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Biological and pharmacological activities of amaryllidaceae alkaloids

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Amaryllidaceae family consists of about 75 genera and 1100 species that occur wide-spread in the tropics and warm temperate regions of the world. Since the first isolation of lycorine, more than 500 amaryllidaceae alkaloids have been isolated over the past three decades. The enormous numbers of diverse amaryllidaceae alkaloids are classified into different groups mainly according to their structural features. The representative alkaloids are norbelladine, lycorine, hippeastrine, narwedine, haemanthamine, pancratistatin, pretazettine, montanine, galanthindole, cherylline and ismine. Recently, more extensive studies have revealed that amaryllidaceae alkaloids exhibit a wide range of bioactivities, such as antitumor, antiviral, antibacterial, antifungal, antimalarial and analgesic. Acetylcholinesterase (AChE) inhibitory and cytotoxic activities have also been reported. The aim of the present review is to discuss the recent developments on biological and pharmacological activities of amaryllidaceae alkaloids with IC_{50} or EC_{50} values since 2005, supporting the potential therapeutic possibilities for the use of these compounds.

1. Introduction

The family of amaryllidaceae takes its name from the genus *Amaryllis* and consists of about 75 genera, whose 1100 species are widely distributed in the tropics and warm temperate regions of the world.[1] Plants of the amaryllidaceae family have been used for thousands of years as traditional herbal medicine. The earliest evidence of their therapeutic application was discovered in the fourth century B.C.E., when Hippocrates of Cos used the oil from the daffodil, *Narcissus poeticus L.* for the treatment of uterine tumors.[2] Since the isolation of the first alkaloid lycorine **(3)** from *Amaryllida -ceae lycoris* in 1877, more than 500 amaryllidaceae alkaloids representing 11 skeletal types (Fig. 1) have been isolated over the past three decades.[3] The representative alkaloids are norbelladine (**1**), rystilline (**2**), lycorine (**3**), hippeastrine (**4**), narwedine (**5**), galanthamine (**6**), haemanthamine (**7**), pancratistatin (**8**), pretazettine (**9**), montanine (**10**), galanthindole (**11**), cherylline (**12**) and ismine (**13**) (Table 1). Such compounds exhibit a diversity of biological activities including antitumor, antiviral, antibacterial, antifungal, antimalarial, analgesic, acetylcholinesterase (AChE) inhibitory and cytotoxic activities. Among them, galanthamine (**6**) is one of the most significant amaryllidaceae alkaloids, which has been approved as a longer acting anticholinesterase drug in the treatment of Alzheimer's disease.[4] Moreover, lycorine and its derivatives have shown potential therapeutical efficacy in cancers, viral and Alzheimer's disease.[5,6,7] Their importance in medicinal chemistry has brought enormous attention to their synthesis and biological activity studies in the recent past.

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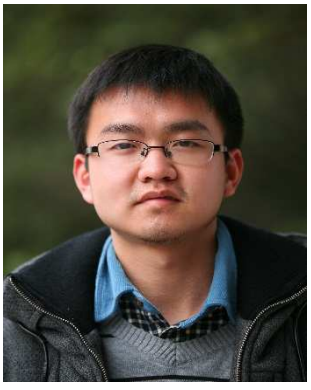

Maomao He obtained his B.Sc. degree in Pharmaceutical Engineering from Wannan Medical College in 2010. He joined Wuhan University School of Pharmaceutical Sciences for obtaining his M.Sc. in Medicinal Chemistry. He is currently pursuing his doctoral studies in Organic Biochemistry and Molecular Biology at the same institution under the guidance of Professor Xuechuan Hong. His doctoral work focuses on the total synthesis and anticancer evaluation of amaryllidaceae alkaloids and erythrina alkaloids.

Chunrong Qu received her B.Sc. in Pharmacy from Southwest University in 2011. She then earned her M.Sc. from Wuhan University in 2013 under the supervision of Professor Xuechuan Hong. She is currently engaging in her Ph.D. research at the same group, where she obtained a sponsorship by ''The Fundamental Research Funds for the Central Universities''. Her research interests include the total synthesis of medermycin, the synthesis of PBRs and applications in molecular imaging.

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Table 1 Ring types and representative amaryllidaceae alkaloids

	Framework-type	Ring type	Representative alkaloids
I	belladine-type	*N*-(3,4-dioxybenzyl)-4-oxyphenethylamine	norbelladine (**1**), rystilline (**2**)
II	lycorine-type	pyrrolo[d,e]phenanthridine	lycorine (**3**)
III	lycorenine-type	2-benzopyrano-[3,4-g]indole	hippeastrine (**4**)
IV	galanthamine-type	6*H*-benzofuro[3a,3,2-e,f]-2-benzazepine	narwedine (**5**), galanthamine (**6**)
V	crinine-type	5,10b-ethanophenanthridine	haemanthamine (**7**)
VI	narcilasine-type	lycoricidine	pancratistatin (**8**)
VII	tazettine-type	2-benzopyrano[3,4-c]indole	pretazettine (**9**)
VIII	montanine-type	5,11-methanomorphanthridine	montanine (**10**)
IX	galanthindole-type	7-phenyloctahydroindole	galanthindole (**11**)
X	cherylline-type	tetrahydroisoquinoline	cherylline (**12**)
XI	ismine-type	2-phenylcyclohexanamine	ismine (**13**)

In past decades, a variety of articles concerning the structure elucidation, biosynthesis and total synthesis and biological activities of amaryllidaceae alkaloids have been reviewed.[1] The intent of this review is to provide an overview of their *in vitro* and *in vivo* biological activities of the amaryllidaceae alkaloid family with their IC_{50} or EC_{50} values since 2005. However, it is inevitable for this review to have some overlap with the contents in previous review articles or chapters of books, especially with several excellent reviews recently published in this area.[1,2,3]

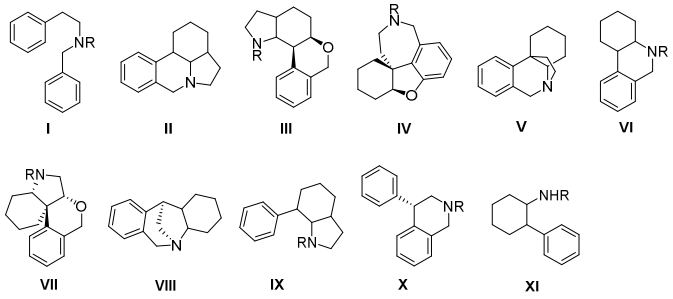

Fig.1 Main skeletal types of the amaryllidaceae alkaloids.

Xuechuan Hong studied chemistry and obtained his PhD from University of Missouri-Columbia (Missouri, USA) in 2005. After a postdoc position at Emory University under the supervision of professor Albert Padwa, he joined Albany Molecular Research Inc as a senior research scientist. Since October 2010, he was appointed a Full Professor at Wuhan University (Wuhan, China). His research interests included targeted therapy for cancer, molecular imaging, drug development.

2. Biological and pharmacological activities

2.1 Anticancer activities

Many amaryllidaceae alkaloids have been reported to exhibit antitumor properties.[5] The first well-known compound with cytostatic effect is lycorine (**3**) isolated in 1877 from the bulb of the *Amaryllidaceae lycoris*.[6] Lycorine (**3**) has been intensively investigated in various preclinical models of human cancers both *in vitro* and *in vivo*.[7] In general, lycorine (**3**) is recognized as a low micromolar antiproliferative agent against multidrug resistant and apoptosis-resistant cancers cells[8,9,10] with selective cell type-dependent cytotoxicity in tumor cells by mitochondrial pathways and inducing apoptosis. Lycorine (**3**) can down-regulate Mcl-1 in human leukemia cells.[11] The apoptosis process includes the regulation of the cell cycle in HL60 and KM3 cell lines, cytochrome-c release and caspase activation.[11] The study has further revealed that lycorine can decrease HDAC enzymatic activities in K562 cells and up-regulate the expression of p53 and its target gene product p21, then inhibit the proliferation of K562 cells.[12] Additionally, lycorine hydrochloride (**20**) can effectively suppress metastatic melanoma C8161 cell-dominant formation of capillary-like tubes *in vitro* and generation of tumor blood vessels *in vivo* with low toxicity. Mechanistic studies have revealed that lycorine hydrochloride (**20**) inhibits melanoma C8161 cell-dominant vasculogenic mimicry by reducing VE-cadherin gene expression and diminishing cell surface exposure of the protein.[8] Recently, lycorine derivative 4-ethyl-5-methyl-5,6-dihydro-[1, 3]dioxolo [4,5-j] phenanthridine (HLY78) is identified as an activator of the Wnt/β-catenin signaling pathway in a Wnt (Wingless and INT-1) ligand–dependent manner. HLY78 can promote LRP6 phosphorylation and Wnt signaling transduction, which has been recognized for its function in breast prostate cancer, and glioblastoma.[13]

The structure-activity relationship (SAR) of lycorine (**3**) has been systematically evaluated by methodically changing different parts of the structure (Fig. 2). It can be seen that conformational freedom of the C-ring, stereochemistry of the C/D-ring junction and free diol functionality in the C-ring in its original configuration in lycorine (**3**) are very crucial for anticancer activities (Table 2).[10,14,15] Lycorine (**3**) has exhibited cytostatic effects rather than cytotoxic effects through impairing the actin cytoskeleton organization in a large panel of apoptosis-resistant cancer cell lines (Table 2). Similarly, the structure

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Table 2 *In vitro* growth inhibitory activities of amaryllidaceae alkaloids on various cancer cell lines

Entry	Comp.	Cell lines, IC_{50} (μM)[a]						Ref.
		A549	OE21	Hs683	U373	SKMEL-28	B16F10	
1	lycorine (**3**)	4.3±0.3	5.1±0.4	6.7±0.3	7.6±0.2	8.5±0.3	6.3±0.4	10
2	haemanthamine (**7**)	4.5±0.6	6.8±0.7	7.0±0.3	7.7±0.5	8.5±0.2	6.8±0.2	17
3	pseudolycorine (**14**)	7.5±0.4	7.7±0.3	7.9±0.2	7.8±0.3	>10	7.5±0.3	10
4	amarbellisine (**15**)	7.2±0.3	6.7±0.2	8.3±0.3	7.3±0.2	8.3±0.2	6.7±0.3	17
5	haemanthidine (**16**)	4.0±0.4	3.7±0.2	4.3±0.2	3.8±0.2	4.2 ± 0.2	3.1±0.2	10
6	lycorine chlorohydrine (**17**)	3.8±0.2	9.6±0.7	3.1±0.3	2.3±0.1	>10	6.9±0.5	10
7	lycorin-2-one (**18**)	9.9±0.5	>10	>10	>10	>10	>10	10
8	1,2-α-epoxy lycorine (**19**)	3.4±0.1	8.5±0.5	3.3±0.2	2.4±0.1	9.5 ± 0.4	4.6±0.2	10
9	lycorine hydrochloride (**20**)	4.3±0.2	4.6±0.1	6.5±0.2	8.6±0.3	8.3± 0.3	5.5±0.2	10
10	anhydro lycorine (**21**)	4.5±0.1	8.8±0.2	7.1±0.3	5.1±0.1	>10	>10	10

[a]All cell lines were cultured in RPMI medium supplemented with 10% heat-inactivated fetal bovine serum. MEM and RPMI cell culture media were supplemented with mM glutamine, 100 μg/mL gentamicin, and pencillin-streptomycin (200 U/mL and 200 μg/mL).

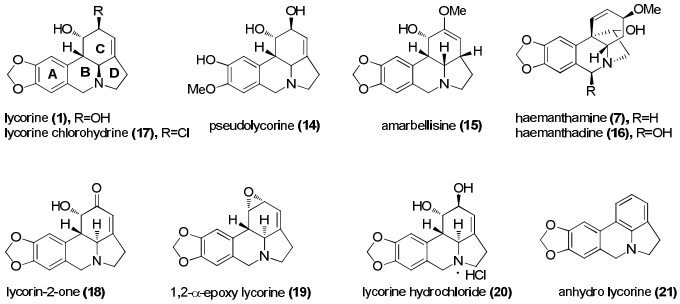

Fig. 2 Structures of lycorine-type and haemanthamine-type amaryllidaceae alkaloids.

feature of an open dioxole ring in pseudolycorine (**14**) is not essential for the antitumor activity. Haemanthamine (**7**) and haemanthidine (**16**) can decrease cell viability, mitochondrial membrane potential and induce apoptosis by declining the percentage of cells in the S phase of the cell cycle.[16] In addition, the biological results have shown that the activity against apoptosis-resistant cancers is also shared by lycorine natural congeners[10,17,18,19] as well as a number of synthetic analogues.[20]

Furthermore, lycorine provides significant therapeutic benefit in mice bearing brain grafts of the B16F10 melanoma model at non-toxic doses.[10] Its potential (*in vitro*) therapeutic ratio has been shown (> 15 times more active against cancer than normal cells) in the literature[21] and its therapeutic potential has been demonstrated in a number of mouse models of human cancers, such as Hey1B ovarian cancer,[22] LLC lung carcinoma,[23] and HL-60 leukemia.[24]

The strong relationship between the compound's lipophilicity and anticancer activities is evidenced in the design of lycorine-based anticancer agents (Table 3). A series of C1, C2-esters are synthesized and many of them have retained the anticancer activity possibly due to intracellular hydrolysis to release the parent lycorine inside the cells.[20] The increase in lipophilicity of C1, C2-esters leads to restoration of activity (**3a-3d**). Non-hydrolyzable C1, C2-ethers lycorine analogues are evaluated as well against a panel of cancer cell lines.[25] Although the SAR analysis does not reveal the activity dependence on any specific structural features present in C1- or C2-ethers or esters, diallyllycorine (**3e**) and silyl ether analogue **3g** were equipotent with lycorine throughout the tested cell lines. Diallyllycorine (**3e**) even is 100 times more potent against the apoptosis-resistant U373 glioblastoma (Table 3).[20] However, C1- or C2-hydroxyls are derivatizated as methyl ethers **3f** leading to a complete loss of the activity apparently due to reduced cell permeability through non-facilitated diffusion.

The β-crinane distichamine (**22**) as well as the phenanthridone narciprimine (**23**), two rare components of amaryllidaceae alkaloids, as shown in Fig. 3, are evaluated for cytotoxic activities against acute lymphoblastic leukemia (CEM) and other cancerous cell lines.[26] As seen in Table 4, distichamine (**22**) is active against all cancer cell lines with IC_{50} values ranging from 2.2 to 14.7 μM. Narciprimine (**23**) is active against CEM cells (IC_{50} value = 13.3 μM), while homolycorine (**25**) is active on CEM, K562 and G-361 cells. As expected from prior observations, lycorine inhibits all six cell lines in a dose-dependent manner (IC_{50} values ranging from 1.6 to 13.0 μM) after 72 h. Similar observations have previously been made for haemanthamine (**7**), which is active across the cell lines with IC_{50} values ranging from 2.1 to 8.1 μM. According to the previous demonstrations, distichamine (**22**) and narciprimine (**23**) can increase the proportion of G_2/M phase cells in a dose-dependent manner. Besides, narciprimine (**23**) and arolycoricidine (**24**) are effective in both type I and type II DNA topoisomerase (cellular targets of a number of chemotherapeutical drugs) reactions in a dose-dependent manner.[27]

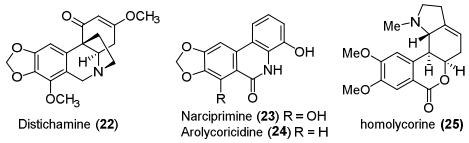

Fig. 3 Structure of distichamine (**22**), narciprimine (**23**), arolycoricidine (**24**) and homolycorine (**25**).

Isocarbostyril amaryllidaceae alkaloids (Fig. 4) are represented by hydroxylated benzophenathridones or isoquinolinone types of structure without basic nitrogen atoms. Pancratistatin (**8**), narciclasine (**27**) and lycoricidine (**28**) are the most widely known compounds against cancer cell lines among this category.[28] The cytotoxicity of narciclasine (**27**) has been evaluated in 60 cancer cell lines by the NCI, and the mean IC_{50} value was 0.046 μM.[29] Lycoricidine (**28**) is 10 times weaker (mean IC_{50} value = 0.33 μM) and pancraistatin (**8**) is 5 times weaker (mean IC_{50} value = 0.26 μM).

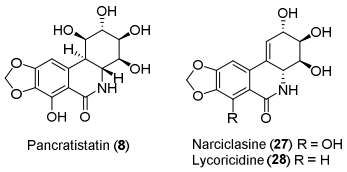

Fig. 4 Structure of pancretistatin (**8**), narciclasine (**27**) and lycoricidine (**28**).

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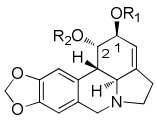
Table 3 *In vitro* growth inhibitory effects of lycorine derivatives on various cancer cell lines



3a-3h

Entry	Comp.	R_1	R_2	Log P^b	GI_{50} *in vitro* values (μM)[a]							Mean ± SEM	Ref.
					Glioma		Carcinoma			Melanoma			
					Hs683	T98G	U373	A549	MCF7	SKMEL-28	B16		
1	**3**	H	H	/	0.9	3	3	0.9	4	4	2	3 ± 1	20
2	**3a**	H	Bz	/	4	32	0.6	32	5	4	8	11 ± 5	20
3	**3b**	Bz	H	/	6	70	1	70	23	60	21	36 ± 10	20
4	**3c**	H	Ac	/	11	2	4	3	4	3	43	9 ± 5	20
5	**3d**	TIPS	Ac	/	36	6	15	18	26	27	34	30	20
6	**3e**	allyl	allyl	3.1	2	4	0.03	4	0.2	6	4	3 ± 1	20
7	**3f**	Me	H	-0.4	36	84	/	31	>100	92	40	>50	25
8	**3g**	TIPS	Allyl	5.9	7.6	6.6	/	4.5	5	2.4	0.8	4.5	25
9	**3h**	H	TIPS[c]	4.8	24	20	/	15	23	24	9.5	19.5	25

[a]The cells were cultured in RPMI media supplemented with 10% heat-inactivated fetal calf, 4 mM glutamine, 100 mg/mL gentamicin and penicillin, streptomycin (200 U/mL and 200 mg/mL). The overall growth level of each cell line was determined using the colorimetric MTT (3-[4, 5-dimethyl thiazol-2-yl]-diphenyl tetrazolium bromide) assay. Each experimental condition was performed in six replicates. [b]The log P values were calculated using the Calculate Molecular Properties protocol, launched from within Discovery Studio 3.5. 25. [c]TIPS = triisopropylsilyl.

Table 4 *In vitro* growth inhibitory effects of distichamine (**22**) and narciprimine (**23**) from Calcein AM assays using the specified cancerous cell lines

Entry	Comp.	Cell lines, IC_{50} (μM) [a,b]						Ref.
		CEM	K562	MCF7	HeLa	G-361	BJ	
1	lycorine (**3**)	1.6 ± 0.0	3.6 ± 1.2	13.0 ± 2.9	10.6 ± 0.9	5.0 ± 0.3	1.9 ± 0.1	26
2	galanthamine (**6**)[c]	>50	>50	>50	>50	>50	>50	26
3	haemanthamine (**7**)	2.1 ± 0.4	3.4 ± 1.6	8.1 ± 3.3	7.0 ± 2.2	3.7 ± 0.4	2.7 ± 0.2	26
4	distichamine (**22**)	4.5 ± 1.6	4.1 ± 0.9	2.3 ± 0.8	2.2 ± 0.1	14.7 ± 0.1	10.5 ± 1.9	26
5	narciprimine (**23**)	13.3 ± 2.5	>50	>50	>50	>50	7.9 ± 0.2	26
6	homolycorine (**25**)	15.0 ± 5.3	19.4 ± 0.8	>50	>50	32.9 ± 6.0	20.8 ± 2.3	26
7	staurosporine (**26**)[c]	0.023 ± 0.002	nt	0.064 ± 0.002	0.175 ± 0.007	nt	0.02 ± 0.0	26

[a]All cells were treated for 72 h with serial concentrations of samples. [b]Values are means of at least three independent experiments performed in triplicate, with standard deviation as indicated (nt = not tested). [c]Staurosporine and galanthamine used as positive and negative controls, respectively.

Apart from foregoing alkaloids in the family of amaryllidaceae, narciclasine (**27**) has been demonstrated to possess antitumor efficacy, which is originally isolated from *Narcissus pseudonarcissus*, with its antimitotic and displaying colchicine-like effects in 1967.[30] Narciclasine (**27**) is a potentially promising GTPase agent against brain tumors including gliomas and brain metastases. It has displayed the cytostatic activity instead of cytotoxic activities *in vivo* (IC_{50} values = 30-90 nM) in experimental models of brain cancers. The possible mechanism for the cytostatic activity involves the impairment of actin cytoskeleton organization by targeting both Rho pathway and the elongation factor eEF1A.[31] Patrick Kiss et al. have reported that narciclasine can impair cancer cell proliferation and migration at concentrations >1μM and is approximately 250-fold less sensitive to normal human fibroblast cell. Additionally, narciclasine (**27**) can induce apoptosis-mediated cytotoxic effects by triggering the activation of initiator caspases (caspase-8 and caspase-10) of the death receptor pathway in human PC-3 prostate and MCF-7 breast cancer cells.[32] The molecular docking data have shown that narciclasine (**27**) directly binds to human recombinant and yeast-purified eEF1A.[33, 34]

To explore the structure-activity relationship of narciclasine and related products, reactive positions of narciclasine (R_1, R_2, R_3, R_4, R_5) have been modified and partial *in vitro* antitumor activity data are summarized in Table 5. The data suggest that the double bond between C-10b and C1, a free lactam carbonyl function at position R_4 and a free phenolic hydroxyl group at position R_5 seem therefore to be necessary for antitumor activities. Any modifications made to positions R_4 and R_5 lead to compounds devoid of antitumor activity *in vitro* (**27a**, **27b**). Though ester **27c** has a ~10 fold weaker activity, mono-esterification of the hydroxyl at position R_1 (**27d**, **27e**) possibly improves or maintaines *in vitro* antitumor activity of narciclasine (Table 5). This may be attributed to the hydrolysis of esters to narciclasine. The elimination of the ester group in the allylic position, subsequent aromatization of ring C possibly results in the formation of narciprimine *in vitro*.[31] Accordingly, Kiss and co-workers[32] have evaluated potential prodrugs of narciclasine with respect to their high oral bioavailability and anti-tumor activity *in vivo*. Indeed narciclasine 4-O-β-D-glucopyranoside (**27i**) has comparable anticancer activity to narciclasine[35] and has <20% degradation in 1 h at pH 2 and 92% stability at physiological pH 7.4. After oral administration, prodrug (**27i**) (81132 ng.min/mL) is found to increase the absolute bioavailability of narciclasine (54812 ng. min/mL) to ~52%. Compound (**27i**) significantly increases survival times in two human GBM models (Hs683 and GL-19) at a dose level of 1 mg/kg/day while narciclasine (**27**) failed in either model at this dose.[31] Thus, it is considered to be a suitable candidate for further evaluation by both the intravenous and oral routes.

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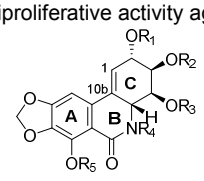
Table 5 Various narciclasine derivatives and their *in vitro* antiproliferative activity against human cancer cell lines



27a-27i

Entry	Comp.	Reaction Site[a]					Cell lines, IC_{50} (μM)[b]							Stability[c](%)	Ref.
		R_1	R_2	R_3	R_4	R_5	PC-3	U373	BxPC3	LoVo	A549	MCF-7	Median		
1	**27**	H	H	H	H	H	0.03	0.03	0.03	0.09	0.03	0.05	0.03	100	31
2	**27a**	H	H	H	H	Et	>10	>10	>10	>10	>10	>10	>10	96	31
3	**27b**	H	H	H	Et	H	>10	>10	>10	>10	>10	>10	>10	100	31
4	**27c**	Ac	H	H	H	H	0.1	0.1	0.07	0.3	0.1	0.4	0.1	20	31
5	**27d**	Bz	H	H	H	H	0.03	0.03	0.004	0.05	0.04	0.2	0.04	82	31
6	**27e**	^{i}PrCO	H	H	H	H	0.005	0.03	0.003	0.05	0.03	0.03	0.03	29	31
7	**27f**	Ac	H	H	H	Ac	0.3	0.06	0.4	0.3	0.3	0.4	0.3	56	31
8	**27g**	H	H	SO3Na	H	H	>10	>10	>10	>10	>10	>10	>10	100	31
9	**27h**	OTBS		$C(Me)_2$	H	H	>10	2	8	3	>10	4	>10	nd[d]	31
10	**27i**	H	H	H	H		0.8	0.7	1	2	2	2	1.5	92	31

[a]Chemical modification to each of 1's reaction sites. [b]The *in vitro* antiproliferative activities of the compounds are reported as IC_{50} values (nM) determined using the MTT colorimetric assay. [c]The stability of products was measured by HPLC analysis following incubation in a physiological solution at 37 °C over 7 days. Results are expressed as the % of the incubated compound recovered. [d]nd: not determined.

Table 6 *In vitro* growth inhibitory effects of pancratistatin and its derivatives on various cancer cell lines

Entry	Comp.	Cell lines, IC_{50} (μM)[a]				Ref.
		BXPC-3	DU-145	NCI-H460	MCF-7	
1	**8**	0.061	0.046	0.098	0.071	28
2	**27**	0.05±0.01	0.05±0.04	0.04±0.01	0.04±0.01	40
3	**28**	0.77±0.01	1.10±0.20	0.40±0.01	0.86±0.06	40
4	**29a**	0.34±0.05	0.72±0.27	0.53±0.01	1.81±1.20	40
5	**29b**	0.22±0.01	0.09±0.01	0.09±0.01	0.24±0.01	40
6	**29c**	0.07±0.01	0.06±0.01	0.07±0.01	0.52±0.47	40
7	**30**	0.039	0.021	0.03	0.017	28
8	**31**	0.0044	0.00049	0.00023	0.00072	28
9	**32**	14	>10.2	10.8	10.8	28

[a]Concentration required to reduce the viability of cells by 50%, after 48 h of treatment with indicated compounds, relative to DMSO control; ±SD from two independent experiments, each performed in four replicates, determined by MTT assay.

Another important isocarbostyril amaryllidaceae alkaloid, pancratistatin (**8**), discovered by Pettit in 1984[36] exhibits potential anticancer activities. So far, the mechanism of pancratistatin's anticancer potential is not exactly elucidated. Pancratistatin (**8**) can induce the apoptosis in cancer cells, produce less cytotoxic effect on the normal cells, associate with the up-regulation of Fas, increase in caspase-3, flip phosphatidyl serine and destabilize mitochondrial membrane potential.[37] Kekre's group has reported that caspase-3 activation and exposure of phosphatidyl serine on the outer leaflet of the plasma membrane are earlier than ROS and DNA fragmentation, which means pancratistatin wouldn't cause DNA double-strand breaks or DNA damage prior to the execution phase of apoptosis in cancer cells.[38] Instead, pancratistatin can selectively induce the cell death in human colon tumor xenografts' study independent of Bax and caspase activation by targeting HT-29 cancer cell *in vivo*.[39]

The intensive research work has been done in the searching of higher anti-cancer pancratistatin derivatives (Fig. 5). The pancratistatin's SAR data indicate that the presence of large hydrophobic C-1 substituents can increase anticancer activities, and the 7-hydroxy group is an important part of the cytotoxic pharmacophore as well as the full amino inositol motif (Table 6).[40] Removal of another oxygen in ring A of pancratistatin further lowers the potency 10 times relative to 7-deoxypancratistatin and 100 times relative to pancratistatin. Ingrassia and co-workers have confirmed that a *trans*-B/C ring junction and C2-, C3-, C4- hydroxyl groups are necessary to maintain pancratistatin's potent cytotoxicity. Otherwise, the *cis* B/C fusion stereochemistry leads to the reduction or complete loss of potency in isocarbostyril compounds. However, methoxy-substuituted crinane skeleton instead of polygydroxylated lycorane part of pancratistatin, has shown higher binding affinity to target proteins by caspase-3 activity in jurkat cells.[41]

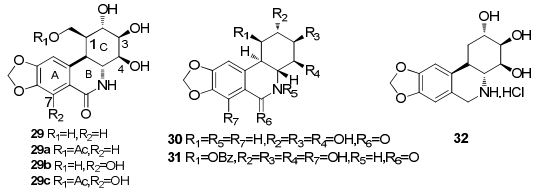


Fig. 5 Structure of pancratistatin analogues.

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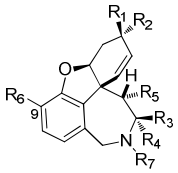
2.2 Acetylcholinesterase (AChE) inhibitory activities

Acetylcholinesterase enzyme serves as a major position in transmissions of nerve pulse by hydrolysis of acetylcholine (ACh). It has been demonstrated that ACh plays a great role in functioning human's brain with releasing basal forebrain choline. ACh deficit will lead to several neurodegenerative disorders such as Alzheimer's disease (AD). Although huge efforts have been made to investigate its pathophysiology, AD remains incurable and affects more than 36 million people around the world.[42] The typical pathological hallmarks of AD are intracellular neurofibrillary tangles, amyloid β-peptide (Aβ) plaques and loss of central cholinergic function. The current medications used for the symptomatic AD treatment such as tacrine, galanthamine, rivastigmine and donepezil mainly belong to cholinesterase inhibitors. Because cholinesterase inhibitors can slow down neurodegenerative progression *via* inhibiting AChE-induced Aβ aggregation, inhibitors of AChE and BuChE also provide additional benefits for AD treatment.[43,44]

Galanthamine (**6**), an amaryllidaceae alkaloid, is originally isolated from *Galanthus nivalis L.* in the 1940s. Galanthamine hydrobromide, under the generic name Reminyl, is the first amaryllidaceae alkaloid to be approved as a prescription drug by FDA due to its high inhibitory efficacy and both reversible and selective activity on AChE. Compared to most of the amaryllidaceae alkaloids, galanthamine displays IC_{50} value of 1.5μM, exceeds over 20 folds to other alkaloids in inhibitory potency (Table 7). As an exception, sanguinine (**6d**), a 9-hydroxy analogue, shows 10 times more active than galanthamine (Table 7, entry 5). The others (entry 2-4, 6-7) show a relative lower in hibitory activities.[45,46,47] It seems that

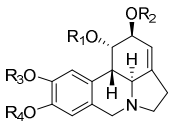
Table 7 AChE inhibitory activities of galanthamine analogues



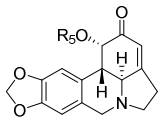
6a-6f

Entry	Compounds	R_1	R_2	R_3	R_4	R_5	R_6	R_7	IC_{50} (μM)	Ref
1	galanthamine (**6**)	OH	H	H	H	H	OMe	Me	1.5±0.2	45
2	leucotamine (**6a**)	$OCOCH_2CHOHCH_3$	H	H	H	H	H	Me	5.3±0.9	45
3	**6b**	$OCOCH_2CHOHCH_3$	H	H	H	H	OMe	Me	6.0±0.7	45
4	**6c**	$OCOCH_2CHOHCH_3$	H	H	OH	H	OMe	Me	3.5±1.1	46
5	sanguinine (**6d**)	OH	H	H	H	H	OH	Me	0.1±0.01	46
6	**6e**	OH	H	H	H	OH	OMe	Me	1.61±0.21	46
7	**6f**	H	OH	H	H	H	OMe	H	9.60±0.65	46

Table 8 AChE inhibitory activities of lycorine derivatives



3m-3v



3w-3x

Entry	Comp.	R_1	R_2	R_3	R_4	R_5	IC_{50}/Ki (μM)	Ref.
1	**3l**	benzoyl	benzoyl	$-CH_2-$		/	>50	48
2	**3b**	H	benzoyl	$-CH_2-$		/	>50	48
3	**3m**	cinnamoyl	cinnamoyl	$-CH_2-$		/	46.76±0.95	48
4	**3n**	H	cinnamoyl	$-CH_2-$		/	>50	48
5	**3o**	cinnamoyl	H	$-CH_2-$		/	>50	48
6	**3p**	$MeSCH_2-$	TBS	$-CH_2-$		/	11.4±0.66	48
7	**3**	H	H	$-CH_2-$		/	na	49[a]
8	**3c**	Ac	H	$-CH_2-$		/	0.43±0.02	49
9	**3q**	H	TBS	$-CH_2-$		/	0.86±0.03	49, 50
10	**3r**	Ac	TBS	$-CH_2-$		/	0.34±0.08	49, 50
11	**3s**	Bz	TBS	$-CH_2-$		/	0.39±0.03	49, 50
12	**3t**	Bz	Ac	$-CH_2-$		/	0.97±0.10	49, 50
13	**3a**	Bz	H	$-CH_2-$		/	0.54±0.03	49, 50
14	**3u**	Ac	Ac	$-CH_2-$		/	211±10	49
15	pseudolycorine **(14)**	H	H	Me	H	/	152.32±32.06	46
16	**3v**	H	Ac	Me	H	/	nd[b]	46
17	lycorin-2-one **(18)**	/	/	/	/	H	>50	48
18	**3w**	/	/	/	/	Ac	>50	48
19	**3x**	/	/	/	/	cinnamoyl	>50	48

[a]Values are means of three experiments, na = not active at 10 >μM, determined by Ki. [b]nd= not detected.

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Table 9 AChE inhibitory activities of other amaryllidaceae alkaloids

Entry	Compounds	IC_{50} (μM)	Ref.
1	crinine (**33**)	461 ± 14	52
2	epibuphanisine (**34**)	547 ± 5	52
3	crinamidine (**35**)	300 ± 27	52
4	hamayne (**36**)	553 ± 3	52
5	3-*O*-acetylhamayne (**36a**)	594 ± 8	52
6	epivittatine (**37**)	239 ± 9	52
7	crinamine (**38**)	697 ± 12	52
8	6-hydroxycrinamine (**38a**)	490 ± 7	52
9	8a-ethoxycrinamine (**38b**)	1145 ± 87	52
10	*N*-desmethyl-8α-ethoxyprecriwelline (**39**)	234 ± 13	52
11	*N*-desmethyl-8β-ethoxyprecriwelline (**39a**)	419 ± 8	52
12	tazettine (**9a**)	705 ± 63	52
13	cherylline (**12**)	407 ± 32	52
14	1-*O*-acetyllycorine (**3c**)	0.96 ±0.04	52
15	lycorine (**3**)	213 ±1	52
16	galanthamine (**6**)	1.9 ±0.16	52

a -OH group or protected -OH group in its allylic position (R_1) is crucial for the activities. However, its clinical application is impeded by rare nature existence. To date, a considerable amount of sanguinine has been extracted from *Pancratium Illyricum L*. in Italy by Iannello group.[45] Nevertheless, the unexpected significant inhibitory effect of sanguinine has brought some hypotheses toward the binding mechanism between galanthamine and AChE with potential modifications of its analogues.

Among the subclasses of amaryllidaceae alkaloids, lycorine (**3**) and its derivatives are investigated intensively for their anti-AChE activities (Fig. 6). Ungiminorine (**3i**) is found to be inhibitory against AChE with IC_{50} value of 0.35 μM, which is about 6-10 times stronger than galanthamine.[47] Assoanine (**3j**) also shows very strong inhibitory activity against AChE with IC_{50} values of 3.87 μM, another analogue oxoassoanine (**3k**) shows moderate inhibitory activity against AChE (IC_{50} = 47.21 μM).[46]

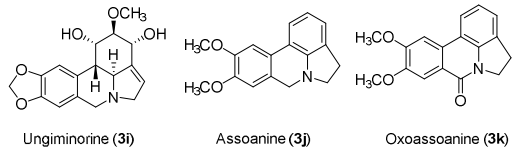

Fig. 6 Structure of lycorine analogues.

Subsequent modification of lycorine has been reported by several groups.[48,49,50,51] Some of results are summarized in Table 8. The data have shown that compound 2-*O*-tert-butyldimethylsilyl-1-*O*-(methylthio)methyllycorine (**3p**) is a dual inhibitor of human acetylcholinesterase (hAChE) and butyrylcholinesterase (hBChE) with IC_{50} values of 11.40±0.66 μM and 4.17±0.29 μM respectively (entries 1-6, 16-18) and the acylated/etherified derivatives of lycorine and lycorin-2-one are more potent against hBChE than hAChE.[48] New C1 and C2 functionalized analogs in the lycorine subset are synthesized and systematically studied.[49,50] A pronounced spike in activity from the inactive parent lycorine **3** to the 2-TBS analog **3q** (Ki = 0.86 μM) is observed. Acetylation or benzoylation of **3q** afforded **3r** (Ki = 0.34 μM) and **3s** (Ki = 0.39 μM) respectively. The lipophilic C2 silyl group binding at the enzyme active site apparently plays an important role against AChE. Desilylation of **3t** to the alcohol **3a** (Ki = 0.54 M) and subsequent acetylation to compounds **3t** (Ki = 0.97 μM), seems to be the least efficacious of the series. Moreover, 1-acetyllycorine (**3c**) has shown a potent inhibitor of AChE (Ki = 0.43 μM) while lycorine (**3**), 2-acetyllycorine (**3c**), and 1, 2-diacetyllycorine (**3u**) exhibits no or very weak inhibitory activity.[49,50,51]

Amaryllidaceae alkaloids having several different ring types are evaluated for their AChE inhibitory activities (Table 9 and Fig. 7). As known before, lycorine-type alkaloids are the most active alkaloids with 1-*O*-acetyllycorine exhibiting inhibitory effects two-fold more potent than that of galanthamine. In addi-tion, crinine (**33**), crinamidine (**35**), epivittatine (**37**), 6-hydroxycrinamine (**38a**), *N*-desmethyl-8α-ethoxypretazettine (**39**), *N*-desmethyl-8β-ethoxypretazettine (**39a**), lycorine (**3**), tazettine (**9a**) and cherylline (**12**) have weak activity.[52]

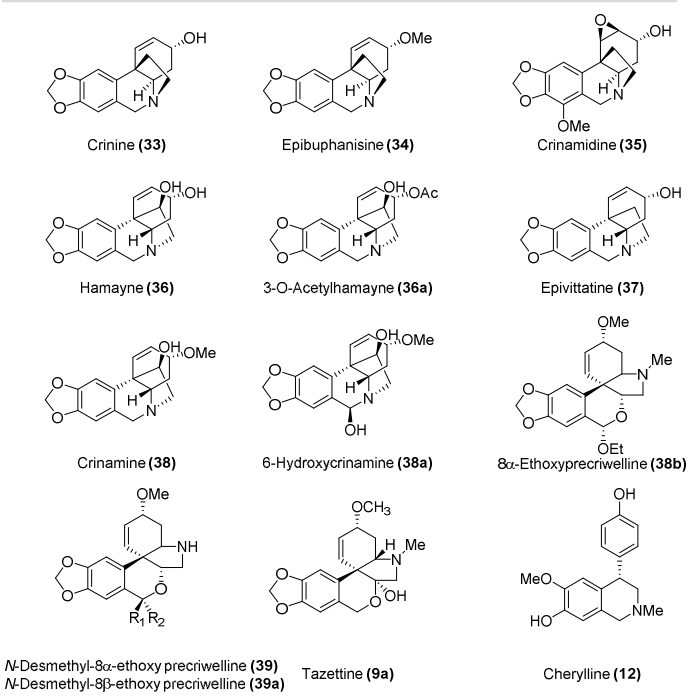

Fig. 7 Other types of amaryllidaceae alkaloids.

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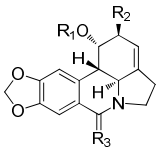
Table 10 Antiflaviviral activities of lycorine analogues

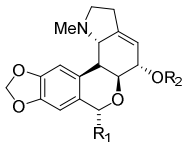


3y-3z

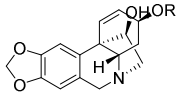
Entry	Comp.	R_1	R_2	R_3	EC_{50} (μM)[a]	CC_{50} (μM)[b]	Ref.
1	lycorine (**3**)	H	OH	H	0.23	24	54
2	**3y**	Ac	OAc	O	>300	>300	54
3	**3u**	Ac	OAc	H	1.49	110	54
4	**3c**	Ac	OH	H	0.86	66	54
5	**3z**	Ac	O	H	0.19	>300	54
6	**3q**	H	OTBS	H	0.73	78	54

[a]EC_{50} values were derived from viral titer reduction assays. Vero cells were infected with *West Nile* virus (0.1 MOI) in the presence of various concentrations of each compound. Viral titers at 42 h. [b]CC_{50} values were derived from Vero cells using an MTT assay.

Table 11 Anti-influenza virus (N5H1) effects of amaryllidaceae alkaloids



R_1 = OEt, R_2 = Me (**4a**)
R_1 = OMe, R_2 = Me (**4b**)



R = H, haemanthamine (**7**)
R = Me (**7a**)

Entry	Compounds	EC_{50}(μM)	CC_{50}(μM)	Ref.
1	**3**	<0.46	20.9	55
2	**7**	1.48	50	55
3	**7a**	6.7	>278	55
4	**4**	47.5	>317	55
5	**4a**	2.06	14.37	56
6	**4b**	0.69	4.79	56

2.3 Antiviral activities

The preliminary evaluation of lycorine, pancratistatin, narciclasines and their derivatives gives antiviral screening data that indicate the presence of *in vitro/in vivo* activities against *Flaviviridae, Bunyaviridae* and *Japanese encephalitis*.[53] Several lycorine analogues are further screened for anti-*West Nile* virus activity and cytotoxicity (Table 10). When C7 of lycorine is oxidized to a carbonyl group (entry 2), compound **3y** has lower antiviral potency. However, when one or two hydroxyl groups at the C1 and C2 positions of lycorine are substituted with other functional groups, all analogues show higher CC_{50} values than that of parental lycorine. Among them, compound **3z** (entry 5) has higher potency (EC_{50} value = 0.19 μM) and lower cytotoxicity (CC_{50} >300 μM, the highest tested concentration). It is reported that lycorine inhibited flaviviruses mainly through suppression of viral RNA synthesis. Furthermore, the data reveal that the 2K peptide plays a direct role in flavivirus RNA replication and lycorine strongly hinders RNA synthesis in flaviviruses and weakly inhibits viral protein translation.[54]

Amaryllidaceae alkaloids lycorine (**3**), hippeastrine (**4**), hemanthamine (**7**) and their analogous (Table 11) exhibit moderate to good anti-influenza activities. Particularly, lycorine and hemanthamine derivative **7a** show strong activities against influenza A virus N5H1 *in vitro*. Compound **4** has lower antiviral activity than other five compounds. Mechanistic studies show that none of these amaryllidaceae alkaloids affected the activity of the ribonucleoprotein (RNP) complex in the viral generation and replication. Instead, lycorine (**3**) delays the export of nucleoprotein from nuclear and compound **7** can block the migration from nuclear to cytoplasm in single and multiple replications.[55,56] It has been demonstrated that lycorine can inhibit the cytopathic effect (CPE) induced by severe acute respiratory syndrome-associated coronavirus (SARS-CoV). The EC_{50} value is 15.7±1.2 nM.[57] Lycorine (**3**) is also screened for antiviral activity against poliovirus (PV) using a cellular fluorescence resonance energy transfer (FRET) assay and the results have shown that it can reduce 1 log10 unit of virus titer at 2.5μM without cytotoxicity.[58] Zhang and his co-workers have reported that lycorine can inhibit human enterovirus 71 (EV71) infection in rhabdomyosarcoma (RD) cells.[5]

2.4 Antibacterial activities

The CH_2Cl_2/MeOH extract, two isolated compounds **40** and **40a** from leaves of *Crinum purpurascens* are screened for antibacterial activity. The extracts have shown remarkable potencies against all the bacteria strains used (Table 12). However, none of isolated compounds have better activity than that of the crude extract.[60]

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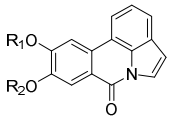
Table 12 Antibacterial activities of extracts and the isolated compounds from *Crinum purpurascens*

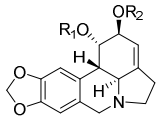


40, 40a

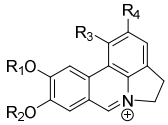
Entry	Comp.	Parameters	Bacteria strain[a]						Ref.
			EC	PA	KP	SA	ST	SPB	
1	R_1 = H, R_2 = Me (**40**)	MIC	NA	NA	NA	NA	NA	NA	60
		MBC	NA	NA	NA	NA	NA	NA	
2	R_1+R_2=-CH_2- (**40a**)	MIC	250	250	200	200	250	250	60
		MBC	300	300	300	300	300	300	
3	DCM/MeOH extract	MIC	4	3	4	4	6	6	60
		MBC	8	7	10	7	12	10	
4	ciprofloxacin (**41**)	MIC	2	2	2	2	NT	NT	60
		MBC	5	5	8	5	NT	NT	

[a]EC = *Escherichia coli*; KP = *Klebsiella pneumoniae*; ST = *Salmonella typhi*; PA = *Pseudomonas aeruginosa*; SA = *Staphylococcus aureus*; SPB = *Salmonella paratyphi B*; NT = Not tested; NA= Not active; ND= Not determined; MIC= Minimum Inhibitory Concentration; MBC= Minimum Bactericidal Concentration.

Table 13 Results of the bioassay evaluation of lycorine analogues for toxicity toward *Flavobacterium columnare* ALM-00-173



3aa-3ac



42-44

Entry	Comp.	R_1	R_2	R_3	R_4	24h IC_{50}[a,b]	MIC[b]	24h IC_{50}		MIC		Ref.
								RDCF[c]	RDCO[d]	RDCF[c]	RDCO[d]	
1	**3u**	Ac	Ac	/	/	2	4	3	3	10	11	61,62
2	**3**	H	H	/	/	27	29	49	47	100	108	63
3	**3c**	Ac	H	/	/	7.3	3.3	10.4	9.9	10	10.8	63
4	**3v**	H	Ac	/	/	17.7	16.6	27.8	26.4	50.5	54.3	63
5	**3aa**	H	*p*-MePhCO	/	/	14.6	4.7	16.4	15.6	10	10.8	63
6	**3aa**	H	*o*-MePhNCO	/	/	11.2	23.2	13.7	13.1	55	59.2	63
7	**3ac**	*o*-MePhNCO	*o*-MePhNCO	/	/	3.0	5.5	2.8	2.6	10.0	10.8	63
8	**42**	Me	Me	H	OH	0.5	1	0.28	0.27	1	1.1	64
9	**43**	Me	Me	OH	H	14.6	10.0	7.6	7.2	10.0	10.8	64
10	**44**	H	Me	H	OH	45.5	100	18.4	19.9	100	107.5	64

[a]24 h 50% Inhibition concentration in mg/l. [b]Minimum inhibitory concentration in mg/l. [c]Relative to drug control florfenicol. [d]Relative to drug control oxytetracycline.

The Gram-negative bacterium *Flavobacterium columnare* which occurs in channel catfish is able to cause *columnaris* disease. Lycorine (**3**), ungeremine (**42**) and their analogues are evaluated using a rapid bioassay for antibacterial activity against two isolates (ALM-00-173 and BioMed) of *Flavobacterium columnare* (Table 13). It has been found that substitution at the C1-*O*- or C2-*O*-position is pivotal to antibacterial efficacy of these compounds. The disubstituted lycorine *O*-analogues have better antibacterial activities than that with only one substitution at either carbon. Notably, the carbamate analogue **3ac** possesses the stronger antibacterial activity toward both *F. columnare* isolates with 24 h IC_{50} value of 3.0 mg/L. On the basis of the 24 h IC_{50} results, ungeremine has higher antibacterial activities than that of lycorine and their analogues, but none of the ungeremine analogues are more active than itself against *F. columnare*. Possibly, the aromatization of the C ring as well as the oxidation to azomethine group of C-7 is the structural feature important to antibacterial activity against *F. columnare*. Besides, the presence of the 1,3-dioxole ring joined to the A ring together with the position of the oxygenation of the C ring also plays a significant role in providing antibacterial activity.[61,62,63,64]

2.5 Other potentially useful activities

2.5.1 Antiparasitic activity

Trichomonas vaginalis is a flagellate protozoan which infects the human urogenital tract and causes vaginitis in women and urethritis in men, the most common non-viral sexually transmitted disease.[65] The cytotoxicity of lycorine in *Trichomonas vaginalis* are different compared with its activities against a variety of tumor cell lines mentioned before. For instance, lycorine arrests of the parasite cell cycle and fails to fulfill the criteria for apoptosis and apoptosis-like death. However, some similarities to paraptotic cell death described for multicellular organisms were observed.[66,67] *Trichomonas vaginalis* nucleoside triphosphate diphosphohydrolase (NTPDase) and ecto-5'-nucleotidase activities are strongly inhibited by lycorine and candimine on 24 h-treated parasites and transcript levels of NTPDase A or B are not altered by the

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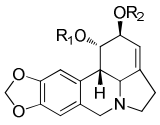
alkaloids.[68] Preliminary structure-activity relationship indicates that the presence of OH groups at C-1 and C-2 positions is not necessary to cell death lycorine induced in *T. vaginalis*. The best antiparasitic activity has been achieved with lycorine esterified at C-2 position with lauroyl group.[69]

2.5.2 Antimalarial activity

Malaria is a mosquito-borne infectious disease of humans and other animals caused by protists (a type of microorganism) of the genus Plasmodium.[70] The increasing resistance to mainstay drugs like chloroquine and controlled use of new artemisinin analogs call for novel discovery of antimalarial agents.[71] Accordingly, eight new synthesized lycorine derivatives among which are isolated from the aerial part and bulbs of *Lycoris traubii* (amaryllidaceae) are evaluated by using the drug-resistant K1 strain and the drug-sensitive FCR3 strain of *P. falciparum for* their in vitro antimalarial activities. All the tested compounds (Fig. 8), including 1,2-di-*O*-butanoyllycorine (**45**), 1-*O*-propanoyllycorine (**45a**), 1-*O*-(3′*R*)-hydroxybutanoyllycorine (**45b**), and new lycorine alkaloid 1-*O*-(3′*S*)-hydroxybutanoyllycorine (**45c**) show high antimalarial activities with IC_{50} values of 0.67, 0.37, 0.60, and 0.62 μg/mL for the K1 strain and of 0.53, 0.30, 0.45, and 0.49 μg/mL for the FCR3 strain of *P. falciparum*, respectively.[72] Additional twenty seven lycorine derivatives are synthesized and further evaluated for their *in vitro* antimalarial activity against chloroquine-sensitive strains of *Plasmodium falciparum*. The best antimalarial activities are achieved with lycorine derivatives that present free hydroxyl groups at C-1 and C-2 or esterified as acetates or isobutyrates. The presence of double bond between C-2-C-3 is also necessary for the expected efficacy.[73] (+)-5, 6-Dehydrolycorine isolated from the bulbs of *Lycoris radiate* has exhibited antimalarial activity with IC_{50} values of 1.9 μM for W-2 strain and of 2.3 μM for D-6 strain *Plasmodium falciparum* as well.[74] In addition, the antimalarial activities of the haemanthamine-type and hippeastrine (**4**) have been reported recently. The structure-activity relationship for haemanthamine derivatives has shown that the presence of the double bond at C-1-C-2 and a free hydroxyl group at C-11 is very important for great inhibitory activity. Compound **7** with two nicotinate groups at C-3 and at C-11 is the most active compound with an IC_{50} value of 0.8±0.06 μM.[75] For lycorenine derivatives from the alkaloid hippeastrine (**4**), the hippeastrine dimers have shown higher antimalarial activity.[76]



$R_1 = R_2 =$ butanoyl (**45**)
$R_1 =$ Propanoyl, $R_2 =$ H (**45a**)
$R_1 =$ (3'S)-Hydroxybutanoyl, $R_2 =$ H (**45b**)
$R_1 =$ (3'R)-Hydroxybutanoyl, $R_2 =$ H (**45c**)

Fig. 8 Structures of lycorine derivatives **45**, **45a**-**45c**

2.5.3 Anticonvulsant and antidepressant activity

The use of amaryllidaceae plants in traditional medicine for CNS activation is extensive such as age-related dementia, epilepsy and depression.[77] In this regard, the isoquinoline montanine (**10**) is originally isolated from *Hippeastrum vittatum*,[78] *Haemanthus L.*[79] which has been evaluated binding to the serotonin transporter protein (SERT) *in vitro*, anticonvulsant, sedative, anxiolytic and antidepressant properties *in vivo*. Montanine exhibits moderate SERT affinity with IC_{50} values of 121.3±3.6 μM or 36.56±1.14 μg/mL (Ki = 66.01 μM). Besides, montanine has a dose-dependent anti-convulsant activity in the PTZ-induced seizure model by blocking the Cl^- channel of $GABA_A$ receptors. Furthermore, results have shown that montanine may act on the BDZ site of the GABA receptor in the mouse's brain and shown anxiolytic-like effects when evaluated in the elevated plus maze. Moreover, montanine can reduce total immobility time and enhance struggling behavior suggesting an antidepressant effect in response to inescapable stress. In contrast with other amaryllidaceae alkaloids, montanine does not affect long-term memory retention when given *i.p*. immediately post-training sessions. For depression study, Pedersen et al.[80] have shown that *Boophone disticha* (*L.f.*) herb extracts can inhibit the serotonin transporter (SERT), noradrenalin transporter (NAT) as well as the dopamine transporter (DAT) with IC_{50} values of 423.8, 77.3 and 93.5 μM respectively. Afterwards, buphanidrine (**46**), distichamine (**22**) and buphanisine (**47**) isolated from bulbs of *Boophone disticha* have IC_{50} values of 62, 199 and 65 μM, respectively, in the SERT-binding assay,[81] which display great potency against depression and potential applications as treatments.

3. Conclusion

In summary, amaryllidaceae has played a significant role for phytochemical based drug discovery. The commercialization of the Alzheimer's drug galanthamine has captured a significant share of the neurodegenerative disease market. Lycorine, pancratistatin and their series of analogues have been intensively investigated in various preclinical models of human cancers both *in vitro* and *in vivo*. In addition, the increasing lipophilic substitution at the C2 group of lycorine apparently enhances the AChE inhibitory activity sharply, some resulting adducts have exhibited inhibitory effects several-fold more potent than that of galanthamine. They may be open a new era on the treatments of various cancers and Alzheimer diseases. Though novel approaches such as phylogenetic method and CADD to screen and obtain targeted alkaloids from amaryllidaceae plants have been reported, further studies of amaryllidaceae alkaloids should focus on SAR and QSAR analysis as well as searching for synthetic methods for the metabolites and their derivatives. To date, literatures have elucidated the mechanisms of amaryllidaceae alkaloids to some extent. However, the fully understanding the mechanism of the amaryllidaceae alkaloids are still a long way to go in the future.

Conflict of Interest

The authors hereby declare no conflicts of interest pertaining to this work.

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