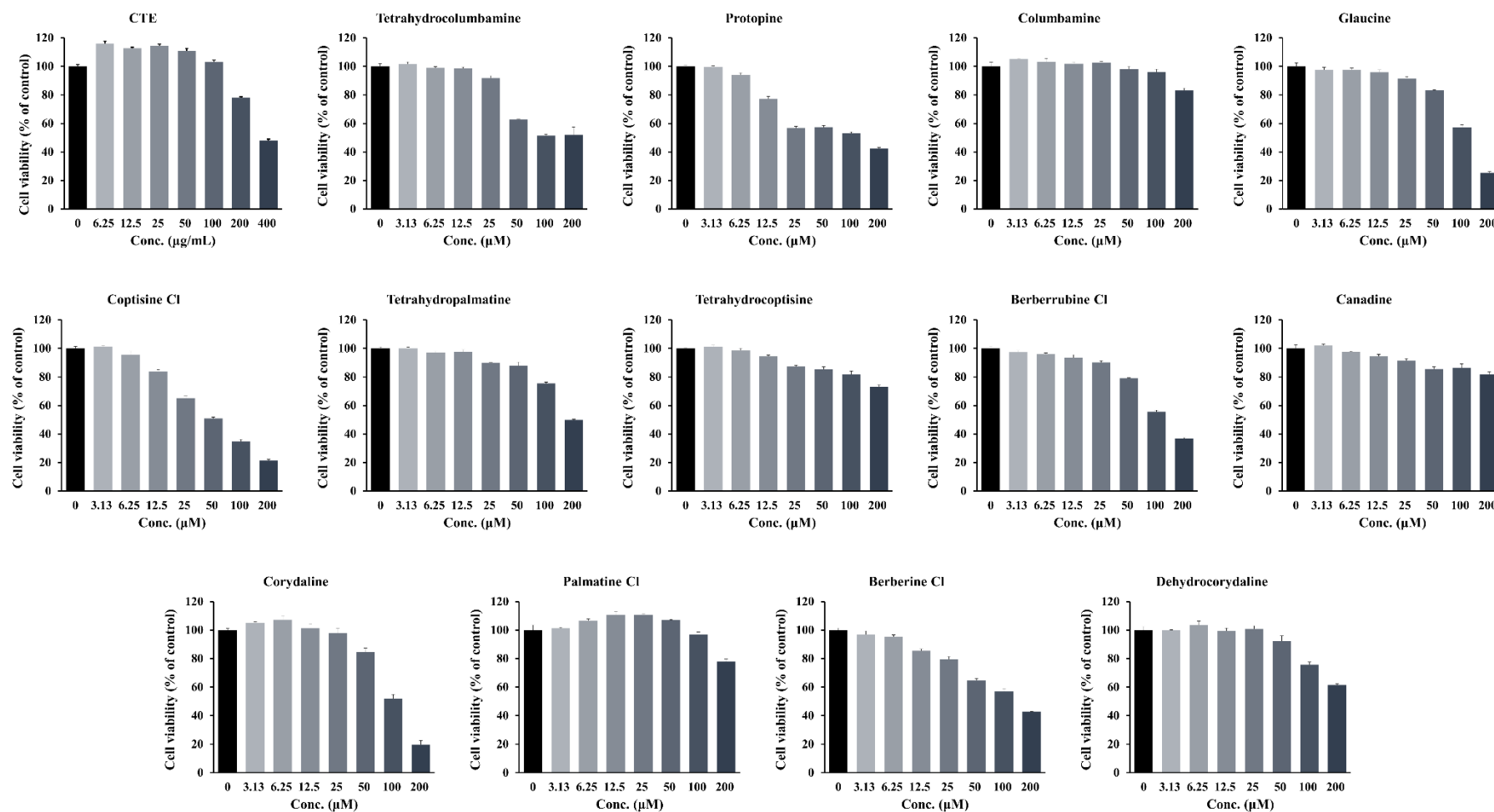


Dehydrocordaline



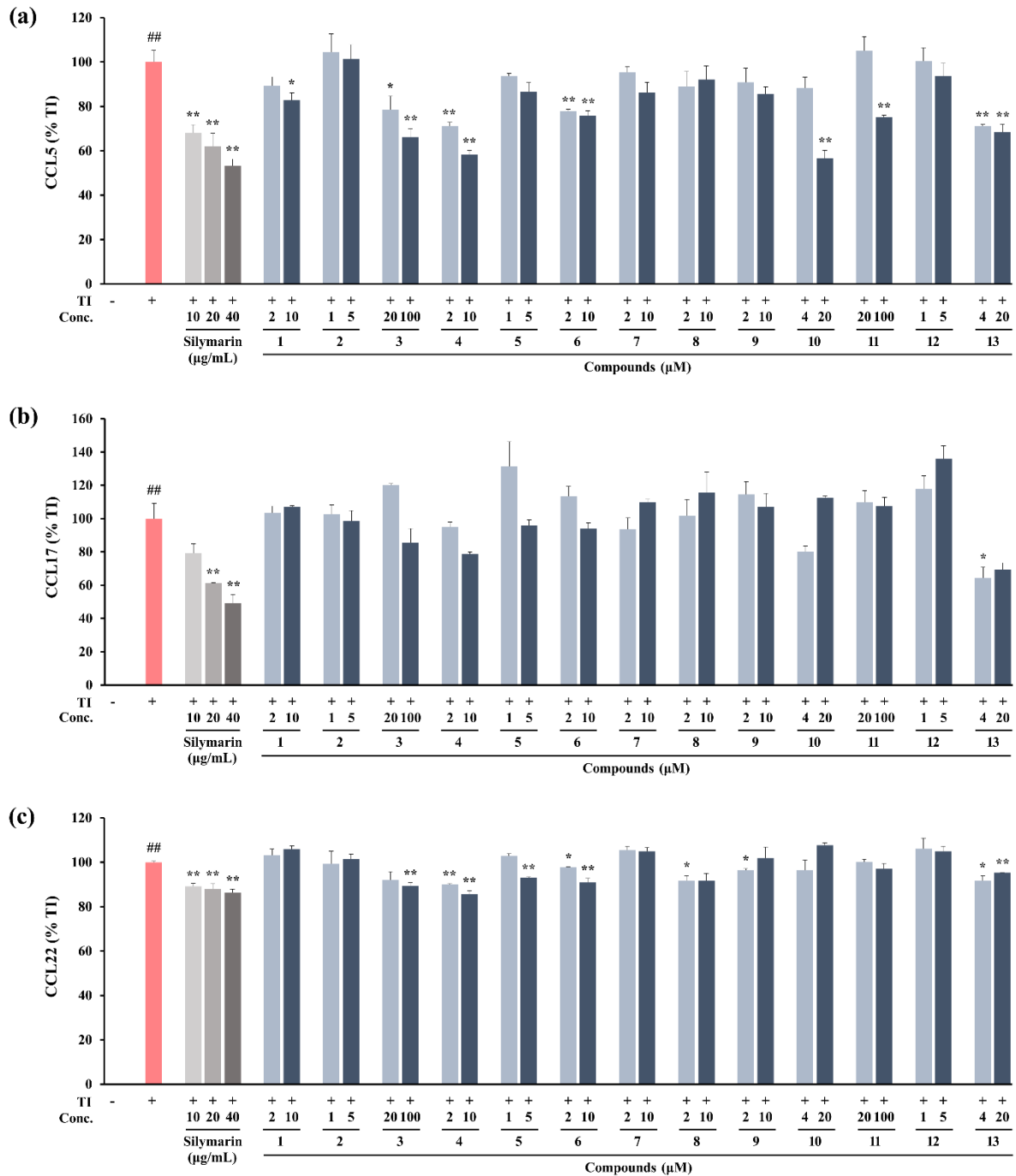
UPLC-MS/MS, ultra-performance liquid chromatography-tandem mass spectrometry.

Figure S2. Cytotoxicity of CTE and its compounds in HaCaT cells.



Data are expressed as means \pm SEMs (n = 4). CTE, Corydalis Tuber 70% ethanol extract.

Figure S3. Effect of compounds included in CTE on chemokine production in TI-stimulated HaCaT cells.



The cells were treated with CTE and stimulated with TI for 24 h. The levels of CCL5 (a), CCL17 (b), and CCL22 (c) in the supernatant were measured. Silymarin was used as a positive control. Data are expressed as means \pm SEMs ($n = 3$). ## $p < 0.01$ vs. normal control; * $p < 0.05$ and ** $p < 0.01$ vs. TI-stimulated cells. CTE, Corydalis Tuber 70%

ethanol extract; TL, TNF- α (10 ng/mL) and IFN- γ (10 ng/mL); **1**, Tetrahydrocolumbamine; **2**, protopine; **3**, columbamine; **4**, glaucine; **5**, coptisine Cl; **6**, tetrahydropalmatine; **7**, tetrahydrocoptisine; **8**, berberrubine Cl; **9**, canadine; **10**, corydaline; **11**, palmatine Cl; **12**, berberine Cl; **13**, dehydrocorydaline.