RSC Advances

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Biologically active isoquinoline alkaloids with drug-like properties from the genus *Corydalis*†

Cite this: *RSC Adv.*, 2014, **4**, 15900

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The genus *Corydalis* (Papaveraceae), comprising more than 400 species in Eurasia and North America, is a rich source of isoquinoline alkaloids with various biological properties including acetylcholinesterase inhibitory effects, anti-proliferative activities, antiviral activities and antiplasmodial activities. Traditionally, some *Corydalis* species have long been used to treat gastric and duodenal ulcers, dysmenorrhoea, rheumatism and cardiac arrhythmia disease and this traditional background so far has proven pharmacological activities resulting in isolation of more than 100 isoquinoline alkaloids from this genus. This overview aims to inform medicinal chemists of the good drug-like properties and versatile biological activities of *Corydalis* alkaloids to stimulate further medicinal chemistry research. Mechanisms of action and structure–activity relationship of *Corydalis* alkaloids are also included.

Received 24th December 2013
Accepted 10th March 2014

DOI: 10.1039/c3ra47944g

www.rsc.org/advances

1. Introduction

Native to the temperate Northern Hemisphere, the genus *Corydalis* consists of more than 400 species in Eurasia and North America.[1] These annual and perennial herbaceous plants with beautiful colorful flowers are a rich source of alkaloids, particularly isoquinoline alkaloids with various biological properties. Traditionally, some *Corydalis* species have long been used for the treatment of different ailments in China, Korea, Japan and other Eastern Asian countries. For example, *C. yanhusuo*, a traditional Chinese medicine, has been used to treat gastric and duodenal ulcers, dysmenorrhoea, rheumatism and cardiac arrhythmia disease.[2,3] Every year new alkaloid structures, together with their biological activities, are reported from the plants of this genus. Some of these alkaloids possess promising biological/pharmacological properties for the

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† Electronic supplementary information (ESI) available. See DOI: 10.1039/c3ra47944g

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treatment of important diseases including cancer, Alzheimer's and microbial infections.

To date, there is no comprehensive review on *Corydalis* alkaloids (CA) and their biological activities. This review deals with chemistry, drug-like features, and biological properties of CA.

Although isoquinoline alkaloids have been reported from many plants, and even other biological sources, however, the diversity of structures in CA has particularly been attractive to natural product researchers.

In today's era of drug development, many bioactive compounds are discovered from nature or combinatorial synthesis by high throughput screening, but only a few bioactive compounds reach the clinical trials phases, often because of the lack of bioavailability. In recent years, several parameters have been introduced for the prediction of drug-like physicochemical properties of drug candidates including intestinal absorption, bioavailability and cell penetration.[4,5] In this regard, the most well-known rule is "rule of five" or Lipinski's rule. According to this rule, an oral drug-like molecule should have an octanol–water partition coefficient (log *P*) of <5, <5 hydrogen bond donors (HBD), <10 hydrogen bond acceptors (HBA) and a molecular weight of <500 Da.[6] Two more requirements for drug-like molecules, proposed by Veber and co-workers, include NROT (number of rotatable bonds) of <10 and a PSA (polar surface area) of <140 Å^2.[7] Considering this description, we evaluated the above parameters for CA in this review to see the eligibility of biologically active CA for being future drugs. Interestingly, most of CA satisfied these criteria for lead-like compounds.

Up to now, different classes of isoquinoline alkaloids have been found in *Corydalis* species including aporphine, protopine, protoberberine, tetrahydroprotoberberine, benzo[*c*]phenanthridine, phthalideisoquinoline, benzylisoquinoline, morphinan, and spirobenzylisoquinoline (Fig. 1). On the basis of these scaffolds, in this review, we identified structure–activity relationships for CA, in order to stimulate further medicinal chemistry researches. Given the generally good drug-like properties, medicinal chemistry programs need to conserve the physic-chemical properties in analogous development.

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2. Biological properties

2.1. Anti-proliferative/anti-cancer activity

Kim *et al.* studied the cytotoxicity of fourteen alkaloids from the tubers *C. ternata* on several tumor cell lines such as A549, SK-OV-3, SK-MEL-2, and HCT-15. They found that demethylcorydalmine (**1**, Fig. 2) had significant cytotoxicity against A549, SK-OV-3, SK-MEL-2, and HCT-15 cell lines with IC_{50} values of 8.34, 5.14, 7.87, and 2.86 μM, respectively.[8] In addition, they found that the benzylisoquinoline alkaloids of *C. ternata* such as *epi*-coryximine (**2**), coryternatines A–C (**3–5**), (*S*)-reticuline (**6**) and (*R*)-reticuline (**7**) were selectively cytotoxic against HCT-15 with IC_{50} values ranging from 23 to 29 μM. Their findings revealed that protoberberine-type alkaloids such as compound **1** and berberine (**8**) showed cytotoxicity against human cell lines (IC_{50} values ranging from 6.27 to 16.59 μM), yet the protopine-type alkaloid such as protopine (**27**) was inactive.[8] In another study, Kim *et al.* found that berberine (**8**) obtained from *C. pallida* possessed cytotoxic activity against HT-1080 (human fibrosarcoma) and SNU-638 (human stomach adenocarcinoma), with IC_{50} values of 3.2 and 3.4 μg ml^{-1}, respectively. The other constituents of this plant were inactive.[9] In 2007, Choi and coworkers showed cytotoxic activity of the benzo[*c*]phenanthridine-type alkaloid corynoline (**10**) from *C. incisa* against A549, SK-OV-3, SK-MEL-2, and HCT-15 with ED_{50} values ranging from 5.27 to 6.14 μM. Compounds **11** and **12** from this plant with benzo[*c*]phenanthridine-type structures exhibited weaker cytotoxicity with ED_{50} values greater than 30 μM.[10]

As shown in Table 1, the strongest cytotoxic activities were observed in the protoberberine and aporphine classes. One of cytotoxicity mechanisms of CA (protoberberine class) have been studied by Cheng *et al.* in 2008. They found that berberine (**8**) and its analogues (**13**, **16–19**) possessed topoisomerase I inhibitory activities. However, among the tested compounds, compound **44**, a morphinadienone alkaloid, and compound **68** showed the best topoisomerase I inhibitory activity.[11]

In Table 1, IC_{50} values of CA were summarized. The IC_{50} values less than 30 μM (or μg ml^{-1}) were considered in the table as significant cytotoxic activities. It should be pointed out, however, inter-lab variation on the IC_{50} values is probable, and should be considered for a better comparison.

In total, aporphine, benzylisoquinoline and protoberberine scaffolds of CA showed a higher cytotoxicity rather than other scaffolds. A typical example is berberine (**8**) with a protoberberine skeleton that showed a remarkable cytotoxicity on a wide range of cancer cell lines (Table 1). Many papers have been published regarding the anticancer properties of berberine (**8**) and its molecular targets. But the biological activities of berberine were not limited to anticancer activity. This compound had a large number of biological targets in the body and showed various biological properties. Because of limits on the length of this review, the subject of the biological/pharmacological activities of berberine (**8**) is not expounded, and readers are referred to two recently published reviews on the biological properties of berberine (**8**).[11-13]

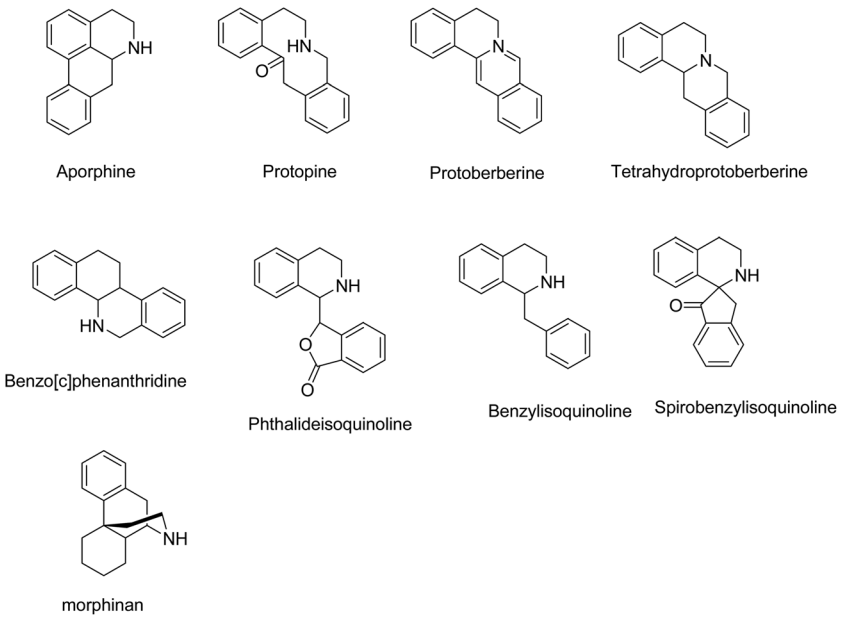

Fig. 1 Chemical structures of different classes of isoquinoline alkaloids from *Corydalis* species.

Multidrug resistance is also a major cause of failure in cancer chemotherapy regimens and finding new scaffolds of natural and synthetic compounds is a hot topic in current multidrug resistance research.[21] Recently, some of isoquinoline alkaloids showed promising activity against MDR.[22,23]

Glaucine (**40**), an aporphine-type alkaloid from *C. yanhusuo*, competitively inhibited two transporters involved in MDR named P-glycoprotein (P-gp) and multidrug resistance-associate protein 1 (MRP1). In MCF-7/ADR cancer cell line, 12.5 μM of glaucine (**40**) reduced the IC_{50} values of adriamycin and mitoxantrone from 41 and 24.19 μg ml^{-1} to 12.6 and 2.11 μg ml^{-1}, respectively.[22]

Chelidonine (**88**), a benzo[*c*]phenanthridine-type alkaloid from *Chelidonium majus*, inhibited P-gp/MDR1 activity in Caco-2 cell line. An alkaloid extract of this plant showed similar results and further identification of alkaloids exhibited berberine (**8**), coptisine (**15**), stylopine (**59**) and protopine (**27**) as major components of alkaloid extract. Mechanistic studies revealed that chelidonine (**88**) and the alkaloid extract (50 μg ml^{-1}) significantly decrease the mRNA levels of P-gp/MDR1, MRP1 and breast cancer resistance protein (BCRP).[23]

2.2. Acetylcholinesterase (AChE) and butyrylcholinesterase (BChE) inhibitory effects

Acetylcholine (ACh) as a neurotransmitter plays an important role in the peripheral and central nervous systems. According to the cholinergic hypothesis, Alzheimer's disease (AD) as the most predominant cause of dementia in the elderly, is a result of decreased levels of ACh in the cerebral cortex. To date, inhibition of AChE and BChE (the enzymes involved in the breakdown of acetylcholine) is the most common approach for treatment of AD. Galantamine from *Galanthus wornowii* is among the first FDA approved drugs for treatment of AD and acts as an AChE inhibitor (AChEI). Natural products are one of the most valuable sources for treatment of AD.[24,25] In Danish and Chinese folk medicines many *Corydalis* species have been used as memory enhancers[26,27] and modern pharmacological experiments have confirmed this effect.

In 2004, Orhan *et al.* demonstrated that chloroform : methanol (1 : 1) extract of *Corydalis solida* subsp. *solida* inhibited AChE with 87.56% inhibitory effect at 1 mg ml^{-1}.[28] In 2006, Adsersen *et al.* reported AChEI properties of aqueous and methanolic extract of 11 plants, used in Danish folk medicine as memory enhancers and *Corydalis* species showed the most potent AChE inhibitory effects.[26] Methanolic extracts of tubers of *C. solida* (L.) Swartz ssp. *slivenensis* and *C. intermedia* (L.) Merat at concentrations of 0.1 mg ml^{-1} resulted in 97% of the enzyme inhibition. In addition, methanolic extracts of tubers of *C. solida* (L.) Swartz ssp. *laxa* and *C. cava* (L.) Schw. et K. at concentration of 0.1 mg ml^{-1} showed 96 and 92% of the enzyme inhibition, respectively.[26] Regarding this ethnobotanical relevance, AChE inhibitory effects of isolated alkaloids from several *Corydalis* species have vastly been investigated. Among these alkaloids, dehydrocorydaline (**14**), coptisine (**15**), berberine (**8**), jatrorrhizine (**20**), and palmatine (**13**), from *C. yanhusuo*, showed potent AChE inhibitory effects with IC_{50} values of 0.62, 1.01, 0.47, 2.08 and 0.74 μM, respectively.[29] Recently, a review on the potential of berberine (**8**) to combat AD was published.[30] Berberine (**8**) combats AD by inhibition of AChE, BChE and monoamine oxidase, by reduction of amyloid-β peptide level, and by lowering cholesterol level.[30] (+)-Canadaline (**9**), canadine (**53**), bulbocapnine (**50**) and corydaline (**52**) from *C. cava*[31,32] showed moderate activity against AChE, while tetrahydropalmatine (**54**) and protopine (**27**) from *C. yanhusuo* were inactive.[29] More information about alkaloids from *Corydalis* species with AChE and BChE inhibitory effects are summarized in Table 3.

In spite of extensive *in vitro* studies on AChE inhibitory effects of CA, *in vivo* studies are limited. Kim *et al.* (1999) found

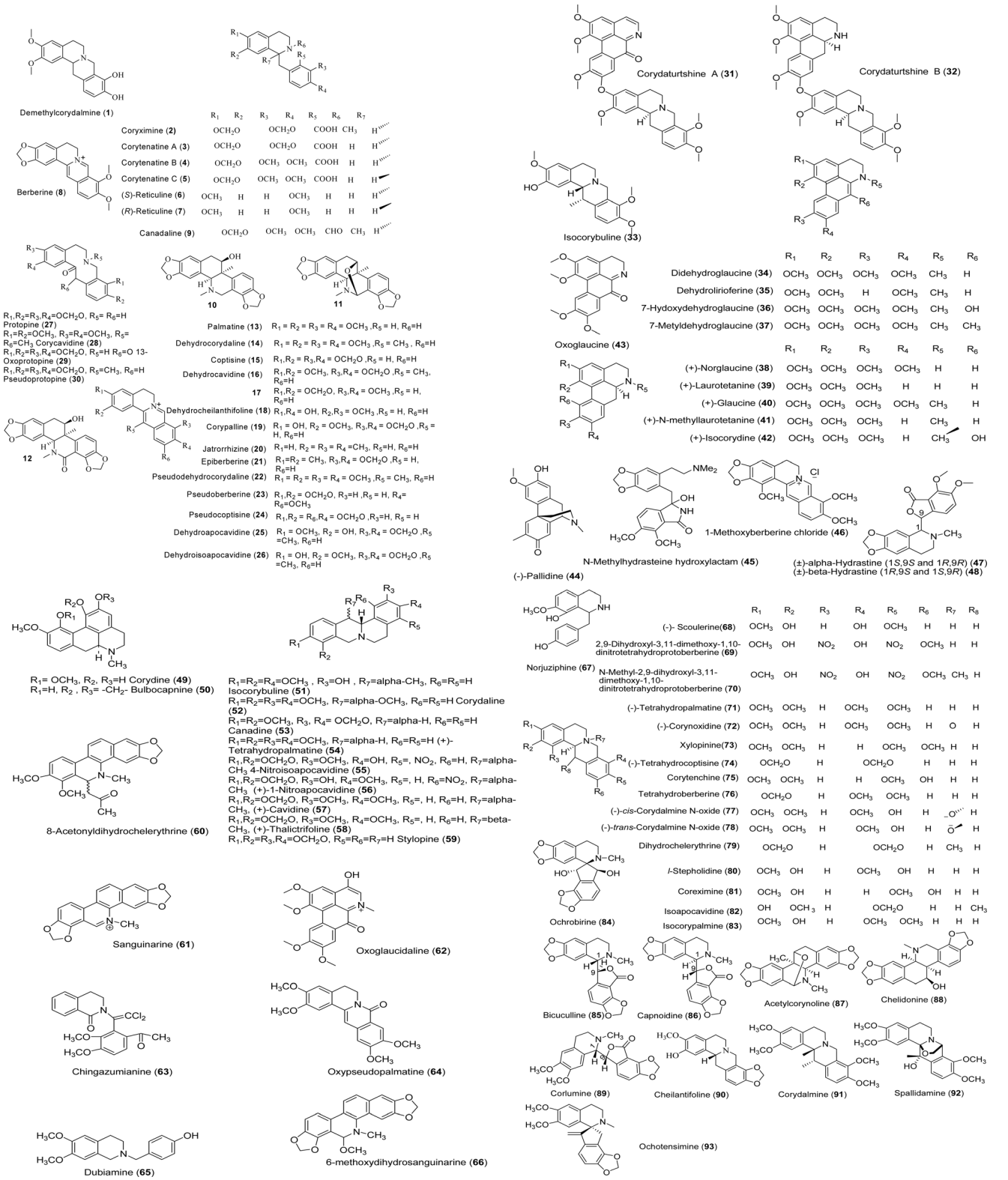

Fig. 2 Chemical structures of biologically active CA.

that protopine (**27**) from *C. ternata* alleviates scopolamine-induced memory impairment in mice.[33] In 2008, Bae *et al.* screened plants used in Korean folk medicine as memory enhancers and they found that *C. turtschaninovii* significantly inhibited AChE activity. Further studies led to the isolation and characterization of 16 alkaloids. Pseudoberberine (**23**) was tested for anti-amnesic activity in mice. The study revealed that pseudoberberine (**23**) (5.0 mg kg^{-1}) significantly reversed

Table 1 Cytotoxic activities of isoquinoline alkaloids from *Corydalis* species against different cancer cell lines. Alkaloids with IC_{50} values greater than 30 μM have not shown in this table

No.	Chemical class	IC_{50}/ED_{50} (cancer cell line)	Reference
1	Protoberberine	8.34 μM (A549), 5.14 μM (SK-OV-3), 7.87 μM (SK-MEL-2), 2.86 μM (HCT-15)	8
2	Benzylisoquinoline	27.75 μM HCT-15	8
3	Benzylisoquinoline	23.29 μM HCT-15	8
4	Benzylisoquinoline	24.58 μM HCT-15	8
5	Benzylisoquinoline	27.63 μM HCT-15	8
6	Benzylisoquinoline	29.44 μM HCT-15	8
7	Benzylisoquinoline	26.83 μM HCT-15	8
8	Protoberberine	6.27 μM (A549), 16.44 μM (SK-OV-3), 13.76 μM (SK-MEL-2), 16.59 μM (HCT-15), 3.2 μg ml (HT-1080), 3.4 μg ml^{-1} (SNU-638), 3.1 μg ml^{-1} (HepG2), 15.2 μg ml^{-1} (Hep3B), 3.3 μg ml^{-1} (SK-Hep1), 13.9 μg ml^{-1} (PLC/PRF/5), 14.1 μg ml^{-1} (K562), 9.0 μg ml^{-1} (U937), 7.9 μg ml^{-1} (P3H1), 0.6 μg ml^{-1} (Raji), 20.0 μM (HL-60), 28.7 μM (HEp-2), 19.6 μM (MCF-7), 0.0149 μg ml^{-1} (B16)	8, 9 and 14–17
10	Benzo[*c*]phenanthridine	5.27 μM (A549), 5.82 μM (SK-OV-3), 5.59 μM (SK-MEL-2), 6.14 μM (HCT-15)	10
11	Benzo[*c*]phenanthridine	17.05 μM (SK-MEL-2)	10
13	Protoberberine	4 μg ml^{-1} (hydrogen iodide salt, KB)	18
14	Protoberberine	4.5 μg ml^{-1} (P388), 4 μg ml^{-1} (L1210), 28.76 μM (SK-MEL-2)	19 and 20
15	Protoberberine	2.97 μg ml^{-1} (P388), 0.36 μg ml^{-1} (HCT-8), 0.32 μg ml^{-1} (A549), 3.5 μg ml^{-1} (HepG2), 5.4 μg ml^{-1} (Hep3B), 1.4 μg ml^{-1} (SK-Hep1), 6.6 μg ml^{-1} (PLC/PRF/5), 7.2 μg ml^{-1} (K562), 1.5 μg ml^{-1} (U937), 10.9 μg ml^{-1} (P3H1), 0.6 μg ml^{-1} (Raji)	14 and 19
31	Aporphine + tetrahydroprotoberberine	12.9 μM (A549), 8.75 μM (SK-OV-3), 12.28 μM (SK-MEL-2), 14.21 μM (HCT-15)	20
32	Aporphine + protoberberine	10.08 μM (SK-MEL-2), 29.31 μM (HCT-15)	20
33	Tetrahydroprotoberberine	22.43 μM (SK-OV-3), 29.61 μM (SK-MEL-2), 15.55 μM (HCT-15)	20
34	Aporphine	4.51 μM (A549), 4.53 μM (SK-OV-3), 4.21 μM (SK-MEL-2), 4.76 μM (HCT-15)	20
35	Aporphine	12.9 μM (A549), 4.53 μM (SK-OV-3), 4.21 μM (SK-MEL-2), 4.76 μM (HCT-15)	20
36	Aporphine	15.94 μM (A549), 12.29 μM (SK-OV-3), 5.99 μM (SK-MEL-2), 15.15 μM (HCT-15)	20
37	Aporphine	15.90 μM (SK-OV-3), 20.86 μM (SK-MEL-2), 27.70 μM (HCT-15)	20
38	Aporphine	19.38 μM (SK-OV-3), 21.97 μM (HCT-15)	20
39	Aporphine	26.19 μM (SK-OV-3), 26.64 μM (SK-MEL-2), 18.60 μM (HCT-15)	20
40	Aporphine	26.76 μM (A549), 21.57 μM (SK-OV-3), 20.39 μM (SK-MEL-2), 18.63 μM (HCT-15)	20
41	Aporphine	12.61 μM (A549), 13.64 μM (SK-OV-3), 11.21 μM (SK-MEL-2), 15.06 μM (HCT-15)	20
43	Aporphine	12.21 μM (A549), 7.19 μM (SK-OV-3), 15.40 μM (SK-MEL-2), 13.10 μM (HCT-15)	20

cognitive impairments induced by scopolamine (1.0 mg kg^{-1}, i.p.) in mice.[34]

2.2.1. Structure–AChE inhibitory activity relationship. As it was mentioned in many studies conducted on AChE inhibitory effect of alkaloids from *Corydalis* species,[29,34,37] protoberberine type alkaloids are the most potent inhibitors of AChE. Aromatization of ring C, substitution of OH with OMe at aromatic rings and the presence of quaternary nitrogen increased the activity (Fig. 3). For example, palmatine (**13**) and tetrahydropalmatine (**54**) had similar structures, but aromatization of ring C and the presence of quaternary nitrogen in palmatine (**13**) decreased the IC_{50} value from 268 μM to 0.74 μM. In the case of jatrorrhizine (**20**), substitution of two methoxy groups with OH groups increased the IC_{50} value from 0.62 μM in dehydrocorydaline (**14**) to 2.08 μM.[29] Hence, the protoberberine scaffold can be a good lead structure for AChE inhibitory studies in the future.

2.3. Anti-platelet aggregation

Platelets are blood cells that play a crucial rule in haemostasis, but some pathological conditions including thrombosis and inflammation are attributed to these cells. These pathological conditions lead to cardiovascular disease. Hence, targeting platelets is an effective approach for prevention and treatment of some cardiovascular diseases. Although current anti-platelet aggregation (PA) drugs show serious side effects including dangerous bleeding, finding new drugs in this field is still

Table 2 *In silico* physico-chemical properties of selected CA. For oral bioavailability, NROT, HBA, HBD, *c* log *P*, PSA and MW should less than 10, 10, 5, 5, 140 and 500, respectively. log $D_{5.5}$ is defined as log *P* at pH 5.5 (pH of the small intestine) and is a more appropriate measure than log *P* where oral drug absorption occurs. All physiochemical parameters used for the prediction of drug-likeness were calculated using Marvin Sketch (version 5.11, academic package, Chem Axon)

No.	MW	NROT	HBA	HBD	PSA	*c* log *P*	log $D_{5.5}$	Bioavailability prediction
1	327	2	5	2	62.18	2.86	1.98	✓
2	369	3	7	1	77.46	2.93	0.20	✓
3	355	3	7	2	86.25	2.55	0.08	✓
4	371	5	7	2	86.25	2.61	0.14	✓
5	371	5	7	2	86.25	2.61	0.14	✓
6	283	4	3	1	30.49	3.33	0.17	✓
7	283	4	3	1	30.49	3.33	0.17	✓
8	336	2	4	0	40.80	−1.28	−1.28	✓
9	369	5	6	0	57.23	3.05	1.39	✓
31	660	8	10	0	97.81	5.62	5.33	✗

Table 3 AChE and BChE inhibitory effects of *Corydalis* alkaloids. Alkaloids with weak activities (IC_{50} > 20 μM) were not shown in this table

	IC_{50} (μM) ± SD			
Compound	AChE	BChE	Plant source	Reference
Canadine (**53**)	12.4 ± 0.9		*C. cava*	31
Protopine (**27**)	16.1	ND	*C. speciosa*	35
	14.5 ± 0.5		*C. turtschaninovii*	34
	50		*C. ternata*	33
Berberine (**8**)	0.47 ± 0.01	ND	*C. yanhusuo*	29
	1.87 ± 0.48	>200 μM	*C. saxicola*	36
	3.3		*C. speciosa*	35
	4.7 ± 0.2			
Palmatine (**13**)	0.74 ± 0.06	ND	*C. yanhusuo*	29
	2.20 ± 0.46	>200 μM	*C. saxicola*	36
	5.8		*C. speciosa*	35
	10.4 ± 0.4		*C. turtschaninovii*	34
Jatrorrhizine (**20**)	2.08 ± 0.09	ND	*C. yanhusuo*	29
Coptisine (**15**)	1.01 ± 0.03	ND	*C. yanhusuo*	29
Dehydrocorydaline (**14**)	0.62 ± 0.05	ND	*C. yanhusuo*	29
Dehydrocavidine (**16**)	9.92 ± 0.23	>200 μM	*C. saxicola*	36
2,9-Dihydroxyl-3,11-dimethoxy-1, 10-dinitrotetrahydroprotoberberine (**69**)	8.77 ± 0.20	>200 μM	*C. saxicola*	36
(+)-1-Nitroapocavidine (**56**)	1.70 ± 0.31	>200 μM	*C. saxicola*	36
(+)-Thalictrifoline (**58**)	14.50 ± 0.20	>200 μM	*C. saxicola*	36
Sanguinarine (**61**)	1.93 ± 0.01	>200 μM	*C. saxicola*	36
Stylopine (**59**)	15.8 ± 1.2	ND	*C. turtschaninovii*	34
Epiberberine (**21**)	6.5 ± 0.5	ND	*C. turtschaninovii*	34
Pseudodehydrocorydaline (**22**)	8.4 ± 0.5	ND	*C. turtschaninovii*	34
Pseudocoptisine (**24**)	4.3 ± 0.3	ND	*C. turtschaninovii*	34
Pseudoberberine (**23**)	4.5 ± 0.2	ND	*C. turtschaninovii*	34
Tacrine	0.16 ± 0.01	ND		
Galantamine	4.0 ± 1.4	1.4 ± 0.2		

necessary.[38] Interestingly, some *Corydalis* species have traditionally been used for the treatment of thromboembolism and can be used for finding PA inhibitors.[39]

In 1989, Wu *et al.* found that 100 μg ml^{-1} of protopine (**27**) isolated from *C. tashiroi* and *C. pallida* inhibited platelet aggregation caused by 20 μM of adenosine 5′-diphosphate (ADP), 100 μM of arachidonic acid (AA), 2 ng ml^{-1} of platelet activation factor (PAF) and 10 μg ml^{-1} of collagen, however, was unable to inhibit platelet aggregation caused by 0.1 U ml^{-1} of thrombin in rabbit platelet suspension.[39] In 1994, Wei *et al.* investigated anti-platelet aggregation of tetrahydroberberine (**76**) isolated from *C. ambigua*. Tetrahydroberberine (**76**) (0.50 mg ml^{-1}) showed more than 80% inhibition in PA induced by AA (0.78 mmol l^{-1}), ADP (2 μmol l^{-1}) and collagen (30 μg ml^{-1}).[40] In 2000, Lin *et al.* isolated three known alkaloids with one novel chlorinated alkaloid, chingazumianine (**63**), from

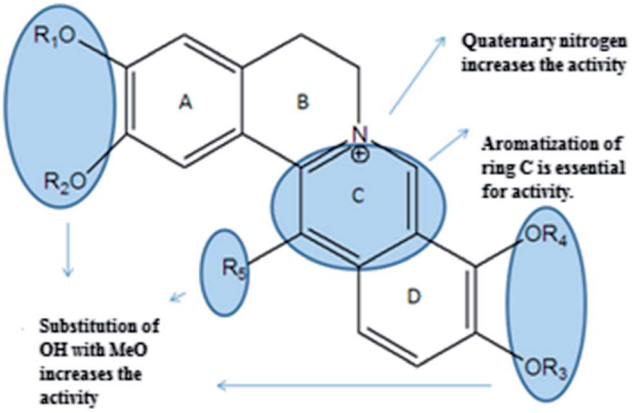

Fig. 3 Structure–AChE inhibitory activity relationship of protoberberine alkaloids from *Corydalis* species.

C. koidzumiana and tested them for inhibition of human PA induced by ADP (20 μM), collagen (10 μg ml^{-1}) and adrenaline (5 μM). In that study, 300 μM of protopine (**27**) completely inhibited PA induced by ADP, collagen and adrenaline. Tetrahydropalmatine (**71**) (300 μM) showed 10% inhibition of PA induced by ADP and collagen, and (83.9%) inhibition of PA caused by adrenaline. Almost similar results were observed for palmatine (**13**). In a lower concentration of 75 μM, these three alkaloids inhibited the secondary-phase aggregation, but not the primary-phase aggregation caused by adrenaline. These findings revealed that inhibition of PA by these alkaloids may be due to inhibition of thromboxane A_2 formation.[41] In 2001, Chen *et al.* evaluated anti-platelet aggregation activity of fourteen alkaloids isolated from *C. tashiroi*. 6-Methoxydihydrosanguinarine (**66**) (50 μg ml^{-1}) and norjuziphine (**67**) (100 μg ml^{-1}) were the most potent PA inhibitors and completely inhibited PA caused by AA (100 μM), collagen (10 μg ml^{-1}) and PAF (2 ng ml^{-1}). (−)-*cis*-Corydalmine *N*-oxide (**77**) and (−)-*trans*-corydalmine *N*-oxide (**78**) (100 μg ml^{-1}) also showed 90% inhibitory activities.[42]

Wu and coworkers studied forty-one isoquinoline alkaloids with different structures for their PA inhibitory activities, however, they could not find any consistency between the structures and PA inhibitory activities of CA.

Overall, some of CA showed potent PA inhibitory activity. For example, 6-methoxydihydrosanguinarine (**66**) showed more potent PA inhibition than aspirin.[42] Moreover, aspirin can only inhibit PA induced by AA, while CA inhibit PA caused by different inducers. Regarding the drug-likeness of CA, it seems that anti-platelet *Corydalis* alkaloids have a good potential for being future anti-platelet drugs.

2.4. Anti-inflammatory properties

Many natural products, particularly alkaloids and terpenes, possessed anti-inflammatory properties. Anti-inflammatory natural products have been fully reviewed by Gautam and Jachak.[43] Barbosa-Filho *et al.* reviewed anti-inflammatory alkaloids isolated in the period of 1907 to 2000.[44] They reviewed 171 alkaloids, of which, 137 alkaloids showed activity. Isoquinoline alkaloids were the most studied chemical structures.[44] The first report on anti-inflammatory alkaloids from *Corydalis* species dates back to 1994, a study of anti-inflammatory activities of methanolic extract and alkaloids from *C. turtschaninovii*.[45] Oral administration of 200 or 500 mg kg^{-1} of methanolic extract from *C. turtschaninovii* in rats inhibited increasing vascular permeability induced by acetic acid, and reduced acute paw edema induced by compound 40/80 (condensation product of *N*-methyl-*p*-methoxyphenethylamine with formaldehyde) and carrageenin. Adjuvant-induced edema in arthritis rats was suppressed by methanolic extract. Also, methanolic extract of *C. turtschaninovii* which contains dehydrocorydaline (**14**), D-glaucine (**40**) and L-tetrahydrocoptisine (**74**) inhibited histamine release from mast cells in rats.[45] The same group investigated anti-inflammatory activities of dehydrocorydaline (**14**) from *C. turtschaninovii* on acute inflammatory models. Orally administered dehydrocorydaline (**14**) (0.125 mmol kg^{-1}) inhibited increasing vascular permeability induced by acetic acid, inflammation induced by serotonin and bradykinin, ear edema induced by AA, prostaglandine E_2 and leukotriene C_4, and acute paw edema induced by carrageenin. However, dehydrocorydaline (**14**) was unable to inhibit the edema induced by histamine.[46] Tetrahydropalmatine (**71**), an isoquinoline alkaloid from *C. yanhusuo* and many other plants, possess anti-inflammatory properties. Oh *et al.* studied effects of this compound on lipopolysaccharide-induced interleukin (IL)-8 production in the human monocytic cell line THP-1. Pretreatment of THP-1 cells with 0.2, 1 or 2 mM of tetrahydropalmatine (**71**) significantly inhibited IL-8 secretion induced by lipopolysaccharide. Their studies revealed the inhibitory mechanism of tetrahydropalmatine (**71**) on IL-8 formation *via* the mitogen-activated protein kinases (MAPKs) and the NF-κB signaling pathway inhibition.[47] Pseudocoptisine (**24**) another isoquinoline alkaloid isolated from *C. turtschaninovii* inhibited pro-inflammatory mediators in lipopolysaccharide-stimulated murine macrophage RAW 264.7 cells. Pseudocoptisine (**24**) (30, 60 and 90 μM) significantly inhibited the production of IL-6 and TNF-α and reduced the levels of nitric oxide (NO) and cyclooxygenase 2. The mechanism of action of pseudocoptisine (**24**) involved in suppression of the production and mRNA expressions of inflammatory cytokines and inhibition of NF-κB.[48] In another study, effects of 11 alkaloids isolated from *C. impatiens* on inhibition of TNF-α and NO production have been evaluated. Ochotensimine (**93**) showed the most potent TNF-α and NO inhibition with IC_{50} values of 1.64 and 1.14 μM, respectively.[49] Overall, it seems that anti-inflammatory activity of CA is mainly due to inhibition of mediators and enzymes involving in inflammation process.

2.5. Antiviral activities

In 2006, Orhan *et al.* studied antiviral activity of 33 isoquinoline alkaloids belonging to *Fumaria* and *Corydalis* on *Herpes simplex* (HSV) and *Parainfluenza* (PI-3) viruses. Protopine (**27**) was the most active alkaloid against PI-3 with minimum cytophatogenic effect inhibitory concentration of 1 μg ml^{-1} and maximum of

32 μg ml^{-1} in Vero cells which was comparable with oseltamivir as the reference with minimum and maximum concentrations of 0.25 and 32 μg ml^{-1}, respectively.[50] However, (−)-stylopine (**59**) and berberine (**8**) were inactive against both viruses. Also, (+)-bulbocapnine (**50**), palmatine (**13**), dehydrocorydaline (**14**) and dehydrocavidine (**16**) were totally inactive against HSV.

Traditional background of *C. saxicola* in treatment of hepatitis has attracted scientists' attention to study anti-hepatitis B virus (HBV) activity of this plant. A study on antiviral activity of this plant was conducted by Wu and co-workers. Dihydrochelerythrine (**79**) exhibited the most potent inhibitory activities against hepatitis B surface antigen (HBsAg) and hepatitis B e antigen (HBeAg) secretion with an IC_{50} value < 0.05 μM in Hep G 2.2.15 cell line while lamivudine as positive control showed IC_{50} values of 15.37 and 44.85 μM against HBsAg and HBeAg secretion, respectively.[51] Dihydrochelerythrine (**79**) gave hope for finding new anti-HBV drugs in the future.[52] Similar experiments were conducted by Li *et al.* Dehydrocavidine (**16**), dehydroapocavidine (**25**) and dehydroisoapocavidine (**26**) (15.6–250 μg ml^{-1}) showed anti-HBsAg activity with average inhibition percentage of 53%, 59% and 54% and anti-HBeAg activity with average inhibition percentage of 41%, 43% and 43%, respectively.[53] Recently, Zeng *et al.* evaluated anti-HBV virus activity of dehydrocheilanthifoline (**18**). After six days of treatment, dehydrocheilanthifoline (**18**) showed an inhibitory effect on HBsAg and HBeAg secretion from Hep G 2.2.15 cell line with IC_{50} values of 15.84 and 17.12 μM, respectively. In addition, they found that dehydrocheilanthifoline (**18**) acted through inhibition of extracellular DNA, intracellular DNA and covalently closed circular DNA.[54] However, further studies are needed to understand the exact mechanism of action of this alkaloid.

2.5.1. Structure–antiviral activity relationship. It seems that for anti-HSV activity, protoberberine-type alkaloids display higher antiviral activities than the other scaffolds and the presence of a quaternary nitrogen atom decreases the activity.[50]

Reviewing alkaloids isolated from *C. saxicola* revealed that protoberberine-type alkaloids are the most active alkaloid against HBV. The study showed that the introduction of quaternary nitrogen increased the anti-HBV activity. Interestingly, the presence of methyl group at C13 position of protoberberine-type alkaloids is vital for the activity (Fig. 4). For example, dehydroisoapocavidine (**26**) and berberine (**8**) have similar structures, however, dehydroisoapocavidine (**26**) had an extra methyl group at C13 position and exhibited potent anti-HBV activity, while berberine (**8**) was inactive.[53]

Further studies are necessary to unravel more aspects of structure–antiviral activity relationship of isoquinoline alkaloids, particularly protopine- and protoberberine-type scaffold.

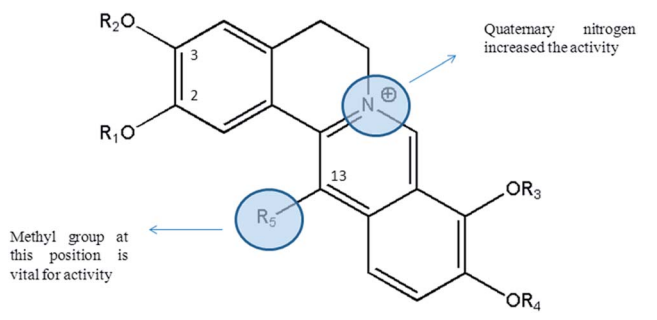

Fig. 4 Structure–antiviral activity relationship of protoberberine alkaloids. Protoberberine structure was the best scaffold for anti-HBV properties.

2.6. Cardiovascular activities

2.6.1. Inhibition of low density lipoprotein (LDL) oxidation. Lee and co-workers isolated nine alkaloids from *C. turtschaninovii* and studied their inhibition activity against LDL oxidation. (±)-Demethylcorydalmine (**1**), (+)-isocorydine (**42**) and (+)-stylopine (**59**) were the most active alkaloids with IC_{50} values of 2.1, 2.4 and 2.0 μM, respectively, while probucol as a positive control showed weaker activity than the alkaloids with an IC_{50} value of 6.8 μM.[55] This study revealed that some CA might have a potential of LDL protection against oxidative stress, which is a key process in atherosclerosis and other cardiovascular diseases. Although, the aforementioned alkaloids (except demethylcorydalmine, **1**) contain no phenolic hydroxyl required for direct radical scavenging activity, it seems that these alkaloids exert its antioxidant properties against LDL oxidation *via* direct ROS and RNS scavenging activity as well as binding to catalyzing metal ions and reducing the concentration of metal ions in lipid peroxidation. The mentioned mechanism of action has been proved for berberine (**8**) in previous studies.[56]

2.6.2. Antihypertensive activities. Chu *et al.* studied antihypertensive activity of DL-tetrahydropalmatine (**54**), a protoberberine-type alkaloid isolated from *C. racemosa*.[57] DL-Tetrahydropalmatine (**54**) (5, 10, 15 and 20 mg kg^{-1}) significantly reduced the plasma concentration of noradrenaline and mean blood pressure after intraperitoneal (i.p.) injection in spontaneously hypertensive rats. In addition, intracerebroventricular administration of 0.05, 0.1, 0.5 and 1 mg 10 μl^{-1} of DL-tetrahydropalmatine (**54**) reduced the plasma concentration of noradrenaline and mean blood pressure. These findings suggested the mechanism of action of DL-tetrahydropalmatine (**54**) through decreasing the sympathetic tone.[58] A more detailed mechanistic study was carried out by Lin and co-workers. They revealed that DL-tetrahydropalmatine (**54**) acted through the 5-HT_2 and/or D_2-receptor antagonism in the hypothalamus to decrease the blood pressure.[58] Also, another study conducted on total alkaloids of *Corydalis* plants, showed the reduction of blood pressure, and relaxation of aortic muscle precontracted by noradrenaline in rabbits.[59] Unfortunately, some severe cardiac and neurological toxic effects were reported after consumption of *C. racemosa* and these toxic effects are attributed to DL-tetrahydropalmatine (**54**), which had similar toxic effects possibly by inhibition of calcium efflux.[60]

2.6.3. Cardioprotective activities. Xuan *et al.* studied protective effects of L-tetrahydropalmatine (**71**), L-stepholidine (**80**) and tetrahydroberberine (**76**) on experimental myocardial infarction (MI) in the left coronary artery ligated-rats.[61] L-Stepholidine (**80**) showed the best protective activity. Intravenous administration of 2.5 mg kg^{-1} of this alkaloid led to rapid

reduction in systolic blood pressure and diastolic blood pressure with values of 39.5% and 48.5%, respectively.[61]

In traditional medicine of China, Korea and Japan, *C. yanhusuo* has been used in patients suffered from cardiovascular diseases.[62] The first report on beneficial effects of *C. yanhusuo* in heart failure after myocardial infarction dates back to 2006.[62] Coronary artery ligated-rats were treated 8 weeks by orally administered ethanolic extract of *C. yanhusuo*. At doses 100 and 200 mg kg^{-1}, *C. yanhusuo* significantly decreased left ventricular end-diastolic pressure from 19 ± 5 mmHg to 12 ± 2 and 9 ± 3 mmHg, respectively, as well as reduction in the plasma concentration of atrial natriuretic peptide. MI leads to hypertrophy of ventricles and increasing in weights of both ventricles and lungs. Treatment with 200 mg kg^{-1} of *C. yanhusuo* significantly reduced the weights of ventricles and lungs as well as infarct size in rats.[62] The molecular mechanism of action of *C. yanhusuo* was provided by Ling and co-workers. They suggested that *C. yanhusuo* acted by the inhibition of myocardial apoptosis through modulation of the Bcl-2 family and this may provide the basis for novel therapeutic strategy in MI.[63] Another study concerned the salutary effects of *C. yanhusuo* extract on cardiac hypertrophy due to pressure overload induced by transverse abdominal aorta constriction in rats.[64] 200 mg kg^{-1} of *C. yanhusuo* significantly reduced cardiac hypertrophy and heart rate in rats compared with vehicle-treated animals.[64] The study was not led to the isolation and purification of bioactive alkaloids.

Further studies on L-tetrahydropalmatine (**71**), an active constituent of *C. yanhusuo*, showed cardioprotective activity in myocardial ischaemia-reperfusion injury induced by 30 min occlusion of the left anterior descending coronary artery and 6 h reperfusion in rats.[65] L-Tetrahydropalmatine (**71**) (20 and 40 mg kg^{-1}) reduced the infarct area/risk area ratio from 36.3 ± 3.9% in case of vehicle to 20.5 ± 2.2 and 18.6 ± 2.2%, respectively, while 10 mg kg^{-1} of this alkaloid was not effective. L-Tetrahydropalmatine (**71**) at doses of 10, 20 and 40 mg kg^{-1} significantly increased ejection fraction and fractional shortening when compared with sham-operated group. Moreover, mechanism of action of L-tetrahydropalmatine (**71**) involved in the reduction of iNOS-mediated NO biosynthesis and TNF-α.[65]

2.7. Antiplasmodial activities

In Bhutanese traditional medicine, some of *Corydalis* species have been used as febrifuge and antimalarial drugs including *C. bhutanica, C. crispa* and *C. dubia*.[66] Recent efforts led to the isolation of some isoquinoline alkaloids from these *Corydalis* species with highly significant antimalarial activities. The extract of *C. bhutanica* did not show antiplasmodial activity even at the highest concentration of 25 μg ml^{-1}. However, the chloroform extract of *C. crispa* showed significant activity against *Plasmodium falciparum* strains TM4/8.2 (a wild type chloroquine and antifolate sensitive strain) and K1CB1 (multidrug resistance strain) with IC_{50} values of 2.11 μg ml^{-1} and 1.89 μg ml^{-1}, respectively. Similarly, the chloroform extract of *C. dubia* exhibited strong activity against TM4/8.2 and K1CB1 strains with IC_{50} values of 2.21 μg ml^{-1} and 2.84 μg ml^{-1}, respectively.[66] Further investigations on *C. dubia* resulted in the isolation of eight alkaloids. Among them, scoulerine (**68**), cheilanthifoline (**63**) and protopine (**27**) showed remarkable antiplasmodial activities against TM4/8.2 strain with IC_{50} values of 1.78, 0.9 and 1.45 μg ml^{-1}, respectively.[68] Almost similar results were observed for K1CB1 strain, however, scoulerine (**68**) was the most active alkaloid with an IC_{50} value of 1.04 μg ml^{-1}. Chloroquine, which is a well-known antimalarial drug, exhibited

Table 4 Antiplasmodial activity of extracts and alkaloids of some *Corydalis* species

	Antiplasmodial IC_{50} (μg ml^{-1})		
Samples	TM4/8.2	K1CB1	Plant source(s)
MeOH extract	11.53	13.24	*C. crispa*
	6.38	9.50	*C. dubia*
	1.00	2.56	*C. calliantha*
	>6.25	>6.25	*C. bhutanica*
CH_2Cl_2 extract	12.09	11.27	*C. crispa*
	>6.25	>6.25	*C. bhutanica*
Hexane extract	14.57	15.82	*C. crispa*
	>1.56	>1.56	*C. bhutanica*
$CHCl_3$ extract	2.11	1.89	*C. crispa*
	2.21	2.84	*C. dubia*
	>6.25	>6.25	*C. bhutanica*
H_2O extract	>25	>25	*C. bhutanica, C. dubia and C. crispa*
Crude alkaloid extract	0.33	0.63	*C. calliantha*
Protopine (**27**)	1.45	1.38	*C. crispa*
13-Oxoprotopine (**29**)	>4.6	>4.6	*C. crispa*
Stylopine (**59**)	>4.0	>4.0	*C. crispa*
Coreximine (**81**)	5.56	6.87	*C. crispa*
Scoulerine (**68**)	1.78	1.04	*C. dubia*
Cheilanthifoline (**90**)	0.90	1.22	*C. calliantha*

IC_{50} values of 0.010 and 0.089 μg ml^{-1} against TM4/8.2 and K1CB1 strains, respectively.[66]

Based upon the results from previous studies,[66] Wangchuk and co-workers isolated nine isoquinoline alkaloids from *C. crispa*. Among the tested alkaloids coreximine (**81**) exhibited a potent antimalarial activity against TM4/8.2 and K1CB1 strains of *P. falciparum* with IC_{50} values of 5.56 and 6.87 μg ml^{-1}, respectively.[69] For more information please see Table 4.

2.7.1. Structure–antiplasmodial activity relationship. From different scaffolds of isoquinoline alkaloids, which have been tested for antimalarial activity, including tetrahydroprotoberberine, benzylisoquinoline, phthalideisoquinoline and spirobenzylisoquinoline, only tetrahydroprotoberberine alkaloids showed remarkable activities. Scoulerine (**68**), cheilanthifoline (**90**) and coreximine (**81**) were the most active alkaloids. It seems that presence of methylenedioxy-isoquinoline moiety in tetrahydroprotoberberine alkaloids removes the activity, while formation of methylenedioxy group at benzyl moiety increases the activity. For example, cheilanthifoline (**90**) with additional methylenedioxy group at benzyl moiety showed more potent antimalarial activity when compared with scoulerine (**68**). Stylopine (**59**) with methylenedioxy-isoquinoline moiety was inactive.[67–69]

2.8. Miscellaneous activities

2.8.1. Sedative and anticonvulsant activities. It is well established that some of phthalideisoquinoline alkaloids with 1*S*, 9*R* isomeric form compete with GABA for its receptor, while other isomeric forms are ineffective.[70,71] Bicuculline (**85**) is a well-known example of CA that inhibits the GABA receptor.[72] Isocoryne (β-hydrastine, **48**), a phthalideisoquinoline alkaloid isolated from *C. pseudoadunca*, inhibited the GABA induced currents of rat neurons in a similar manner with bicuculline (**85**) and corlumine (**89**), but with a stronger inhibition activity.[72] DL-Tetrahydropalmatine (**54**), another alkaloid isolated from many *Corydalis* species, showed inhibitory activity on epileptic attack in rats induced by picrotoxin.[73,74] Picrotoxin acted by attenuation of GABA mediated inhibition as well as increasing amygdaloid dopamine release.[73] Chang *et al.* demonstrated that DL-tetrahydropalmatine (**54**) acted through inhibition of amygdaloid dopamine release.[73] Concomitant administration of 30 mg kg^{-1} of DL-tetrahydropalmatine (**54**) with picrotoxin (3 and 4 mg kg^{-1} s.c.) decreased horizontal motion time, vertical motion time, clockwise turning, anticlockwise turning, total distanced traveled and freezing time in rats as compared with the control group received saline instead of DL-tetrahydropalmatine (**54**).[73] In another study, i.p. administration of 20 and 30 mg kg^{-1} of DL-tetrahydropalmatine (**54**), 30 min before daily electrical kindling stimulation, reduced the behavioral seizure score and the motion responses which normally developed during electrical kindling. Lin and co-workers suggested DL-tetrahydropalmatine (**54**) as an effective antiepileptogenic and anticonvulsant agent.[75]

In animal studies, pure alkaloids of *C. cava* showed sedative properties,[76,77] and further studies showed that this effect was induced by interaction between protoberberine alkaloids and GABAergic system.[78] Protoberberine type alkaloids influenced the binding behavior of $GABA_A$ receptor, exhibited positive cooperation in binding of [^3H] bicuculline (**85**) methochloride ([^3H] BMC) and increased the specific binding of about 21–49%. Corydaline (**52**), isoapocavidine (**82**), isocorypalmine (**83**), scoulerine (**68**) and tetrahydropalmatine (**54**) significantly increased specific binding of [^3H] BMC when compared with diazepam, and isoapocavidine (**82**) exhibited the most potent activity.[77,78]

2.8.2. Antinociceptive activities. There are only a few reports on antinociceptive activities of CA. Wang *et al.* studied the antinociceptive activity of alkaloid extract from *C. yanhusuo*.[79] They used a formalin-induced model to evaluate the antinociceptive effect of the total alkaloids of *C. yanhusuo*. Their findings revealed that nociception was significantly inhibited by a single dose of 150 mg kg^{-1} of total alkaloids. They characterized the alkaloids of the extract by HPLC-DAD-MS-MS, however, this effect was not attributed to a particular alkaloid. Two years later, Choi *et al.* studied the antinociceptive effect of *Corydalis* tuber extract using a neuropathic pain rat model.[80] They found that repeated treatment of this extract decreased mechanical allodynia (a pain due to a stimulus which does not normally provoke pain) and alleviated thermal heat hyperalgesia (an increased sensitivity to pain, which may be caused by damage to pain receptors or peripheral nerves) in the maintenance period (day 14–19). This study was also a mechanistic investigation of the antinociceptive property of a *Corydalis* extract and was not led to identification of a particular alkaloid responsible for this effect.[80] But Cao and coworkers studied the antinociceptive effect of tetrahydropalmatine (**54**). They evaluated the antinociceptive effect of tetrahydropalmatine (**54**) at doses 20, 40 and 60 mg kg^{-1} using different pain models including formalin-induced nociception, writhing test and bee venom-induced hyperalgesia. Their results suggested that tetrahydropalmatine (**54**) more effectively inhibited visceral nociception and hyperalgesia than persistent nociception.[81,82]

2.8.3. Insecticidal activities. Five alkaloids from *C. bulbosa* have been tested against *Drosophila melanogaster*, commonly known as vinegar fly. (±)-Dehydrocorydaline (**14**), (−)-tetrahydroberberine (**76**) and (−)-tetrahydrocoptisin (**74**) exhibited the most potent larvicidal activity with LC_{50} values of 0.23 μM, 0.91 μM and 1.70 μM diet concentration, respectively.[83] (−)-Tetrahydroberberine (**76**) was the most potent alkaloid with a LD_{50} value of 2.5 μg per adult fly.[83] In another study, (−)-canadine (**53**) from *C. bulbosa* showed larvicidal activity with a LC_{50} value of 0.9 μM diet concentration. The LD_{50} value was 5.0 μg per adult against adults.[84] Protoberberine type alkaloids were the most potent compounds, and the presence of methylenedioxy-isoquinoline moiety was necessary for their activities against *D. melanogaster*.[84]

Park and co-workers evaluated insecticidal activity of alkaloids from *C. turtschaninovii* against *Aphis gossypii*, which is a widely distributed pest of a variety of agricultural crops.[85] At concentration of 1000 ppm, (+)-stylopine (**59**), (+)-corydaline (**52**), demethylcorydalmine (**1**), isocorypalmine (**83**), glaucine (**40**), and pseudoprotopine (**30**) exhibited significant insecticidal activity with inhibition values of 69.7, 46.9, 68.5, 75.5, 80.2, and 78.9%, respectively.[85]

2.8.4. Antifungal activities. Ma *et al.* isolated thirteen alkaloids from *C. incisa* and *C. ambigua* and evaluated their antifungal activity against plant pathogenic fungus *Cladosporium herbarum* using bioautography method.[86] Corynoline (**10**) and acetylcorynoline (**87**) showed the best activity with minimum amount for activity of 3 μg, while other alkaloids were inactive or showed weak activities.[86] *N*-Methylhydrasteine hydroxylactam (**45**) and 1-methoxyberberine chloride (**46**) from *C. longipes* were effective individually and in 1 : 1 mixture against spore germination of some plant pathogenic fungi, including *Alternaria*, *Curvularia*, *Colletotrichum*, *Helminthosporium* and *Ustilago*.[87] Also, (±)-α-hydrastine (**47**) and (±)-β-hydrastine (**48**) from *C. longipes* exhibited remarkable activity against *Helminthosporium echinoclova*.[88]

Berberine (**8**) showed potent antifungal activity with MIC values of 5, 7.81, and 5.89 μg ml^{-1} against *Candida albicans*, *C. tropicalis*, and *C. glabrata*, respectively.[89,90] Berberine (**8**) and its derivatives have attracted considerable attention as antifungal and antibacterial agents. The authors encourage the reader to review the articles provided in this regard.[91–93]

Protopine (**27**), (+)-bulbocapnine (**50**), (−)-canadine (**53**), corydalmine (**91**) and corydaline (**52**) from *Corydalis* species showed potent antifungal activity against *Candida albicans* with an MIC value of 4 μg ml^{-1} as compared with ketoconazole as a standard compound (MIC value = 2 μg ml^{-1}).[50]

2.8.5. Antibacterial activities. Li and co-workers studied *in vitro* anti-*Helicobacter pylori* activity of 30 Chinese herbal medicines used for treating gastric ulcers in traditional medicine of China. Among them, ethanolic extract of *C. yanhusuo* showed moderate antibacterial activity with an MIC value of 60 μg ml^{-1}.[94]

The petroleum ether extract of *C. bungeana*, a constituent of Chinese traditional medicine Zi Hua Di Ding, was tested against *Bacillus subtilis* and *Pseudomonas syringae* and at a concentration of 6.25 μg ml^{-1} exhibited a significant activity against *B. subtilis*.[95] However, no alkaloids isolated from *C. bungeana* showed antibacterial activity against *B. subtilis*.[95]

A more comprehensive study on antibacterial activity of CA was conducted by Orhan and co-workers (2007).[50] Thirty three isoquinoline alkaloids from *Fumaria* and *Corydalis* species were tested against *Escherichia coli*, *Pseudomonas aeruginosa*, *Proteus mirabilis*, *Klebsiella pneumoniae*, *Acinetobacter baumannii*, *Staphylococcus aureus* and *Bacillus subtilis*. Some of them showed a moderate activity (MIC = 8 μg ml^{-1}) against *K. pneumonia* and *A. baumannii*, while the other alkaloids showed a weak activity (MIC ≥ 32 μg ml^{-1}) against the other bacteria.[50]

Other biological activities have sporadically been reported from CA includes antiallergic activity,[96] hepatoprotective activity,[97,98] attenuation of cocaine self-administration and cocaine-induced reinstatement in rats,[99] antiangiogenic activity,[100] hypothermic effects,[101] enhancement of gastrointestinal motor function[102] and cancer chemopreventive activity.[103] The results and mechanism of action are summarized in Table 5.

3. Drug-like properties of CA

According to "rule of five" or Lipinski's rule an oral drug-like molecule should have an octanol–water partition coefficient (log *P*) of <5, <5 hydrogen bond donors (HBD), <10 hydrogen bond acceptors (HBA) and a molecular weight of <500 Da.[6] Later on, two more requirements for drug-like molecules, proposed by Veber and co-workers, include NROT (number of rotatable bonds) of <10 and a PSA (polar surface area) of <140 $Å^2$.[7] This rule describes physico-chemical properties crucial for a drug's pharmacokinetics in the human body, including their absorption, distribution, metabolism, and excretion (ADME). However,

Table 5 Tabulated overview of some miscellaneous properties of *Corydalis* alkaloids

Alkaloid	Biological activity	Result(s)	Mechanism of action(s)
Dehydrocavidine (**16**) (0.5 and 1 mg kg^{-1})	Hepatoprotective activity	Significantly reversed the elevating serum enzymes (ALT, AST, LDH and ALP) induced by CCl_4	Free radical scavenging activity, promoting collagenolysis and regulating fibrosis-related genes
Dehydrocorydaline (**14**) (0.5 mmol kg^{-1} p.o.)	Antiallergic activity	Inhibited 48 h homologous passive cutaneous anaphylaxis (type 1 allergic model)	Inhibition of antibody-mediated and cell-mediated allergic reactions
DL-Tetrahydropalmatine (**54**) (10–50 mg kg^{-1} i.p.)	Hypothermic activity	Induced hypothermia in rats	Acts through serotoninergic mechanisms to induce body temperature
C. yanhusuo alkaloids and berberine (**8**)	Anti-angiogenic activity	Showed powerful anti-angiogenic activity	Suppressed the upregulation of matrix metalloproteinase 2 induced by VEGF at mRNA and protein levels
L-Tetrahydropalmatine (**71**) 3.75, and 7.5 mg kg^{-1}, i.p.	Treatment of cocaine addiction	Blocking both cocaine-induced reinforcing effects and cocaine craving	Acts on dopamine receptors
Tetrahydroberberine (**76**)	Enhances gastrointestinal motor function	Significant acceleration of gastric emptying	Acts on dopamine D_2 and/or serotonin 5-HT_{1A} receptors
Spallidamine (**92**) and 17 other alkaloids	Cancer chemopreventive activity	Strong cancer chemopreventive activity comparable with β-carotene as positive standard	Not reported

the rule does not predict pharmacological activity/potency of a compound. Compounds do not violate more than two criteria are considered as drug-like compounds. For example, compounds **69** and **70** violated PSA criteria, however, they are still considered as drug-like compounds (Table 2).

All CA (except compound **31**) interestingly satisfied the drug-like criteria (For more information about drug-like properties of other alkaloids, please see Table 2 in ESI†).

It is important to know, the world of drug-like compounds is quite limited, and filters for differentiating nondrug and drug-like compounds could certainly assist in drug discovery programs. The drug-like properties of CA ensure that CA will show acceptable ADME and translate effectively into clinical trials. The importance of drug-like properties encouraged researchers to design protocols that front-loaded of the desired physico-chemical properties at the first stage of isolation of bioactive natural products.[104]

In conclusion, drug-like properties of CA accompanied by their various biological activities make CA promising lead compounds in medicinal chemistry field.

4. Conclusion and future perspectives

Many natural drugs are discovered from medicinal plants that have a background of medicinal applications in the past. *Corydalis* species (CA) consists of more than 400 species in Eurasia and North America. Some *Corydalis* species such as *C. yanhusuo* has been traditionally used for the treatment of different diseases. *Corydalis* species are rich source of alkaloids, particularly isoquinoline alkaloids, with various biological activities including anti-cancer, acetylcholinesterase inhibitory, antimalarial, antiplatelet and antimicrobial activities. To date, different classes of isoquinoline alkaloids have been identified from this genus including aporphine, protopine, protoberberine, tetrahydroprotoberberine, benzo[*c*]phenanthridine, phthalideisoquinoline, benzylisoquinoline, morphinan (with a rearrangement in isoquinoline scaffold) and spirobenzylisoquinoline.

As we mentioned in this paper, most of CA satisfied the criteria for lead-like compounds. This means, if we would find a bioactive CA that it could show an acceptable efficacy in *in vivo* studies, and would not have a remarkable toxicity, the compound has a great chance to translate effectively into clinically meaningful activity in patients. For instance, in response to the growing mass of *in vitro* and *in vivo* evidence for berberine's antihyperglycemic and antihyperlipidemic efficacy, a number of clinical trials have recently addressed the pharmacokinetics, safety, and efficacy of berberine in humans.[105] In the United States, several randomized phase II/III/IV trials are investigating berberine's effects on a range of metabolic disorders. Five ongoing phase III/IV trials are studying berberine's effects on lipid profile. Three phase IV trials are investigating berberine's therapeutic effects in metabolic syndrome. Two phase IV trials are investigating berberine's therapeutic effects in polycystic ovary syndrome. Antihyperglycemic effects of berberine in diabetes mellitus type 2 are being investigated in two phase II/III trials. One phase II trial is studying berberine's therapeutic effects in non-alcoholic fatty liver disease. It seems that other CA can likewise translate into clinical trial if they show safety/efficacy in *in vivo* studies.

It is ultimately recommended that regarding the versatile biological properties of CA, these compounds may have even a broader range of biological applications in the future.

Acknowledgements

The authors would like to thank Dr Seyed Ahmad Emami for his scientific help.

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