European Journal of Medicinal Chemistry 63 (2013) 722–730

Contents lists available at SciVerse ScienceDirect

European Journal of Medicinal Chemistry

journal homepage: http://www.elsevier.com/locate/ejmech

Original article

Preparation and antimalarial activity of semisynthetic lycorenine derivatives

Juan C. Cedrón [a,b,c,1], David Gutiérrez [d], Ninoska Flores [d], Ángel G. Ravelo [a,b,*,1], Ana Estévez-Braun [a,b,*,1]

[a] Instituto Universitario de Bio-Orgánica "Antonio González", Av. Astrofísico Francisco Sánchez 2, 38206 La Laguna, Tenerife, Spain
[b] Instituto Canario de Investigación del Cáncer (ICIC), Spain
[c] Universidad de Ingeniería & Tecnología (UTEC), Av. Cascanueces 2281, Santa Anita, Lima 43, Peru
[d] Instituto de Investigaciones Fármaco Bioquímicas, Facultad de Ciencias Farmacéuticas y Bioquímicas, Universidad Mayor de San Andrés, Av. Saavedra 2024, 2° piso, Miraflores, La Paz, Bolivia

ARTICLE INFO

Article history:
Received 14 December 2012
Received in revised form
8 March 2013
Accepted 9 March 2013
Available online 18 March 2013

Keywords:
Lycorenine
Antiplasmodial activity
Amaryllidaceae alkaloids
Hippeastrine

ABSTRACT

A set of twenty one lycorenine derivatives has been prepared from the alkaloid hippeastrine (**1**). The modifications performed on hippeastrine included some functional group transformations, structural simplification and preparation of dimers. All alkaloids were tested as potential antimalarial agents, being the hippeastrine dimers the most active compounds.

1. Introduction

Malaria is still a main endemic disease in poorer countries, especially in Africa, where 90% of deaths due to malaria occur [1]. In the absence of effective antimalarial vaccines, low molecular weight antimalarial drugs are important weapons against the disease [2]. Quinine, chloroquine, mefloquine, and artemisinin derivatives have played an important role in fighting against malaria. However, widespread drug resistance has made them less effective with artemisinin derivatives as the only exception. Artemisinin based combination therapies (ACT) recommended by World Health Organization will help to delay the emergence of clinical resistance against artemisinins, but reports of increased parasite clearance times have emerged recently [3,4]. Therefore, it is important to discover antimalarials with novel chemical entities that are effective against the multidrug resistant parasite strains.

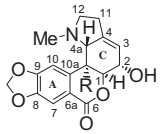
The Amaryllidaceae alkaloids, represented by a large group of selected structures belonging to eight skeletons, exhibit a wide range of biological activities, including antimalarial [5]. For example, lycorine, ungeremine and haemanthamine are the most active alkaloids against both choloroquine-sensitive and chloroquine-resistant strains of *Plasmodium falciparum* [6,7]. Alkaloids from six of the eight Amaryllidaceae alkaloids have been evaluated as antimalaric agents, showing that alkaloids with the lycorine-type are the most active [8]. In previous publications our group has reported the antimalarial activity of the haemanthamine- and lycorine-type alkaloids [9,10]. However, to the best of our knowledge, no reports for antimalarial activity have been published for the narciclasine- and lycorenine-type alkaloids. Taking into account this information and considering the large amount of hippeastrine (**1**) isolated during the phytochemical study of *Pancratium canariense* (Amaryllidaceae) [11], we decided to evaluate the antiplasmodial activity of a set of semisynthetic lycorenine derivatives, and establish some structure–activity relationships. This is the first report of the antimalarial activity of lycorenine-type alkaloids, although other bioactivities, such as cytotoxic [12] and antifungic [13] have been reported.

2. Results and discussion

Sixteen alkaloids were isolated from the bulbs of *P. canariense* [11,14]. Two of them, hippeastrine (**1**) (1.35 g) and pancratinine A (**2**) (4.5 mg) present a lycorenine skeleton (Fig. 1).

* Corresponding authors. Instituto Universitario de Bio-Orgánica "Antonio González", Av. Astrofísico Francisco Sánchez 2, 38206 La Laguna, Tenerife, Spain.
E-mail address: aestebra@ull.es (A. Estévez-Braun).
[1] http://www.icic.es.

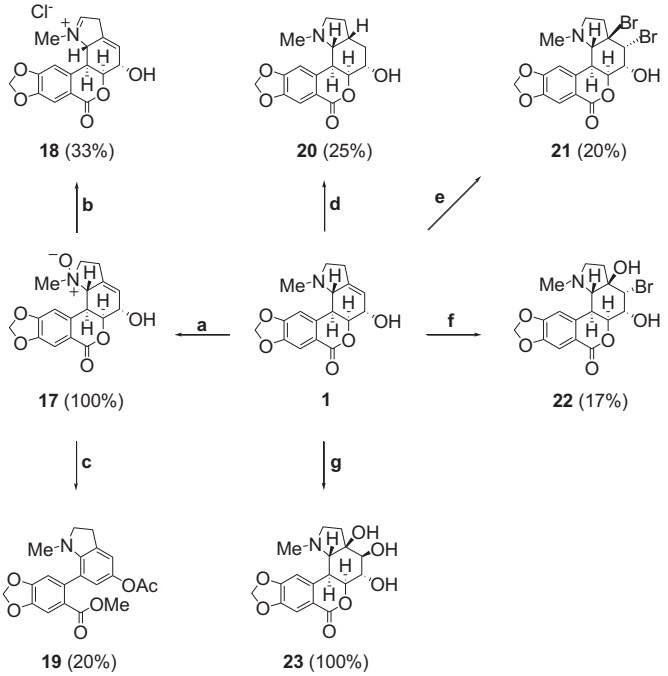
0223-5234/$ – see front matter © 2013 Elsevier Masson SAS. All rights reserved.
http://dx.doi.org/10.1016/j.ejmech.2013.03.018



Hippeastrine (**1**): R = H
Pancratinine A (**2**): R = OH

Fig. 1. Structures of lycorenine-type alkaloids isolated from *Pancratium canariense*.

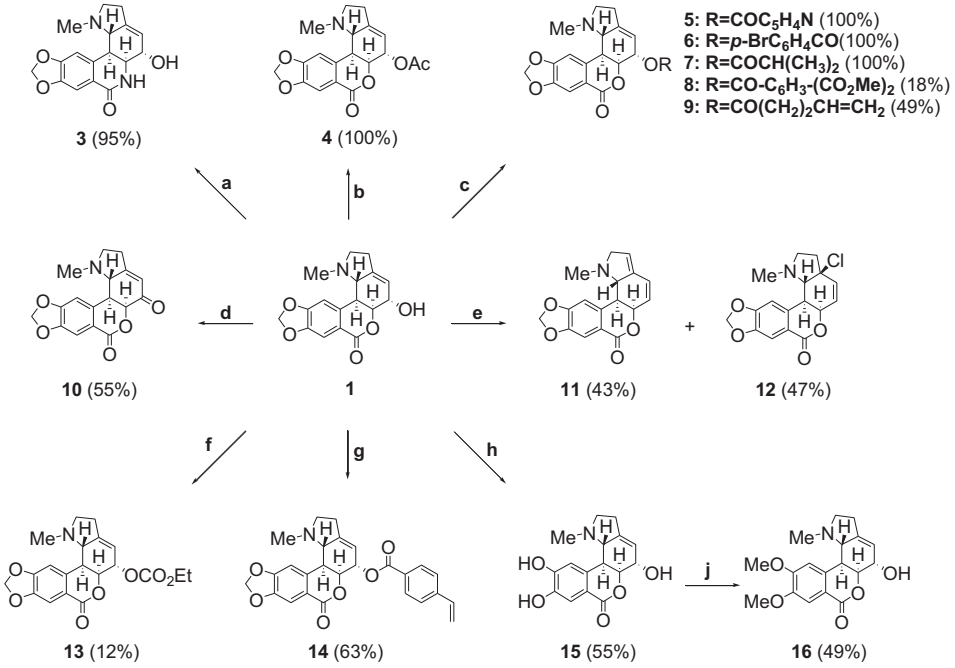
Because of the large amount of **1** isolated, several modifications were achieved on its structure, as shown in Schemes 1–3. For the most part, the transformations were carried out on the hydroxyl group at C-2 or on the double bond presents in ring C. Transformation of the lactone into lactam was made by using ammonium acetate in acetic acid. Thus, compound **3** was obtained in high yield with the same B/C *cis* ring fusion as **1**. Compound **4** was obtained quantitatively by acetylation of **1** with Ac_2O/Py, while compounds **5**–**9** were prepared by acylation of **1** with several acyl chlorides of different size, lipophilicity and stereoelectronic properties. Oxidation of the hydroxyl group at C-2 of **1** to yield the cyclohexanone **10** was only achieved when the Jones reagent [15] was employed, since the use of other oxidants, such as PCC [16], sulphur trioxide [17] or manganese dioxide [18] was unsuccessful. Treatment of **1** with thionyl chloride yielded two products, **11** and **12**, which have a double bond at C-2–C-3 due to the corresponding elimination of the hydroxyl group at C-2. The β orientation for the chlorine group of **12** was established for the value of the chemical shift of H-4a in **12** respect to that in **1**, in agreement with the data published for 6-membered ring with chorine atom and hydrogen in *syn* or *anti* disposition [19]. With the aim of eliminate the NMe group, **1** was reacted with ethyl chloroformiate, a useful reagent for this purpose [20]. However, only derivative **13** was obtained, showing no demethylation, but formation of a carbonate at C-2. Another modification achieved at the hydroxyl group was the esterification with *p*-vinylbenzoic acid, yielding **14**. The methylenedioxy group of



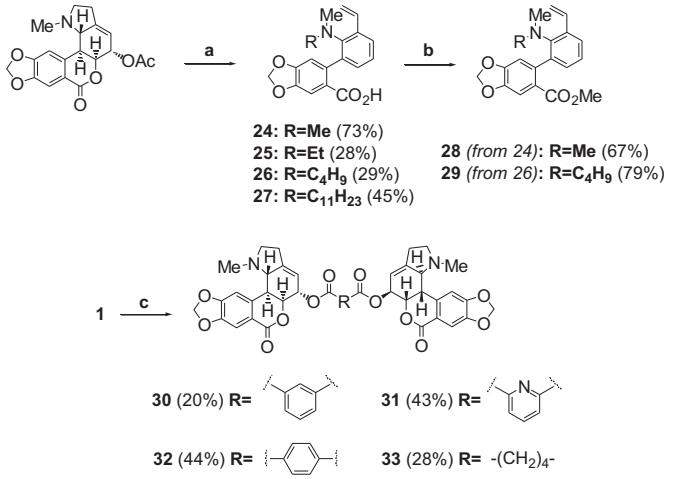
Scheme 2. Reagents and conditions: (a) 30% H_2O_2, MeOH, 24 h; (b) 1. HCl 12 M, MeOH, 30 min; 2. $FeSO_4{\cdot}7H_2O$, MeOH, 0 °C; 3. EDTA 0.1 M, 30 min; (c) Ac_2O, DCM, reflux; (d) H_2, 10% Pd/C, THF; (e) Br_2, DCM, 24 h; (f) $CH_3CONHBr$, $SnCl_4$, H_2O, CH_3CN, 0 °C; (g) OsO_4, NMO, 24 h.

1 was also modified. Reaction of **1** with BBr_3 produced catechol **15**, which reacted with trimethylsilyldiazomethane to yield 2α-hydroxyhomolycorine **16**, an alkaloid isolated previously from *Leucojum vernum* [21].

The nitrogen atom of **1** was also modified, as shown in Scheme 2. The N-oxide **17** was quantitatively obtained by reaction with



Scheme 1. Reagents and conditions: (a) $CH_3CO_2NH_4$, CH_3CO_2H, reflux; (b) Ac_2O, py; (c) RCl, NEt_3, DCM; (d) Jones reagent, acetone; (e) $SOCl_2$, DCM, reflux; (f) $ClCO_2Et$, $KOH_{(aq)}$, DCM; (g) *p*-vinylbenzoic acid, DCC, DCM; (h) BBr_3, DCM; (j) Me_3SiCHN_2, MeOH.



Scheme 3. Reagents and conditions: (a) 1. RX, CH_3CN, 24 h; 2. *t*-BuOK/*t*-BuOH, reflux, 4 h; (b) Me_3SiCHN_2, MeOH; (c) 0.5 equiv acyl chloride, NEt_3, 24 h.

hydrogen peroxide. Using **17** as starting material, we tried to obtain the corresponding N-demethylated derivative. Thus, Polonovski reaction was performed [22]. In this case, the ammonium derivative **18** was formed in 33% yield. Another reagent used for this type of transformation is acetic anhydride. When **17** was refluxed with Ac_2O no demethylation was observed, being isolated only the achiral compound **19** together with 2-acetylhippeastrine **4**. **19** could be formed by lactone opening followed by aromatization.

Several modifications were also carried out on the double bond C-3–C-4 of **1**. Hydrogenation of **1** in the presence of 10% Pd/C yielded **20** in low yield (25%), showing a *cis* union for the C and D rings. The *cis* fusion was established by the value of the coupling constant between H-4a and H-4 (J = 5.8 Hz). Since the absolute configuration of hippeastrine (**1**) is known [11,14], the stereochemistry at C-4 was established as 4*R*. Bromination of the double bond yielded the dibrominated derivative **21**. Due to the small coupling constant (J = 3.3 Hz) between the axial hydrogen H-2 and H-3, this last hydrogen must have an equatorial disposition and, therefore, the bromine atom at C-3 is axial. According to the mechanism for bromination of double bonds, the bromine at C-4 is β-axial, and the stereochemistry at C-3 and C-4 was established as 3*S*,4*S*. Haloamidation of the double bond following the procedure described by Corey [23] yielded compound **22**. In this case a bromohydroxylation occurred instead of the expected bromoamidation, probably because the cyclic bromonium ion intermediate was opened by the water used instead of acetonitrile. In the ^{1}H NMR spectrum of compound **22**, the hydrogen H-3 appeared as a singlet at δ 4.80, indicating an almost null coupling with the axial hydrogen H-2. Therefore, H-3 is equatorial and the bromine at C-3 is α-axial, and in agreement with the established mechanism of this reaction, the hydroxyl at C-4 presents an *anti* orientation with respect to the bromine. Thus, the absolute stereochemistry at C-3, C-4 was determined as 3*R*, 4*R*. Treatment of **1** with osmium tetraoxide produced quantitatively the *cis*-diol **23**. The orientation of the hydroxyl groups was determined as β on the basis of the large value for the coupling constant of H-3 (J = 10.3 Hz), indicating a *trans* disposition with H-2, and consequently the absolute stereochemistry 3*S*,4*R*.

Hofmann elimination [24] was performed on compound **4** in order to achieve a structural simplification. Thus, when 2-acetylhippeastrine was refluxed with different alkyl halides and then treated with *t*-BuOK/*t*-BuOH the corresponding biphenyl derivatives **24**–**27** were obtained. Under these reaction conditions, the D ring and the lactone ring openings occurred and the aromatization of the C ring was also detected. The treatment of compounds **24** and **26** with trimethylsilyldiazomethane yielded the methyl esters **28** and **29**, respectively.

Clivimine is the only dimeric alkaloid known isolated from Amaryllidaceae [25]. This compound consists of two units of clivonine (another lycorenine-type alkaloid) joined through 2,6-dimethyl-pyridine-3,5-dicarboxylic acid [26]. With the aim of preparing hippeastrine dimers, we decided to react **1** with some aromatic diacyl dichlorides such as pyridine-2,6-dicarboxylic acid, isophthalic acid, terephthalic acid and with the aliphatic hexanedioic acid. Under usual acylation conditions, four dimers **30**–**33** were obtained as shown in Scheme 3.

Results of the antiplasmodial evaluation against F-32 Tanzania (chloroquine sensitive) strains of *P. falciparum* are summarized in Table 1. Only six lycorenine derivatives (**8**, **15**, **30**–**33**) showed antiplasmodial activity with IC_{50} values less than 10 μM. The dimers **30** and **32** resulted be the most active compounds being 9.8-fold more active than the monomer (**1**).

From the obtained results some structure–activity relationships can be outlined. The presence of a free hydroxyl group at C-2 seems important for the activity since the acylated derivatives (**4**–**7**, **9**, **10**, **13** and **14**) were less active than hippeastrine. The oxidation of the hydroxyl group also led to a less active compound (**1** vs. **10**). Concerning the modifications on the double bond at C-3–C-4 of the ring C, derivatives **20**–**23** were less active than **1**, indicating that this double bond is important for the activity. A simplification of the licorenine skeleton achieved by the formation of the biphenyls **24**–**29** produced a drastic loss of activity. The N-oxide derivative **17**

Table 1
In vitro activity of compounds **1**–**33** against *Plasmodium falciparum* F32.

Compound	IC_{50} (μM)[a]
1	12.7 ± 1.0
2	60.4 ± 6.0
3	18.8 ± 2.0
4	58.8 ± 8.0
5	49.8 ± 5.0
6	42.2 ± 8.0
7	57.1 ± 8.0
8	7.4 ± 0.5
9	55.4 ± 7.0
10	79.8 ± 12.0
11	94.1 ± 10.0
12	84.0 ± 12.0
13	61.8 ± 8.0
14	49.4 ± 4.0
15	9.9 ± 1.0
16	84.5 ± 9.0
17	87.6 ± 6.0
18	>100
19	70.4 ± 5.0
20	78.8 ± 6.0
21	58.7 ± 8.0
22	58.3 ± 4.0
23	68.7 ± 8.0
24	67.5 ± 6.0
25	86.1 ± 9.0
26	>100
27	>100
28	67.6 ± 6.0
29	76.2 ± 8.0
30	1.3 ± 0.1
31	3.9 ± 0.9
32	1.3 ± 0.1
33	4.0 ± 1.0
Chloroquine	0.03

[a] Data are expressed as mean standard deviation of three determinations.

was inactive, indicating the importance of the unshared electron pair on the nitrogen of hippeastrine. On the other hand, the replacement of the methylenedioxy moiety by hydroxyl groups (compound **15**) produced a slight improvement in the antiplasmodial activity.

3. Conclusion

A set of 21 lycorenine derivatives has been prepared through selective transformations. The modifications performed on the structure of hippeastrine (**1**) indicate that only the elimination of the methylenedioxy group produces a slight increase of the activity. However, when different dimers of hippeastrine were evaluated, the activity increased 10-fold compared to the single alkaloid. The fact that dimers are significantly more potent than monomers may suggest improved binding to their specific target than the monomers or a possible hydrolysis of the dimer, giving place to two molecules of **1** during the biological evaluation. Our results represent a starting point for preparing new lycorenine-type derivatives with improved antiplasmodial activity.

4. Experimental

4.1. General

IR spectra were obtained using a Bruker IFS28/55 spectrophotometer. Optical rotations were measured with a Perkin–Elmer 241 automatic polarimeter. ^{1}H and ^{13}C NMR spectra were recorded, unless any other indication, in $CDCl_3$ or MeOD at 300 and 75 MHz respectively. 2D NMR experiments were conducted on a Bruker WP-400 SY NMR spectrometer at 400 MHz. High- and low-resolution mass spectra were obtained on a VG Autospec spectrometer. Analtech Silica Gel GF preparative layer with UV254 was used for TLC. Silica gel (0.2–0.63 mm) was employed for column chromatography. Silica gel 60 (Merck) was used on a Harrison Research 7924T Chromatotron.

4.2. Biological assays

F-32 Tanzania (chloroquine sensitive) strains of *P. falciparum* were cultured according to Trager and Jensen [27] on glucose-enriched RPMI 1640 medium, supplemented with 10% human serum at 37 °C. After 24 h of incubation at 37 °C, the medium was replaced by fresh medium supplemented with the compound to be evaluated in DMSO at three different concentrations (0.1, 1 and 10 μg/mL) and incubation was continued for further 48 h. On the third day of the test, a blood smear was taken from each well and parasitaemia was calculated for each concentration of sample compared to the control. IC_{50} values were determined graphically by plotting concentrations vs. percent inhibition. Chloroquine (0.03 μM) was used as a positive control. All tests were performed in triplicate.

4.3. Chemistry

Alkaloids **1** and **2** were extracted from the bulbs of *P. canariense* as described in Ref. [11].

4.3.1. Hippeastrine lactam (*3*)

20 mg (0.064 mmol) of **1** and 100 mg (20 equiv) of ammonium acetate dissolved in 5 mL of acetic acid were refluxed for 2 h. The solvent was then evaporated and the residue was purified by preparative TLC using DCM:MeOH 17:3 as eluent. 19 mg (85%) of **3** was obtained as an amorphous white solid. $[\alpha]_D^{20} = +57.6$ (*c* 0.27, EtOH). ^{1}H NMR ($CDCl_3$) δ 7.45 (1H, s, H-7), 7.03 (1H, s, H-10), 6.06 (2H, s, OCH_2O), 5.73 (1H, s, H-3), 4.60 (1H, s, H-1), 4.34 (1H, s, H-2), 3.35 (1H, m, H-10b), 3.23 (1H, d, $J = 9.6$ Hz, H-12), 2.86 (1H, d, $J = 9.5$ Hz, H-4a), 2.58 (2H, br s, H-11), 2.40 (1H, dd, $J = 9.7$ Hz, $J = 18.4$ Hz, H-12), 2.00 (3H, s, NMe) ^{13}C NMR ($CDCl_3$) δ 164.3 (s, C-6), 151.7 (s, C-9), 147.8 (s, C-8), 142.4 (s, C-4), 138.4 (s, C-10a), 119.3 (d, C-3), 118.2 (s, C-6a), 109.7 (d, C-7), 108.5 (d, C-10), 101.9 (t, OCH_2O), 81.6 (d, C-1), 67.1 (d, C-2), 66.2 (d, C-4a), 55.4 (t, C-12), 42.6 (q, NMe), 37.6 (d, C-10b), 27.2 (t, C-11). IR (neat, cm^{-1}): 3438, 1645, 1481, 1387, 1290, 1252, 1121, 1034, 936. HRMS *m*/*z* 314.1273 (calcd for $C_{17}H_{18}N_2O_4$ $[M]^+$ 314.1267).

4.3.2. 2-Acetylhippeastrine (*4*)

To 14 mg (0.044 mmol) of **1** in 1 mL of pyridine, 0.3 mL (3.18 mmol) of acetic anhydride was added. After 3 h of stirring, the solvent was evaporated and the residue was purified by preparative TLC with DCM:MeOH 19:1, yielding 16 mg (100%) of **4** as an amorphous white solid. $[\alpha]_D^{20} = +87.9$ (*c* 1.4, EtOH). ^{1}H NMR ($CDCl_3$) δ 7.46 (1H, s, H-7), 6.97 (1H, s, H-10), 6.06 (2H, s, OCH_2O), 5.59 (1H, s, H-3), 5.42 (1H, s, H-2), 4.57 (1H, s, H-1), 3.16 (1H, m, H-12), 2.82 (1H, d, $J = 9.6$ Hz, H-10b), 2.70 (1H, d, $J = 9.4$ Hz, H-4a), 2.54 (2H, br s, H-11), 2.27 (1H, dd, $J = 9.2$ Hz, $J = 18.1$ Hz, H-12), 2.06 (3H, s, $OCOCH_3$), 2.04 (3H, s, NMe). ^{13}C NMR ($CDCl_3$) δ 169.4 (s, O$\underline{C}$OCH$_3$), 163.8 (s, C-6), 151.7 (s, C-9), 147.8 (s, C-8), 147.7 (s, C-4), 138.5 (s, C-10a), 118.2 (s, C-6a), 114.6 (d, C-3), 109.6 (d, C-7), 108.5 (d, C-10), 101.9 (t, OCH_2O), 79.2 (d, C-1), 68.5 (d, C-2), 66.5 (d, C-4a), 55.9 (t, C-12), 43.3 (q, NMe), 40.7 (d, C-10b), 27.8 (t, C-11), 20.7 (c, OCO$\underline{C}$H$_3$). IR (neat, cm^{-1}): 2926, 2852, 2791, 1728, 1616, 1503, 1481, 1450, 1383, 1292, 1226, 1121, 1031, 939, 755, 655. HRMS *m*/*z* 358.1282 (calcd for $C_{19}H_{20}NO_6$ $[M + 1]^+$ 358.1291).

4.3.3. General procedure for acylation of *1*

To a solution of haemanthamine **1** in 5 mL of dry DCM were added NEt_3 and the corresponding acyl chloride. After 12 h of stirring at rt, the solvent was evaporated and the residue was purified by preparative TLC using DCM:MeOH 9:1 as eluent, yielding the corresponding esters **5**–**9**.

4.3.4. 2-Nicotylhippeastrine (*5*)

Following the procedure described above, 19.5 mg (0.062 mmol) of **1** was treated with 22 μL (2.5 equiv) of NEt_3 and 17 mg (1.5 equiv) of nicotyl chloride. After purification, 26 mg (100%) of **5** was obtained as an amorphous white solid. $[\alpha]_D^{20} = +131.6$ (*c* 1.6, EtOH). ^{1}H NMR ($CDCl_3$) δ 9.18 (1H, s, CC$\underline{H}$N), 8.78 (1H, dd, $J = 1.5$ Hz, $J = 4.8$ Hz, NC$\underline{H}$CHCHC), 8.27 (1H, d, $J = 6.1$ Hz, NCHCHC$\underline{H}$C), 7.48 (1H, s, H-7), 7.39 (1H, dd, $J = 4.8$ Hz, $J = 7.7$ Hz, NCHC$\underline{H}$CHC), 6.97 (1H, s, H-10), 6.07 (1H, br s, OCH_2O), 6.05 (1H, br s, OCH_2O), 5.71 (2H, s, H-2, H-3), 4.73 (1H, s, H-1), 3.18 (1H, m, H-12), 2.91 (1H, d, $J = 10.2$ Hz, H-10b), 2.75 (1H, d, $J = 9.3$ Hz, H-4a), 2.57 (2H, m, H-11), 2.30 (1H, dd, $J = 9.2$ Hz, $J = 18.5$ Hz, H-12), 2.09 (3H, s, NMe). ^{13}C NMR ($CDCl_3$) δ 163.8 (s, C-6), 163.7 (s, C═O), 153.4 (d, C$\underline{C}$HN), 151.7 (s, C-9), 150.7 (d, N$\underline{C}$HCHCHC), 148.3 (s, C-4), 147.9 (s, C-8), 138.3 (s, C-10a), 137.2 (d, NCHCH$\underline{C}$HC), 125.5 (s, $\underline{C}$CHN), 123.2 (d, NCH$\underline{C}$HCHC), 118.1 (s, C-6a), 114.0 (d, C-3), 109.6 (d, C-7), 108.6 (d, C-10), 101.9 (t, OCH_2O), 79.0 (d, C-1), 69.5 (d, C-2), 66.5 (d, C-4a), 55.9 (t, C-12), 43.4 (q, NMe), 40.9 (d, C-10b), 27.9 (t, C-11). IR (neat, cm^{-1}): 2926, 2853, 2791, 1724, 1617, 1592, 1480, 1385, 1254, 1112, 1034, 937, 881, 749, 701, 664. HRMS *m*/*z* 421.1398 (calcd for $C_{23}H_{21}N_2O_6$ $[M]^+$ 421.1400).

4.3.5. 2-(p-Bromobenzoyl)-hippeastrine (*6*)

Following the procedure described above, 18.7 mg (0.6 mmol) of **1** was reacted with 21 μL (2.5 equiv) of NEt_3 and 19.5 mg (1.5 equiv) of *p*-bromobenzoyl chloride to yield, after purification, 29 mg (100%) of **6** as an amorphous white solid. $[\alpha]_D^{20} = +96.6$ (*c* 1.6, EtOH). ^{1}H NMR ($CDCl_3$) δ 7.85 (2H, d, $J = 8.3$ Hz, COC(C$\underline{H}$)$_2$), 7.54 (2H, d, $J = 8.3$ Hz, (C$\underline{H}$)$_2$CBr), 7.46 (1H, s, H-7), 7.00 (1H, s, H-10), 6.06 (2H, s,

OCH_2O), 5.70 (1H, s, H-3), 5.67 (1H, s, H-2), 4.71 (1H, s, H-1), 3.23 (1H, br s, H-12), 2.95 (1H, d, J = 8.3 Hz, H-10b), 2.77 (1H, d, J = 8.4 Hz, H-4a), 2.58 (2H, br s, H-11), 2.34 (1H, dd, J = 9.2 Hz, J = 18.3 Hz, H-12), 2.12 (3H, s, NMe). ^{13}C NMR ($CDCl_3$) δ 164.3 (s, C-6), 163. 8 (s, C=O), 151.7 (s, C-9), 147.9 (s, C-4), 147.4 (s, C-8), 138.2 (s, C-10a), 131.5 (d, COC(CH)$_2$), 131.0 (d, (CH)$_2$CBr), 128.3 (s, COC(CH)$_2$), 128.2 (s, (CH)$_2$CBr), 118.1 (s, C-6a), 114.5 (d, C-3), 109.6 (d, C-7), 108.6 (d, C-10), 102.0 (t, OCH_2O), 78.9 (d, C-1), 69.1 (d, C-2), 66.6 (d, C-4a), 55.9 (t, C-12), 43.3 (q, NMe), 40.7 (d, C-10b), 27.8 (t, C-11). IR (neat, cm^{-1}): 2927, 2790, 1722, 1618, 1590, 1481, 1386, 1293, 1255, 1099, 1036, 1010, 938, 846, 756, 664. HRMS m/z 497.0466 (calcd for $C_{24}H_{20}NO_6Br$ $[M]^+$ 497.0474).

4.3.6. 2-Isobutyrylhippeastrine (**7**)

Following the procedure described above, 15.5 mg (0.05 mmol) of **1** was treated with 18 μL (2.5 equiv) of NEt_3 and 8 μL (1.5 equiv) of isobutyryl chloride. After purification, 18 mg (100%) of **7** were obtained as an amorphous white solid. $[\alpha]_D^{20}$ = +94.0 (c 0.5, EtOH). ^{1}H NMR ($CDCl_3$) δ 7.46 (1H, s, H-7), 7.00 (1H, s, H-10), 6.07 (2H, s, OCH_2O), 5.59 (1H, s, H-3), 5.42 (1H, s, H-2), 4.55 (1H, s, H-1), 3.23 (1H, br s, H-12), 2.86 (1H, d, J = 8.7 Hz, H-10b), 2.73 (1H, d, J = 8.7 Hz, H-4a), 2.50 (3H, m, H-11, CH(CH$_3$)$_2$), 2.27 (1H, dd, J = 9.5 Hz, J = 18.1 Hz, H-12), 2.10 (3H, s, NMe), 1.13 (3H, d, J = 4.3 Hz, CH(CH$_3$)$_2$), 1.08 (3H, d, J = 4.3 Hz, CH(CH$_3$)$_2$). ^{13}C NMR ($CDCl_3$) δ 175.5 (s, C=O), 163.8 (s, C-6), 151.7 (s, C-9), 147.9 (s, C-8, C-4), 138.3 (s, C-10a), 118.2 (s, C-6a), 115.0 (d, C-3), 109.6 (d, C-7), 108.6 (d, C-10), 101.9 (t, OCH_2O), 79.2 (d, C-1), 68.1 (d, C-2), 66.6 (d, C-4a), 55.9 (t, C-12), 43.3 (q, NMe), 33.7 (d, C-10b), 31.5 (d, CH(CH$_3$)$_2$), 27.7 (t, C-11), 18.6 (q, CH(CH$_3$)$_2$). IR (neat, cm^{-1}): 2928, 2789, 1731, 1617, 1481, 1386, 1291, 1251, 1189, 1151, 1119, 1035, 939, 755, 663. HRMS m/z 385.1585 (calcd for $C_{21}H_{23}NO_6$ $[M]^+$ 385.1525).

4.3.7. 2-[3,5-Bis(methoxycarbonyl)benzoyl]hippeastrine (**8**)

Following the procedure described above, 35 mg (0.11 mmol) of **1** was treated with 8 μL (0.5 equiv) of NEt_3 and 6.6 μL (0.33 equiv) of trimesyl chloride. After purification, 11.3 mg (18%) of **8** were obtained as an amorphous white solid. $[\alpha]_D^{20}$ = +108.9 (c 0.28, MeOH). ^{1}H NMR ($CDCl_3$) δ 8.85 (1H, s, CH(CCO$_2$Me)$_2$), 8.79 (2H, s, COC(CH)$_2$), 7.48 (1H, s, H-7), 7.02 (1H, s, H-10), 6.08 (1H, br s, OCH_2O), 6.06 (1H, br s, OCH_2O), 5.74 (2H, s, H-2, H-3), 4.76 (1H, s, H-1), 3.97 (6H, s, OMe), 3.21 (1H, br s, H-12), 2.97 (1H, br s, H-10b), 2.79 (1H, s, H-4a), 2.61 (2H, d, J = 5.4 Hz, H-11), 2.32 (1H, m, H-12), 2.12 (3H, s, NMe). ^{13}C NMR ($CDCl_3$) δ 165.0 (s, CO$_2$Me), 163.8 (s, C-6), 163.5 (s, C=O), 151.7 (s, C-9), 147.8 (s, C-8), 138.2 (s, C-10a), 134.7 (d, CH(CCO$_2$Me)$_2$), 134.4 (d, COC(CH)$_2$), 131,0 (s, CH(CCO$_2$Me)$_2$), 130.4 (s, COC(CH)$_2$), 118.1 (s, C-6a), 114.2 (d, C-3), 109.6 (d, C-7), 108.7 (d, C-10), 101.9 (t, OCH_2O), 78.9 (d, C-1), 69.6 (d, C-2), 66.5 (d, C-4a), 55.9 (t, C-12), 52.4 (q, OMe), 43.3 (q, NMe), 27.8 (t, C-11). IR (neat, cm^{-1}): 3057, 2954, 2850, 1730, 1615, 1449, 1385, 1292, 1119, 1036, 999, 937, 783, 738, 700. HRMS m/z 534.1389 (calcd for $C_{28}H_{24}NO_{10}$ $[M - 1]^+$ 534.1400).

4.3.8. 2-(4-Pentenoyl)hippeastrine (**9**)

Following the procedure described above, 20 mg (0.064 mmol) of **1** was treated with 23 μL (2.5 equiv) of NEt_3 and 14 μL (2 equiv) of 4-pentenoyl chloride. After purification, 12.3 mg (49%) of **9** were obtained as an amorphous white solid. $[\alpha]_D^{20}$ = +94.3 (c 0.07, MeOH). ^{1}H NMR ($CDCl_3$) δ 7.46 (1H, s, H-7), 6.97 (1H, s, H-10), 6.13 (2H, s, OCH_2O), 5.78 (1H, m, CH_2CH_2CH=CH_2), 5.58 (1H, s, H-3), 5.44 (1H, s, H-2), 5.01 (1H, d, J = 18.0 Hz, CH_2CH_2CH=CH_2), 4.96 (1H, d, J = 10.5 Hz, CH_2CH_2CH=CH_2), 4.56 (1H, s, H-1), 3.19 (1H, br s, H-12), 2.83 (1H, d, J = 8.6 Hz, H-10b), 2.70 (1H, d, J = 8.6 Hz, H-4a), 2.54 (2H, s, H-11), 2.38 (5H, m, CH_2CH_2CH=CH_2, H-12), 2.07 (3H, s, NMe). ^{13}C NMR ($CDCl_3$) δ 171.4 (s, C=O), 163.8 (s, C-6), 151.6 (s, C-9), 147.8 (s, C-4), 147.8 (s, C-8), 138.4 (s, C-10a), 136.1 (d, CH_2CH_2CH=CH_2), 118.1 (s, C-6a), 115.4 (t, CH_2CH_2CH=CH_2), 114.6 (d, C-3), 109.6 (d, C-7), 108.4 (d, C-10), 101.9 (t, OCH_2O), 79.1 (d, C-1), 68.3 (d, C-2), 66.5 (d, C-4a), 55.8 (t, C-12), 43.3 (q, NMe), 40.6 (d, C-10b), 33.2 (t, CH_2CH_2CH=CH_2), 28.5 (t, CH_2CH_2CH=CH_2), 27.7 (t, C-11). IR (neat, cm^{-1}): 2922, 2850, 1729, 1615, 1481, 1291, 1251, 1160, 1118, 1034, 936, 785, 652, 610. HRMS m/z 398.1606 (calcd for $C_{22}H_{24}NO_6$ $[M + 1]^+$ 398.1604).

4.3.9. 2-Oxohippeastrine (**10**)

To a solution of 37.9 mg (0.12 mmol) of **1** in acetone (2 mL) in an ice bath, was added the Jones reagent dropwise, until the solution turned orange. The reaction mixture was stirred for 30 min and then isopropanol was added. The solution was filtered through Florisil and the residue was concentrated and purified by preparative TLC using DCM:MeOH 9:1 as eluent, to yield 20 mg (55%) of **10** as an amorphous white solid. $[\alpha]_D^{20}$ = +16.0 (c 0.3, EtOH). ^{1}H NMR ($CDCl_3$) δ 7.50 (1H, s, H-7), 6.92 (1H, s, H-10), 6.10 (2H, s, OCH_2O), 6.09 (1H, s, H-3), 4.59 (1H, s, H-1), 3.28 (1H, m, H-4a), 3.18 (2H, s, H-12, H-10b), 2.80 (2H, d, J = 8.4 Hz, H-11), 2.42 (1H, dd, J = 9.3 Hz, J = 18.4 Hz, H-12), 2.07 (3H, s, NMe). ^{13}C NMR ($CDCl_3$) δ 188.5 (s, C-2), 163.0 (s, C-6), 152.1 (s, C-9), 148.3 (s, C-8), 135.8 (s, C-10a), 121.1 (d, C-3), 117.8 (s, C-6a), 109.7 (d, C-7), 108.4 (d, C-10), 102.1 (t, OCH_2O), 77.8 (d, C-1), 67.4 (d, C-4a), 56.2 (t, C-12), 45.5 (d, C-10b), 43.3 (q, NMe), 29.7 (t, C-11). IR (neat, cm^{-1}): 2957, 2927, 2855, 1730, 1646, 1454, 1386, 1251, 1170, 1065, 1035, 938, 755. HRMS m/z 313.0962 (calcd for $C_{17}H_{15}NO_5$ $[M]^+$ 313.0950).

4.3.10. Reaction of hippeastrine with thionyl chloride

To a solution of 20 mg (0.064 mmol) of **1** in 5 mL of dry DCM, 100 μL (20 equiv) of thionyl chloride were added. The mixture was refluxed for 5 h. Then, the solvent was evaporated and the residue was purified using preparative TLC with DCM:MeOH 9:1 as eluent to yield 8 mg (43%) of **11** and 10 mg (47%) of **12**.

4.3.11. 3,4-Dihydro-2-deoxy-2,3,4,11-tetradehydrohippeastrine (**11**)

White solid. $[\alpha]_D^{20}$ = +52.5 (c 0.8, EtOH). ^{1}H NMR ($CDCl_3$) δ 7.53 (1H, s, H-7), 6.98 (1H, s, H-10), 6.57 (1H, d, J = 9.5 Hz, H-3), 6.06 (2H, s, OCH_2O), 5.99 (1H, dd, J = 5.7 Hz, J = 9.6 Hz, H-11), 5.67 (1H, d, J = 1.9 Hz, H-2), 4.97 (1H, dd, J = 2.0 Hz, J = 3.3 Hz, H-1), 3.96 (1H, d, J = 15.4 Hz, H-12), 3.55 (1H, m, H-4a), 3.30 (1H, d, J = 15.5 Hz, H-12), 2.80 (1H, dd, J = 3.2 Hz, J = 7.5 Hz, H-10b), 2.04 (3H, s, NMe). ^{13}C NMR ($CDCl_3$) δ 164.1 (s, C-6), 151.8 (s, C-9), 147.6 (s, C-8), 137.3 (s, C-4), 137.0 (C-10a), 127.6 (d, C-3), 124.5 (d, C-2), 124.0 (d, C-11), 117.4 (s, C-6a), 109.3 (d, C-7), 108.6 (d, C-10), 101.7 (t, OCH_2O), 74.4 (d, C-1), 69.7 (d, C-4a), 63.3 (t, C-12), 45.1 (d, C-10b), 42.8 (q, NMe). IR (neat, cm^{-1}): 3054, 2918, 1712, 1617, 1481, 1390, 1334, 1254, 1122, 1038, 935, 821, 734, 662. HRMS m/z 297.1007 (calcd for $C_{17}H_{15}NO_4$ $[M]^+$ 297.1001).

4.3.12. 4S-Chloro-2,3-dehydro-3,4-dihydro-2-deoxyhippeastrine (**12**)

White solid. $[\alpha]_D^{20}$ = +19.7 (c 1.0, EtOH). ^{1}H NMR ($CDCl_3$) δ 7.54 (1H, s, H-7), 7.00 (1H, s, H-10), 6.23 (1H, d, J = 9.8 Hz, H-3), 6.06 (1H, br s, OCH_2O), 6.05 (1H, br s, OCH_2O), 5.97 (1H, dd, J = 5.5 Hz, J = 9.8 Hz, H-2), 4.82 (1H, dd, J = 3.5 Hz, J = 5.5 Hz, H-1), 3.30 (1H, dd, J = 7.1 Hz, J = 8.7 Hz, H-12), 3.21 (1H, d, J = 10.4 Hz, H-4a), 2.97 (1H, m, H-11), 2.57 (1H, dd, J = 3.2 Hz, J = 10.4 Hz, H-10b), 2.33 (1H, dd, J = 5.5 Hz, J = 13.3 Hz, H-12), 2.24 (1H, m, H-11), 2.19 (3H, s, NMe). ^{13}C NMR ($CDCl_3$) δ 164.3 (s, C-6), 151.6 (s, C-9), 147.6 (s, C-8), 137.0 (s, C-10a), 135.8 (d, C-3), 122.8 (d, C-2), 117.5 (s, C-6a), 109.3 (d, C-7), 108.9 (d, C-10), 101.7 (t, OCH_2O), 73.4 (d, C-1), 71.8 (d, C-4a), 64.6 (s, C-4), 53.6 (t, C-12), 44.4 (q, NMe), 43.8 (d, C-10b), 40.5 (t, C-11). IR (neat, cm^{-1}): 3023, 1644, 1479, 1387, 1293, 1256, 1121, 1035, 551. HRMS m/z 333.0762 (calcd for $C_{17}H_{16}NO_4Cl$ $[M]^+$ 333.0768).

*4.3.13. Ethyl hippeastrinyl carbonate (**13**)*

A solution of 30 mg (0.095 mmol) of **1** in 4 mL of DCM was treated with 46 μL (5 equiv) of ethyl chloroformiate and 4 mL of a 3 M solution of potassium hydroxide. The mixture was stirred at rt for 24 h. After that time, both phases were separated ant the aqueous phase was extracted with DCM. The organic phases were combined, dried with magnesium sulphate and concentrated. Further purification by preparative TLC using DCM:MeOH 9:1 as eluent yielded 4.1 mg (12%) of **13** as an amorphous white solid. $[\alpha]_D^{20} = +48.2$ (*c* 0.35, MeOH). ^{1}H NMR ($CDCl_3$) δ 7.47 (1H, s, H-7), 7.04 (1H, s, H-10), 6.08 (1H, br s, OCH_2O), 6.07 (1H, br s, OCH_2O), 5.70 (1H, s, H-3), 5.30 (1H, s, H-2), 4.68 (1H, s, H-1), 4.20 (2H, q, $J = 7.0$ Hz, OCH_2CH_3), 3.29 (1H, br s, H-12), 2.98 (1H, br s, H-10b), 2.81 (1H, m, H-4a), 2.60 (2H, d, $J = 11.0$ Hz, H-11), 2.35 (1H, dd, $J = 8.8$ Hz, $J = 17.6$ Hz, H-12), 2.11 (3H, s, NMe), 1.30 (3H, t, $J = 7.0$ Hz, OCH_2CH_3). ^{13}C NMR ($CDCl_3$) δ 163.7 (s, C-6), 153.7 (s, OCO_2), 151.8 (s, C-9), 147.7 (s, C-8), 143.2 (s, C-4), 138.5 (s, C-10a), 124.6 (s, C-6a), 117.9 (d, C-3), 109.6 (d, C-7), 108.6 (d, C-10), 101.9 (t, OCH_2O), 78.8 (d, C-1), 71.5 (d, C-2), 66.7 (d, C-4a), 64.3 (t, OCH_2CH_3), 55.8 (t, C-12), 43.2 (q, NMe), 38.9 (d, C-10b), 27.6 (t, C-11), 13.9 (q, OCH_2CH_3). IR (neat, cm^{-1}): 2923, 2852, 1740, 1615, 1481, 1385, 1250, 1197, 1117, 1033, 1007, 938, 875, 784, 670. HRMS *m*/*z* 388.1406 (calcd for $C_{20}H_{22}NO_7$ $[M]^+$ 388.1396).

*4.3.14. 2-(p-Vinylbenzoyl)-hippeastrine (**14**)*

To a solution of 20 mg (0.064 mmol) of **1** in 3 mL of DCM were added 26.5 mg (2 equiv) of 1,3-dicyclohexylcarbodiimide (DCC) and 19 mg (2 equiv) of *p*-vinylbenzoic acid. The mixture was stirred for 24 h. Then, the solvent was evaporated. Further purification of the residue by preparative TLC using DCM:MeOH 9:1 as eluent yielded 17.7 mg (63%) of **14** as an amorphous white solid. $[\alpha]_D^{20} = +228.6$ (*c* 0.07, MeOH). ^{1}H NMR ($CDCl_3$) δ 7.94 (2H, d, $J = 8.2$ Hz, $COC(CH)_2$), 7.46 (2H, d, $J = 8.2$ Hz, $(CH)_2CCH{=}CH_2$), 7.42 (1H, s, H-7), 6.98 (1H, s, H-10), 6.73 (1H, dd, $J = 10.9$ Hz, $J = 17.5$ Hz, $(CH)_2CCH{=}CH_2$), 6.08 (1H, br s, OCH_2O), 6.07 (1H, br s, OCH_2O), 5.85 (1H, d, $J = 17.5$ Hz, $(CH)_2CCH{=}CH_2$), 5.70 (2H, s, H-2, H-3), 5.38 (1H, d, $J = 10.9$ Hz, $(CH)_2CCH{=}CH_2$), 4.73 (1H, s, H-1), 3.19 (1H, m, H-12), 2.93 (1H, d, $J = 9.2$ Hz, H-10b), 2.74 (1H, d, $J = 9.2$ Hz, H-4a), 2.59 (2H, m, H-11), 2.30 (1H, dd, $J = 9.1$ Hz, $J = 18.3$ Hz, H-12), 2.10 (3H, s, NMe). ^{13}C NMR ($CDCl_3$) δ 164.8 (s, C═O), 163.9 (s, C-6), 151.6 (s, C-9), 147.8 (s, C-8), 147.7 (s, C-4), 142.1 (s, $COC(CH)_2$), 138.5 (s, C-10a), 135.6 (d, $(CH)_2CCH{=}CH_2$), 129.8 (d, $COC(CH)_2$), 128.4 (s, $(CH)_2CCH{=}CH_2$), 125.9 (d, $(CH)_2CCH{=}CH_2$), 118.2 (s, C-6a), 116.5 (t, $(CH)_2CCH{=}CH_2$), 114.5 (d, C-3), 109.6 (d, C-7), 108.5 (d, C-10), 101.9 (t, OCH_2O), 79.1 (d, C-1), 68.7 (d, C-2), 66.5 (d, C-4a), 55.9 (t, C-12), 43.3 (q, NMe), 40.9 (d, C-10b), 27.8 (t, C-11). IR (neat, cm^{-1}): 2924, 2360, 1714, 1611, 1480, 1255, 1179, 1098, 1036, 937, 782, 735. HRMS *m*/*z* 446.1605 (calcd for $C_{26}H_{24}NO_6$ $[M + 1]^+$ 446.1604).

*4.3.15. 8,9-Norhippeastrine (**15**)*

To a solution of 27.2 mg (0.086 mmol) of **1** in 8 mL of dry DCM, at 0 °C, was added dropwise 0.13 mL (1.5 equiv) of a solution 1 M of BBr_3 in DCM. After 5 h stirring, the solvent was removed and the residue was chromatographed using preparative TLC with DCM:MeOH 4:1. 14 mg (55%) of **15** were obtained as an amorphous white solid. $[\alpha]_D^{20} = +8.7$ (*c* 0.75, EtOH). ^{1}H NMR (MeOD) δ 7.43 (1H, s, H-7), 6.98 (1H, s, H-10), 5.78 (1H, s, H-3), 4.59 (1H, s, H-1), 4.27 (1H, s, H-2), 3.42 (1H, br s, H-12), 3.18 (1H, d, $J = 9.7$ Hz, H-10b), 3.00 (1H, d, $J = 9.9$ Hz, H-4a), 2.72 (3H, m, H-11, H-12), 2.34 (3H, s, NMe). ^{13}C NMR (MeOD) δ 164.9 (s, C-6), 148.5 (s, C-9), 146.2 (s, C-8), 145.5 (s, C-4), 138.7 (s, C-10a), 122.5 (d, C-3), 118.9 (s, C-6a), 116.0 (d, C-7), 114.5 (d, C-10), 81.2 (d, C-1), 67.5 (d, C-2), 65.6 (d, C-4a), 55.3 (t, C-12), 40.4 (q, NMe), 34.6 (d, C-10b), 25.8 (t, C-11). IR (neat, cm^{-1}): 3444, 2925, 2854, 1644, 1459, 1381, 1265, 576. HRMS *m*/*z* 304.1181 (calcd for $C_{16}H_{18}NO_5$ $[M + 1]^+$ 304.1185).

*4.3.16. 8,9-Nor-8,9-dimethoxyhippeastrine (**16**)*

To 15 mg (0.05 mmol) of **15** in 5 mL of MeOH was added 1 mL of a 2 M solution of trimethylsilyldiazomethane in Et_2O. The mixture was reacted for 24 h. After that period, the solvent was evaporated and the residue was purified by preparative TLC using DCM:MeOH 9:1 as eluent. 8 mg (49%) of **16** were obtained as an amorphous white solid. $[\alpha]_D^{20} = +89.4$ (*c* 0.5, MeOH). ^{1}H NMR ($CDCl_3$) δ 7.52 (1H, s, H-7), 7.04 (1H, s, H-10), 5.68 (1H, s, H-3), 4.64 (1H, s, H-1), 4.41 (1H, s, H-2), 3.94 (3H, s, OMe), 3.92 (3H, s, OMe), 3.20 (1H, ddd, $J = 5.0$ Hz, $J = 9.3$ Hz, $J = 9.3$ Hz, H-12), 2.94 (1H, d, $J = 9.5$ Hz, H-10b), 2.71 (1H, d, $J = 9.5$ Hz, H-4a), 2.55 (2H, br s, H-11), 2.28 (1H, dd, $J = 9.5$ Hz, $J = 18.7$ Hz, H-12), 2.05 (3H, s, NMe). ^{13}C NMR ($CDCl_3$) δ 164.9 (s, C-6), 153.0 (s, C-9), 148.8 (s, C-8), 144.9 (s, C-4), 136.7 (s, C-10a), 118.4 (d, C-3), 116.4 (s, C-6a), 111.7 (d, C-7), 110.7 (d, C-10), 82.2 (d, C-1), 66.8 (d, C-2), 66.8 (d, C-4a), 56.2 (q, OMe), 55.9 (t, C-12), 55.9 (q, OMe), 43.2 (q, NMe), 39.1 (d, C-10b), 27.6 (t, C-11). IR (neat, cm^{-1}): 3400, 2926, 2338, 1716, 1602, 1512, 1461, 1361, 1299, 1220, 1158, 1071, 1024, 975, 878, 733, 640. HRMS *m*/*z* 330.1365 (calcd for $C_{18}H_{20}NO_5$ $[M - 1]^+$ 330.1341).

*4.3.17. Hippeastrine N-oxide (**17**)*

To a solution of 25 mg (0.08 mmol) of **1** in 2 mL of MeOH, at 0 °C, 0.1 mL (11 equiv) of 30% H_2O_2 were added dropwise. The mixture was reacted for 24 h. Then, an excess of manganese dioxide was added and the resulting mixture was filtered through Celite. After evaporation of the solvent, 26 mg (100%) of **17** was obtained as an amorphous white solid. $[\alpha]_D^{20} = +23.0$ (*c* 0.3, MeOH). ^{1}H NMR (MeOD) δ 7.42 (1H, s, H-7), 7.29 (1H, s, H-10), 6.13 (2H, s, OCH_2O), 5.90 (1H, s, H-3), 4.66 (1H, s, H-1), 4.12 (1H, d, $J = 9.5$ Hz, H-2), 3.70 (2H, m, H-12), 3.55 (1H, t, $J = 9.3$ Hz, H-10b), 3.30 (1H, s, H-4a), 3.00 (3H, s, NMe), 2.90 (1H, m, H-11), 2.77 (1H, m, H-11). ^{13}C NMR (MeOD) δ 164.4 (s, C-6), 152.2 (s, C-9), 148.8 (s, C-8), 138.0 (s, C-4), 137.4 (s, C-10a), 122.0 (d, C-3), 118.1 (s, C-6a), 108.9 (d, C-7), 108.7 (d, C-10), 102.5 (t, OCH_2O), 81.6 (d, C-1), 76.7 (d, C-2), 69.1 (t, C-12), 65.3 (t, C-4a), 54.7 (q, NMe), 32.6 (d, C-10b), 24.9 (t, C-11). IR (neat, cm^{-1}): 3405, 2925, 1718, 1619, 1506, 1487, 1455, 1389, 1296, 1257, 1124, 1040, 960, 940, 892, 785, 739. HRMS *m*/*z* 331.1060 (calcd for $C_{17}H_{17}NO_6$ $[M]^+$ 331.1056).

*4.3.18. Preparation of the ammonium derivative (**18**)*

To 25 mg (0.075 mmol) of **17** dissolved in 2 mL of MeOH was added dropwise HCl 12 M until a pH = 1 was reached. After 30 min of stirring, the solvent was evaporated and the residue was dissolved in MeOH and 50 mg (2 equiv) of iron (II) sulphate heptahydrate, at 0 °C. The mixture was reacted for 2 h. After that period, the solvent was removed and the residue was dissolved in 2 mL of a basic solution (pH = 10) of ethylenediamine tetraacetate. This solution was stirred for 30 min and then was extracted with DCM. The organic phase was dried over anhydrous magnesium sulphate and then filtered and concentrated. Further purification of the residue on preparative TLC with DCM:MeOH 9:1 and 1% NH_3 as eluent yielded 10 mg (33%) of **18** as an amorphous white solid. $[\alpha]_D^{20} = -14.4$ (*c* 0.54, EtOH). ^{1}H NMR ($CDCl_3$) δ 7.49 (1H, s, H-7), 6.69 (1H, br s, H-12), 6.31 (1H, s, H-10), 6.02 (2H, s, OCH_2O), 5.98 (1H, d, $J = 2.7$ Hz, H-3), 4.68 (1H, dd, $J = 4.2$ Hz, $J = 5.7$ Hz, H-1), 4.51 (1H, d, $J = 5.7$ Hz, H-4a), 4.13 (1H, dd, $J = 6.8$ Hz, $J = 11.3$ Hz, H-2), 3.62 (3H, s, NMe), 3.59 (1H, br s, H-10b), 3.05 (1H, dd, $J = 6.7$ Hz, 8.9 Hz, H-11), 2.59 (1H, dd, $J = 6.7$ Hz, $J = 8.9$ Hz, H-11). ^{13}C NMR ($CDCl_3$) δ 162.8 (s, C-6), 152.7 (s, C-9), 147.0 (s, C-8), 137.0 (s, C-10a), 123.5 (d, C-12), 123.4 (d, C-3), 117.2 (s, C-6a), 115.0 (s, C-4), 109.5 (d, C-7), 106.7 (d, C-10), 101.8 (t, OCH_2O), 81.6 (d, C-1), 66.5 (d, C-2), 33.8 (d, C-4a), 33.7 (d, C-10b), 33.6 (q, NMe), 29.8 (t, C-11). IR (neat, cm^{-1}): 3442, 2924, 1645, 1482, 1385, 1267, 1034, 569. HRMS *m*/*z* 313.0953 (calcd for $C_{17}H_{15}NO_5$ $[M - 1]^+$ 313.0950).

*4.3.19. Methyl 6-[5-(acetyloxy)-1-methyl-2,3-dihydro-1H-indol-7-yl]-1,3-benzodioxole-5-carboxylate (**19**)*

To a solution of 29.2 mg (0.08 mmol) of **17** in 3 mL of DCM was added 11 μL (2 equiv) of acetic anhydride. After 24 h of reflux, the solvent was evaporated and the residue was purified using a chromatotron with DCM:MeOH 19:1 as eluent to obtain 16 mg (51%) of **4** and 6.4 mg (20%) of **19** as an amorphous white solid. ^{1}H NMR ($CDCl_3$) δ 7.36 (1H, s, H-7), 6.83 (1H, s, H-10), 6.79 (1H, d, J = 3.7 Hz, H-3), 6.47 (1H, d, J = 2.0 Hz, H-1), 6.05 (2H, s, OCH_2O), 3.62 (3H, s, OMe), 3.28 (2H, m, H-12), 2.95 (2H, t, J = 8.2 Hz, H-11), 2.28 (3H, s, NMe), 2.22 (3H, s, $OCOCH_3$). ^{13}C NMR ($CDCl_3$) δ 169.9 (s, $O\underline{C}OCH_3$), 166.9 (s, C-6), 149.7 (s, C-9), 146.6 (s, C-8), 142.2 (s, C-4), 141.2 (s, C-2), 136.0 (s, C-10a), 132.1 (s, C-10b), 131.9 (s, C-4a), 124.0 (s, C-6a), 121.3 (d, C-1), 116.6 (d, C-3), 111.0 (d, C-10), 109.4 (d, C-7), 101.6 (t, OCH_2O), 57.2 (t, C-12), 51.7 (q, OMe), 38.9 (q, NMe), 28.5 (t, C-11), 13.8 (c, $OCO\underline{C}H_3$). IR (neat, cm^{-1}): 2921, 2852, 2360, 1755, 1726, 1615, 1484, 1436, 1372, 1210, 1123, 1034, 930, 874, 733, 670. HRMS m/z 369.1224 (calcd for $C_{20}H_{19}NO_6$ [M]$^+$ 369.1212).

*4.3.20. 3,4R-Dihydrohippeastrine (**20**)*

14.9 mg (0.047 mmol) of **1** in 4 mL of THF were hydrogenated in the presence of catalytic amounts of 10% Pd/C. The reaction mixture was stirred for 5 days. The solution was then filtered through Celite and the solvent was evaporated. After purification by preparative TLC with DCM:MeOH 9:1 as eluent, 3.5 mg (25%) of **20** were obtained as an amorphous white solid. $[\alpha]_D^{20} = -6.4$ (c 0.7, EtOH). ^{1}H NMR ($CDCl_3$) δ 7.51 (1H, s, H-7), 6.95 (1H, s, H-10), 6.05 (2H, s, OCH_2O), 4.64 (1H, t, J = 4.9 Hz, H-1), 3.97 (1H, m, H-2), 3.38 (1H, t, J = 5.0 Hz, H-10b), 3.30 (1H, br s, H-12), 2.70 (1H, dd, J = 5.6 Hz, J = 5.8 Hz, H-4a), 2.34 (2H, m, H-12, H-4), 2.26 (3H, s, NMe), 2.00 (2H, m, H-11), 1.63 (2H, m, H-3). ^{13}C NMR ($CDCl_3$) δ 163.9 (s, C-6), 152.3 (s, C-9), 147.0 (s, C-8), 137.0 (s, C-10a), 118.3 (s, C-6a), 109.5 (d, C-7), 106.4 (d, C-10), 101.8 (t, OCH_2O), 81.0 (d, C-1), 68.4 (d, C-2), 66.7 (d, C-4a), 54.8 (t, C-12), 41.6 (q, NMe), 36.2 (d, C-10b), 34.6 (d, C-4), 33.3 (t, C-3), 29.9 (t, C-11). IR (neat, cm^{-1}): 3403, 2925, 2852, 1710, 1617, 1482, 1450, 1263, 1123, 1036, 934, 757, 651. HRMS m/z 317.1238 (calcd for $C_{17}H_{19}NO_5$ [M]$^+$ 317.1263).

*4.3.21. 3S,4S-Dibromohippeastrine (**21**)*

A solution of 20 mg (0.063 mmol) of **1** in 5 mL of dry DCM was treated with 4 μL (1 equiv) of Br_2. The reaction mixture was stirred for 24 h. Then, the solvent was evaporated and the residue was purified by preparative TLC using DCM:MeOH 9:1 as eluent to obtain 6 mg (20%) of **20** as an amorphous orange solid. $[\alpha]_D^{20} = -13.5$ (c 0.6, EtOH). ^{1}H NMR ($CDCl_3$) δ 7.51 (1H, s, H-7), 6.98 (1H, s, H-10), 6.07 (2H, s, OCH_2O), 5.07 (1H, d, J = 3.3 Hz, H-3), 4.67 (1H, t, J = 3.0 Hz, H-1), 4.30 (1H, t, J = 3.3 Hz, H-2), 3.36 (1H, m, H-12), 2.90 (2H, m, H-4a, H-10b), 2.74 (2H, m, H-11), 2.44 (1H, m, H-12), 2.34 (3H, s, NMe). ^{13}C NMR ($CDCl_3$) δ 164.0 (s, C-6), 152.3 (s, C-9), 148.0 (s, C-8), 138.2 (s, C-10a), 117.5 (s, C-6a), 109.8 (d, C-7), 108.7 (d, C-10), 102.0 (t, OCH_2O), 77.6 (d, C-1), 71.3 (d, C-2), 68.5 (s, C-4), 66.2 (d, C-3), 66.0 (C-4a), 52.9 (d, C-10b), 45.7 (q, NMe), 41.4 (t, C-12), 29.5 (t, C-11). IR (neat, cm^{-1}): 3439, 1645, 1480, 1419, 1288, 1252, 1117, 1030, 604. HRMS m/z 470.9429 (calcd for $C_{17}H_{17}NO_5Br_2$ [M]$^+$ 476.9433).

*4.3.22. 3R-Bromo-4R-hydroxyhippeastrine (**22**)*

20 mg (0.064 mmol) of **1** was added to mixture of 10 mg (1.2 equiv) of *N*-bromoacetamide, 26 μL (0.4 equiv) of a 1 M solution of tin chloride ($SnCl_4$) in DCM, and 1.4 μL (1.2 equiv) of H_2O in 5 mL of MeCN at 0 °C. After 2 h stirring, water was added and the mixture was extracted with DCM. The organic phase was concentrated and purified by preparative TLC, using DCM:MeOH 9:1 as eluent, to yield 4.4 mg (17%) of **22**. White solid. $[\alpha]_D^{20} = -3.3$ (c 0.3, MeOH). ^{1}H NMR (MeOD) δ 7.41 (1H, s, H-7), 7.09 (1H, s, H-10), 6.10 (2H, s, OCH_2O), 4.80 (1H, s, H-3), 4.72 (1H, s, H-1), 4.62 (2H, s, −OH, H-2), 3.63 (2H, m, H-10b, H-12), 3.08 (1H, d, J = 10.9 Hz, H-4a), 2.83 (1H, dd, J = 10.0 Hz, J = 23.0 Hz, H-11), 2.60 (1H, t, J = 10.0 Hz, H-12), 2.14 (1H, dd, J = 6.3 Hz, J = 13.4 Hz, H-11), 1.85 (3H, s, NMe). ^{13}C NMR (MeOD) δ 163.7 (s, C-6), 152.8 (s, C-9), 148.3 (s, C-8), 136.5 (s, C-10a), 117.6 (s, C-6a), 108.4 (d, C-10), 108.2 (d, C-7), 102.3 (t, OCH_2O), 81.4 (d, C-1), 77.5 (s, C-4), 73.5 (d, C-2), 66.9 (d, C-4a), 53.6 (t, C-12), 52.2 (d, C-3), 43.4 (q, NMe), 38.4 (t, C-11), 38.3 (d, C-10b). IR (neat, cm^{-1}): 3431, 2922, 2853, 1718, 1616, 1481, 1450, 1392, 1290, 1255, 1202, 1110, 1035, 934, 883, 734, 667. HRMS m/z 434.0017 (calcd for $C_{17}H_{18}NO_6BrNa$ [M + Na]$^+$ 434.0038).

*4.3.23. 3S,4R-Dihydroxyhippeastrine (**23**)*

A solution of 20 mg (0.064 mmol) of **1** in 5 mL of *t*-butanol/THF/H_2O 7:2:1 was treated with 24.5 mg (3.3 equiv) of N-methylmorpholine N-oxide (NMO) and 0.4 mg (2.5% mol) of osmium tetraoxide. The reaction mixture was stirred for 24 h. Then, a saturated solution of sodium bisulphate was added and the mixture was extracted several times with DCM. The organic phases were dried over anhydrous $MgSO_4$, filtered and concentrated to afford 22 mg (100%) of **23** as an amorphous white solid. $[\alpha]_D^{20} = +18.8$ (c 1.5, MeOH). ^{1}H NMR (MeOD) δ 7.38 (1H, s, H-7), 7.05 (1H, s, H-10), 6.06 (2H, s, OCH_2O), 4.63 (1H, t, J = 4.2 Hz, H-1), 3.97 (1H, dd, J = 4.2 Hz, J = 10.3 Hz, H-2), 3.55 (1H, d, J = 10.3 Hz, H-3), 3.05 (2H, m, H-10b, H-11), 2.60 (2H, m, H-4a, H-12), 1.98 (3H, s, NMe), 1.96 (2H, m, H-11, H-12). ^{13}C NMR (MeOD) δ 164.5 (s, C-6), 152.2 (s, C-9), 147.5 (s, C-8), 136.6 (s, C-10a), 117.2 (s, C-6a), 108.3 (d, C-10), 107.9 (d, C-7), 102.0 (t, OCH_2O), 80.4 (s, C-4), 77.8 (d, C-1), 74.4 (d, C-4a), 72.3 (d, C-3), 71.8 (d, C-2), 54.2 (t, C-12), 41.9 (q, NMe), 38.4 (d, C-10b), 37.1 (t, C-11). IR (neat, cm^{-1}): 3398, 2924, 1705, 1616, 1482, 1450, 1393, 1261, 1123, 1037, 938, 883, 736, 651. HRMS m/z 349.1169 (calcd for $C_{17}H_{19}NO_7$ [M]$^+$ 349.1162).

*4.3.24. General procedure for the preparation of biphenyl derivatives **24**–**27***

A solution of **4** (2-acetylhippeastrine) in 5 mL of CH_3CN was treated with an excess of the corresponding alkyl halide. After 24 h of reflux, the solvent was evaporated and the residue was dissolved in 5 mL of *t*-butanol. An excess of potassium *t*-butoxide (10 equiv) was added and the mixture was refluxed for 4 h. Then a saturated solution of NH_4Cl was added and the mixture was extracted with DCM. The organic phase was concentrated and purified by preparative TLC using DCM:MeOH 99:1 as eluent.

*4.3.25. 6-[2-(Dimethylamino)-3-vinylphenyl]-1,3-benzodioxole-5-carboxylic acid (**24**)*

Following the general procedure, 35.8 mg (0.1 mmol) of **4** was treated with 0.5 mL (8 mmol) of methyl iodide, to yield 22.6 mg (73%) of **24** as an amorphous white solid. ^{1}H NMR ($CDCl_3$) δ 7.42 (2H, s, H-3, H-7), 6.96 (3H, m, H-1, H-2, H-11), 6.62 (1H, s, H-10), 6.05 (2H, s, OCH_2O), 5.61 (1H, d, J = 17.5 Hz, H-12), 5.25 (1H, d, J = 9.7 Hz, H-12), 2.52 (6H, s, NMe). ^{13}C NMR ($CDCl_3$) δ 162.3 (s, C-6), 150.4 (s, C-9), 147.2 (s, C-4a), 146.2 (s, C-8), 138.5 (s, C-10b), 138.3 (s, C-10a), 135.4 (d, C-11), 135.2 (s, C-4), 129.6 (d, C-1), 126.6 (d, C-3), 123.2 (d, C-2), 122.4 (s, C-6a), 113.4 (t, C-12), 111.3 (d, C-10), 110.1 (d, C-7), 101.7 (t, OCH_2O), 43.2 (q, NMe). IR (neat, cm^{-1}): 2912, 1668, 1615, 1489, 1440, 1408, 1359, 1270, 1234, 1142, 1086, 936, 905, 879, 803, 762, 608. HRMS m/z 311.1159 (calcd for $C_{18}H_{17}NO_4$ [M]$^+$ 311.1158).

*4.3.26. 6-[2-(Ethylmethylamino)-3-vinylphenyl]-1,3-benzodioxole-5-carboxylic acid (**25**)*

Following the general procedure, 43.2 mg (0.12 mmol) of **4** was treated with 0.5 mL (6.7 mmol) of ethyl bromide, to yield 11 mg (28%) of **25** as an amorphous white solid. ^{1}H NMR ($CDCl_3$) δ 7.43 (1H, s, H-7), 7.41 (1H, d, J = 7.2 Hz, H-3), 6.97 (3H, m, H-1, H-2, H-11),

6.61 (1H, s, H-10), 6.06 (1H, br s, OCH_2O), 6.04 (1H, br s, OCH_2O), 5.60 (1H, d, $J = 17.5$ Hz, H-12), 5.23 (1H, d, $J = 10.9$ Hz, H-12), 2.69 (2H, m, NCH_2CH_3), 2.55 (3H, s, NMe), 0.88 (3H, t, $J = 7.0$ Hz, NCH_2CH_3). ^{13}C NMR ($CDCl_3$) δ 162.7 (s, C-6), 150.2 (s, C-9), 147.6 (s, C-4a), 146.2 (s, C-8), 139.0 (s, C-10b), 138.7 (s, C-10a), 135.8 (s, C-4), 135.6 (d, C-11), 129.7 (d, C-1), 126.4 (d, C-3), 123.3 (d, C-2), 122.7 (s, C-6a), 113.3 (t, C-12), 110.1 (d, C-7), 101.6 (t, OCH_2O), 49.7 (t, NCH_2CH_3), 39.9 (q, NMe), 13.3 (q, NCH_2CH_3). IR (neat, cm^{-1}): 3394, 3080, 2971, 1685, 1615, 1486, 1443, 1407, 1235, 1135, 1084, 1037, 933, 806, 762, 737. HRMS *m/z* 325.1305 (calcd for $C_{19}H_{19}NO_4$ $[M]^+$ 325.1314).

4.3.27. 6-[2-(Butylmethylamino)-3-vinylphenyl]-1,3-benzodioxole-5-carboxylic acid (**26**)

Following the general procedure described above, 53.1 mg (0.15 mmol) of **4** was treated with 0.5 mL (4.66 mmol) of 1-bromobutane, to yield 14.7 mg (29%) of **26** as an amorphous white solid. ^1H NMR ($CDCl_3$) δ 7.43 (1H, s, H-7), 7.42 (1H, d, $J = 7.2$ Hz, H-3), 6.98 (3H, m, H-1, H-2, H-11), 6.60 (1H, s, H-10), 6.06 (1H, br s, OCH_2O), 6.04 (1H, br s, OCH_2O), 5.61 (1H, dd, $J = 1.1$ Hz, $J = 17.5$ Hz, H-12), 5.23 (1H, d, $J = 10.9$ Hz, H-12), 2.59 (3H, s, NMe), 2.57 (2H, m, $NCH_2CH_2CH_2CH_3$), 1.27 (2H, m, $NCH_2CH_2CH_2CH_3$), 1.06 (2H, m, $NCH_2CH_2CH_2CH_3$), 0.75 (3H, t, $J = 7.2$ Hz, $NCH_2CH_2CH_2CH_3$). ^{13}C NMR ($CDCl_3$) δ 164.6 (s, C-6), 150.2 (s, C-9), 147.8 (s, C-4a), 146.2 (s, C-8), 139.2 (s, C-10b), 139.0 (s, C-10a), 135.8 (s, C-4), 135.6 (d, C-11), 129.8 (d, C-1), 126.3 (d, C-3), 123.1 (d, C-2), 122.8 (s, C-6a), 113.2 (t, C-12), 111.3 (d, C-10), 110.2 (d, C-7), 101.6 (t, OCH_2O), 55.2 (t, $NCH_2CH_2CH_2CH_3$), 40.9 (q, NMe), 30.2 (t, $NCH_2CH_2CH_2CH_3$), 19.8 (t, $NCH_2CH_2CH_2CH_3$), 13.6 (q, $NCH_2CH_2CH_2CH_3$). IR (neat, cm^{-1}): 2957, 2928, 1687, 1615, 1486, 1443, 1407, 1235, 1137, 1082, 1038, 934, 806, 762, 737, 673. HRMS *m/z* 353.1612 (calcd for $C_{21}H_{23}NO_4$ $[M]^+$ 353.1627).

4.3.28. 6-[2-(Methylundecylamino)-3-vinylphenyl]-1,3-benzodioxole-5-carboxylic acid (**27**)

Following the general procedure described above, 42.5 mg (0.12 mmol) of **4** was treated with 0.5 mL (2.24 mmol) of 1-bromoundecane, to yield 24 mg (45%) of **27** as an amorphous white solid. ^1H NMR ($CDCl_3$) δ 7.43 (1H, d, $J = 8.1$ Hz, H-3), 7.42 (1H, s, H-7), 6.97 (3H, m, H-1, H-2, H-11), 6.62 (1H, s, H-10), 6.06 (1H, br s, OCH_2O), 6.04 (1H, br s, OCH_2O), 5.61 (1H, d, $J = 17.6$ Hz, H-12), 5.22 (1H, d, $J = 11.4$ Hz, H-12), 3.95 (2H, t, $J = 6.3$ Hz, $NCH_2(CH_2)_9CH_3$), 2.59 (3H, s, NMe), 1.25 (18H, s, $NCH_2(CH_2)_9CH_3$), 0.87 (3H, t, $J = 5.7$ Hz, $NCH_2(CH_2)_9CH_3$). ^{13}C NMR ($CDCl_3$) δ 167.0 (s, C-6), 149.5 (s, C-9), 148.0 (s, C-4a), 146.1 (s, C-8), 139.4 (s, C-10b), 138.6 (s, C-10a), 136.0 (s, C-4), 135.6 (d, C-11), 129.9 (d, C-1), 125.9 (d, C-3), 123.7 (s, C-6a), 122.9 (d, C-2), 113.0 (t, C-12), 111.1 (d, C-10), 109.8 (d, C-7), 101.5 (t, OCH_2O), 64.7 (t, $NCH_2(CH_2)_9CH_3$), 41.0 (q, NMe), 31.6, 29.2, 29.1, 29.0, 28.3, 28.0, 26.7, 25.6, 22.4 (t, $NCH_2(CH_2)_9CH_3$), 13.9 (q, $NCH_2(CH_2)_9CH_3$). IR (neat, cm^{-1}): 3398, 2924, 2853, 1706, 1619, 1465, 1366, 1247, 1128, 1084, 1039, 937, 905, 807, 760. HRMS *m/z* 451.2713 (calcd for $C_{28}H_{37}NO_4$ $[M]^+$ 451.2723).

4.3.29. General procedure for methylation to obtain derivatives **28** and **29**

To a solution of the corresponding acid in 5 mL of DCM:MeOH 1:1, 0.5 mL of a solution 2 M of trimethylsilyldiazomethane in Et_2O was added. The reaction mixture was stirred for 24 h. Then, the solvent was evaporated and the residue was purified by preparative TLC using DCM:hexane 1:1 as eluent, to yield the corresponding methyl esters **28** and **29**.

4.3.30. Methyl 6-[2-(dimethylamino)-3-vinylphenyl]-1,3-benzodioxole-5-carboxylate (**28**)

11.3 mg (0.036 mmol) of **24** was treated as described in the general procedure, to yield 7.8 mg (67%) of **28**. White solid. ^1H NMR ($CDCl_3$) δ 7.43 (1H, d, $J = 7.7$ Hz, H-3), 7.41 (1H, s, H-7), 6.98 (3H, m, H-1, H-2, H-11), 6.64 (1H, s, H-10), 6.07 (1H, br s OCH_2O), 6.05 (1H, br s, OCH_2O), 5.63 (1H, d, $J = 17.6$ Hz, H-12), 5.25 (1H, d, $J = 10.9$ Hz, H-12), 3.60 (3H, s, OMe), 2.55 (6H, s, NMe). ^{13}C NMR ($CDCl_3$) δ 166.8 (s, C-6), 149.8 (s, C-9), 147.0 (s, C-4a), 146.2 (s, C-8), 138.6 (s, C-10b), 138.5 (s, C-10a), 135.3 (s, C-4), 135.1 (d, C-11), 129.7 (d, C-1), 126.2 (d, C-3), 123.2 (s, C-6a), 123.0 (d, C-2), 113.5 (t, C-12), 111.1 (d, C-10), 109.7 (d, C-7), 101.6 (t, OCH_2O), 51.5 (q, OMe), 43.5 (q, NMe). IR (neat, cm^{-1}): 3060, 2921, 1726, 1617, 1486, 1370, 1245, 1129, 1084, 1037, 930, 880, 759, 728, 675. HRMS *m/z* 325.1302 (calcd for $C_{19}H_{19}NO_4$ $[M]^+$ 325.1314).

4.3.31. Methyl 6-[2-(butylmethylamino)-3-vinylphenyl]-1,3-benzodioxole-5-carboxylate (**29**)

8 mg (0.023 mmol) of **26** was reacted as described in the general procedure, to yield 6.5 mg (79%) of **29** as an amorphous white solid. ^1H NMR ($CDCl_3$) δ 7.45 (1H, d, $J = 7.5$ Hz, H-3), 7.41 (1H, s, H-7), 7.03 (2H, m, H-1, H-11), 6.93 (1H, t, $J = 6.5$ Hz, H-2), 6.63 (1H, s, H-10), 6.07 (1H, br s, OCH_2O), 6.05 (1H, br s, OCH_2O), 5.62 (1H, d, $J = 17.7$ Hz, H-12), 5.22 (1H, d, $J = 10.9$ Hz, H-12), 3.59 (3H, s, OMe), 2.58 (3H, s, NMe), 2.56 (2H, m, $NCH_2CH_2CH_2CH_3$), 1.16 (4H, m, $NCH_2CH_2CH_2CH_3$), 0.77 (3H, t, $J = 7.2$ Hz, $NCH_2CH_2CH_2CH_3$). ^{13}C NMR ($CDCl_3$) δ 166.6 (s, C-6), 149.6 (s, C-9), 148.0 (s, C-4a), 146.3 (s, C-8), 139.3 (s, C-10b), 139.2 (s, C-10a), 135.9 (s, C-4), 135.5 (d, C-11), 129.8 (d, C-1), 125.9 (d, C-3), 123.2 (s, C-6a), 122.9 (d, C-2), 113.0 (t, C-12), 111.2 (d, C-10), 109.6 (d, C-7), 101.5 (t, OCH_2O), 55.1 (t, $NCH_2CH_2CH_2CH_3$), 51.4 (q, OMe), 41.1 (q, NMe), 30.4 (t, $NCH_2CH_2CH_2CH_3$), 19.8 (t, $NCH_2CH_2CH_2CH_3$), 13.6 (q, $NCH_2CH_2CH_2CH_3$). IR (neat, cm^{-1}): 3081, 2926, 1727, 1617, 1485, 1437, 1369, 1244, 1127, 1082, 1037, 931, 878, 804, 761. HRMS *m/z* 367.1784 (calcd for $C_{22}H_{25}NO_4$ $[M]^+$ 367.1784).

4.3.32. General procedure for preparation of dimers **30**–**33**

To a solution of **1** in 5 mL of dry DCM, NEt_3 (1.5–2.5 equiv) and 0.5 equiv of the corresponding acyl dichloride were added. After stirring at rt for 24 h, the solvent was evaporated and the residue was purified by preparative TLC using DCM:MeOH 9:1 as eluent to obtain the corresponding dimers **30**–**33**.

4.3.33. Dihippeastrinyl isophthalate (**30**)

Following the procedure described above, 30 mg (0.095 mmol) of **1** was treated with 20 μL (1.5 equiv) of NEt_3 and 9.7 mg of isophthaloyl chloride. After purification, 7.1 mg (20%) of **30** were obtained as an amorphous white solid. $[\alpha]_D^{20} = +147.3$ (*c* 0.22, MeOH). ^1H NMR ($CDCl_3$) δ 8.58 (1H, s, H-1′), 8.19 (2H, d, $J = 7.6$ Hz, H-3′), 7.51 (1H, t, $J = 7.6$ Hz, H-4′), 7.47 (2H, s, H-7), 7.06 (2H, s, H-10), 6.08 (2H, br s, OCH_2O), 6.06 (2H, br s, OCH_2O), 5.71 (4H, s, H-2, H-3), 4.73 (2H, s, H-1), 3.22 (2H, m, H-12), 2.95 (2H, d, $J = 8.1$ Hz, H-10b), 2.76 (2H, d, $J = 8.1$ Hz, H-4a), 2.60 (4H, m, H-11), 2.30 (2H, dd, $J = 8.9$ Hz, $J = 18.0$ Hz, H-12), 2.10 (6H, s, NMe). ^{13}C NMR ($CDCl_3$) δ 164.1 (s, C=O), 163.8 (s, C-6), 151.7 (s, C-9), 147.8 (s, C-4), 147.8 (s, C-8), 138.3 (s, C-10a), 134.0 (d, C-1′), 130.7 (s, C-2′), 129.9 (d, C-3′), 128.7 (d, C-4′), 118.0 (s, C-6a), 114.2 (d, C-3), 109.5 (d, C-7), 108.7 (d, C-10), 101.9 (t, OCH_2O), 79.0 (d, C-1), 69.2 (d, C-2), 66.5 (d, C-4a), 55.8 (t, C-12), 43.3 (q, NMe), 40.7 (d, C-10b), 27.8 (t, C-11). IR (neat, cm^{-1}): 3054, 2924, 1724, 1614, 1481, 1385, 1292, 1226, 1120, 1035, 938, 732, 564. HRMS *m/z* 761.2357 (calcd for $C_{42}H_{37}N_2O_{12}$ $[M + 1]^+$ 761.2347).

4.3.34. Dihippeastrinyl-pyridine-2,6-dicarboxylate (**31**)

Following the procedure described above, 30 mg (0.095 mmol) of **1** was reacted with 20 μL (1.5 equiv) of NEt_3 and 9.7 mg of 2,6-pyridinedicarbonyl chloride to yield, after purification, 15.5 mg (43%) of **31**. as an amorphous white solid. $[\alpha]_D^{20} = +153.1$ (*c* 0.26, MeOH). ^1H NMR ($CDCl_3$) δ 8.25 (2H, d, $J = 7.7$ Hz, H-2′), 7.98 (1H, t, $J = 7.7$ Hz, H-3′), 7.45 (2H, s, H-7), 7.21 (2H, s, H-10), 6.06 (2H, br s, OCH_2O), 6.05 (2H, br s, OCH_2O), 5.78 (2H, s, H-3), 5.71 (2H, s, H-2),

4.77 (2H, s, H-1), 3.25 (2H, m, H-12), 3.01 (2H, d, $J = 9.3$ Hz, H-10b), 2.72 (2H, d, $J = 9.3$ Hz, H-4a), 2.54 (4H, s, H-11), 2.25 (2H, dd, $J = 9.1$ Hz, $J = 18.5$ Hz, H-12), 2.09 (6H, s, NMe). ^{13}C NMR ($CDCl_3$) δ 163.9 (s, C-6), 163.0 (s, C=O), 151.6 (s, C-9), 148.6 (s, C-4), 147.7 (s, C-1′), 147.6 (s, C-8), 138.8 (s, C-3′), 138.1 (d, C-10a), 128.1 (d, C-2′), 118.0 (s, C-6a), 113.6 (d, C-3), 109.5 (d, C-7), 109.0 (d, C-10), 101.8 (t, OCH_2O), 78.9 (d, C-1), 69.8 (d, C-2), 66.7 (d, C-4a), 55.7 (t, C-12), 43.3 (q, NMe), 40.8 (d, C-10b), 27.8 (t, C-11). IR (neat, cm^{-1}): 2924, 2850, 1723, 1616, 1502, 1481, 1450, 1385, 1291, 1252, 1123, 1034, 937, 843, 783, 734, 699, 651. HRMS m/z 762.2275 (calcd for $C_{41}H_{36}N_3O_{12}$ $[M + 1]^+$ 762.2299).

4.3.35. Dihippeastrinyl terephthalate (**32**)

Following the procedure described above, 25 mg (0.08 mmol) of **1** was treated with 28 μL (2.5 equiv) of NEt_3 and 8.1 mg of terephthaloyl chloride. After purification, 13.2 mg (44%) of **32** were obtained as an amorphous white solid. $[\alpha]_D^{20} = +92.5$ (c 0.43, MeOH). ^{1}H NMR ($CDCl_3$) δ 8.03 (4H, s, H-2′), 7.47 (2H, s, H-7), 6.98 (2H, s, H-10), 6.07 (2H, br s, OCH_2O), 6.05 (2H, br s, OCH_2O), 5.70 (4H, s, H-2, H-3), 4.73 (2H, s, H-1), 3.19 (2H, m, H-12), 2.93 (2H, d, $J = 9.1$ Hz, H-10b), 2.74 (2H, d, $J = 9.1$ Hz, H-4a), 2.59 (4H, m, H-11), 2.30 (2H, dd, $J = 9{:}2$ Hz, $J = 18.5$ Hz, H-12), 2.10 (6H, s, NMe). ^{13}C NMR ($CDCl_3$) δ 164.1 (s, C=O), 163.8 (s, C-6), 151.7 (s, C-9), 147.8 (s, C-4), 147.8 (s, C-8), 138.4 (s, C-10a), 133.4 (s, C-1′), 129.5 (d, C-2′), 118.1 (s, C-6a), 114.3 (d, C-3), 109.6 (d, C-7), 108.6 (d, C-10), 101.9 (t, OCH_2O), 78.9 (d, C-1), 69.3 (d, C-2), 66.5 (d, C-4a), 55.8 (t, C-12), 43.3 (q, NMe), 27.8 (t, C-11). IR (neat, cm^{-1}): 2921, 1722, 1616, 1481, 1386, 1252, 1199, 1102, 1035, 937, 877, 732. HRMS m/z 761.2378 (calcd for $C_{42}H_{37}N_2O_{12}$ $[M + 1]^+$ 761.2347).

4.3.36. Dihippeastrinyl adipate (**33**)

Following the procedure described above, 25 mg (0.08 mmol) of **1** was treated with 28 μL (2.5 equiv) of NEt_3 and 5.8 μL of adipoyl chloride. After purification, 8.2 mg (28%) of **33** were obtained as an amorphous white solid. $[\alpha]_D^{20} = +99.2$ (c 0.38, MeOH). ^{1}H NMR ($CDCl_3$) δ 7.45 (2H, s, H-7), 6.97 (2H, s, H-10), 6.07 (4H, s, OCH_2O), 5.58 (2H, s, H-3), 5.41 (2H, d, $J = 1.6$ Hz, H-2), 4.55 (2H, s, H-1), 3.18 (2H, m, H-12), 2.83 (2H, d, $J = 8.1$ Hz, H-10b), 2.68 (2H, d, $J = 8.1$ Hz, H-4a), 2.53 (4H, s, H-11), 2.29 (4H, $COCH_2(CH_2)_2CH_2CO$), 2.07 (6H, s, NMe), 1.61 (4H, br s, $COCH_2(CH_2)_2CH_2CO$). ^{13}C NMR ($CDCl_3$) δ 171.6 (s, C=O), 163.9 (s, C-6), 151.6 (s, C-9), 147.8 (s, C-8), 147.0 (s, C-4), 138.3 (s, C-10a), 118.0 (s, C-6a), 114.5 (d, C-3), 109.5 (d, C-7), 108.5 (d, C-10), 101.9 (t, OCH_2O), 79.1 (d, C-1), 68.3 (d, C-2), 66.5 (d, C-4a), 55.8 (t, C-12), 43.3 (q, NMe), 33.4 (t, $COCH_2(CH_2)_2CH_2CO$), 27.7 (t, C-11), 23.9 (t, $COCH_2(CH_2)_2CH_2CO$). IR (neat, cm^{-1}): 3055, 2926, 1729, 1615, 1481, 1450, 1385, 1291, 1252, 1122, 1035, 937, 901, 783, 734, 610. HRMS m/z 741.2629 (calcd for $C_{40}H_{41}N_2O_{12}$ $[M + 1]^+$ 741.2660).

Acknowledgements

This work has been partly funded by the Spanish MEC (SAF 2009-13296-C02-01) and by the ICIC (Instituto Canario de Investigación del Cáncer).

Appendix A. Supplementary data

Supplementary data related to this article can be found at http://dx.doi.org/10.1016/j.ejmech.2013.03.018.

References

[1] P. Shetty, Nature 484 (2012) S14–S15.
[2] T. Wu, A.S. Nagle, A.K. Chatterjee, Curr. Med. Chem. 18 (2011) 853–871.
[3] C. Zhu, S.P. Cook, J. Am. Chem. Soc. 134 (2012) 13577–13579.
[4] H. Noedl, L. Chanthap, Y. Se, D. Socheat, S. Peou, K. Schaecher, S. Srivichai, P. Teja-Isavadharm, B. Smith, K. Jongsakul, S. Surasri, M. Fukuda, Trop. Med. Int. Health 12 (2007) 69.
[5] O. Hochino, The Amaryllidaceae alkaloids, in: G. Cordell (Ed.), The Alkaloids, Chemistry and Biology, Academic Press, 1998, p. 323.
[6] E.J. Osorio, S. Berkov, R. Brun, C. Codina, F. Viladomat, F. Cabezas, J. Bastida, Phytochem. Lett. 3 (2010) 161–163.
[7] Y. Toriizuka, E. Kinoshita, N. Kogure, M. Kitajima, A. Ishiyama, K. Otoguro, H. Yamada, S. Omura, H. Takayama, Bioorg. Med. Chem. 16 (2008) 10182–10189.
[8] B. Sener, I. Orhan, J. Satayavivad, Phytother. Res. 17 (2003) 1220–1223.
[9] J.C. Cedrón, D. Gutiérrez, N. Flores, A.G. Ravelo, A. Estévez-Braun, Bioorg. Med. Chem. 20 (2012) 5464–5472.
[10] J.C. Cedrón, D. Gutiérrez, N. Flores, A.G. Ravelo, A. Estévez-Braun, Bioorg. Med. Chem. 18 (2010) 4694–4701.
[11] J.C. Cedrón, J.C. Oberti, A. Estévez-Braun, A.G. Ravelo, M. Del Arco-Aguilar, M. López, J. Nat. Prod. 72 (2009) 112–116.
[12] M. Antoun, N. Mendoza, Y. Rios, G. Proctor, D. Wickramaratne, J. Pezzuto, D. Kinghorn, J. Nat. Prod. 56 (1993) 1423–1425.
[13] A. Evidente, A. Andolfi, A. Abou-Donia, S. Touema, H. Hammoda, E. Shawky, A. Motta, Phytochemistry 65 (2004) 2113–2118.
[14] J.C. Cedrón, M. Del Arco-Aguilar, A. Estévez-Braun, A.G. Ravelo, Chemistry and biology of *Pancratium* alkaloids, in: G.A. Cordell (Ed.), The Alkaloids, Academic Press, 2010, pp. 1–37.
[15] C. Djerassi, R. Ingle, R.A. Bowers, J. Org. Chem. 21 (1956) 1547–1549.
[16] E. Corey, J. Suggs, Tetrahedron Lett. 16 (1975) 2647–2650.
[17] F. Pinacho, R. Carrillo, T. Martín, F. García-Tellado, V.S. Martín, J. Org. Chem. 70 (2005) 10099.
[18] L. Blackburn, X. Wei, J. Taylor, Chem. Commun. 14 (1999) 1337–1338.
[19] L. Jackmam, S. Stemhell, Applications of Nuclear Magnetic Resonance Spectroscopy in Organic Chemistry, second ed., Pergamon Press, Oxford, 1969.
[20] M. Abdel-Monem, P. Portoghese, J. Med. Chem. 15 (1972) 208–210.
[21] P. Forgo, J. Hohmann, J. Nat. Prod. 68 (2005) 1588–1591.
[22] K. McCamley, J. Ripper, R. Singer, P. Scammells, J. Org. Chem. 68 (2003) 9847–9850.
[23] Y. Yeung, X. Gao, E. Corey, J. Am. Chem. Soc. 128 (2006) 9644–9645.
[24] S. Lee, U. Venkatesham, C. Rao, S. Lam, J. Lin, Bioorg. Med. Chem. 15 (2007) 1034–1043.
[25] S. Kobayashi, H. Ishikawa, E. Sasakawa, M. Kihara, T. Shingu, A. Kato, Chem. Pharm. Bull. 28 (1980) 1827–1831.
[26] A. Ali, S. Ross, S. El-Moghazy, J. Nat. Prod. 46 (1983) 350–352.
[27] W. Trager, J. Jensen, Science 193 (1976) 673–675.