

# We are IntechOpen, the world's leading publisher of Open Access books Built by scientists, for scientists

4,800

Open access books available

122,000

International authors and editors

135M

Downloads

Our authors are among the

154

Countries delivered to

TOP 1%

most cited scientists

12.2%

Contributors from top 500 universities



WEB OF SCIENCE™

Selection of our books indexed in the Book Citation Index  
in Web of Science™ Core Collection (BKCI)

Interested in publishing with us?  
Contact [book.department@intechopen.com](mailto:book.department@intechopen.com)

Numbers displayed above are based on latest data collected.  
For more information visit [www.intechopen.com](http://www.intechopen.com)



---

# LC-HRMS for the Identification of $\beta$ -Carboline and Canthinone Alkaloids Isolated from Natural Sources

---

Ana Claudia F. Amaral, Aline de S. Ramos,  
José Luiz P. Ferreira, Arith R. dos Santos,  
Jefferson D. da Cruz, Adélia Viviane M. De Luna,  
Vinicius Vaz C. Nery, Iasmim C. de Lima,  
Marcelo Henrique da C. Chaves and  
Jefferson Rocha de A. Silva

Additional information is available at the end of the chapter

<http://dx.doi.org/10.5772/68075>

---

## Abstract

$\beta$ -carboline and canthinone alkaloids are widely distributed in the Angiosperms. Due to their diverse biological activities, the structures of these alkaloids have been used as important models for the synthesis of novel therapeutic drugs. Combining high-performance liquid chromatography (HPLC) with high-resolution mass spectrometry (HRMS) has provided a valuable tool in the analysis of these alkaloids in, for example, plants, insects, marine creatures, human tissues and body fluids. In this review, we summarized the main  $\beta$ -carboline and canthinone alkaloids studied by liquid chromatography high-resolution mass spectrometry (LC-HRMS) associated with mass analyzers, molecular weight information, mass fragmentation and biological activities, presenting an overview of increasing interest for carboline alkaloids study by LC-HRMS.

**Keywords:** chromatography, indole, mass analyzer, fragmentogram, biological activity, body samples

---

## 1. Introduction

Since ancient times, alkaloids have been used as medicine and in folk medicine for the treatment of different diseases.  $\beta$ -Carboline alkaloids are a group of natural indole alkaloids with different degrees of aromaticity widely distributed in the Angiosperms [1–61]. Canthinones

---

are  $\beta$ -carboline alkaloids that have an additional ring-fusion. Analysis of these alkaloids may be realized by combination of liquid chromatography-high-resolution mass spectrometry (LC-HRMS/MS) to produce information about metabolites contained in complex natural source samples. The LC-HRMS is commonly used as choice technique to analyze and elucidate  $\beta$ -carboline and canthinone alkaloids of the extract mixture and that fact will be approached in this review together with other topics described below.

## 2. Source of $\beta$ -carboline and canthinone alkaloids

In the plant kingdom,  $\beta$ -carboline and canthinone alkaloids are mainly found in Angiosperms, predominantly in Simaroubaceae, Rubiaceae, Rutaceae, Apocynaceae, Amaranthaceae, Annonaceae, Zygophyllaceae and Passifloraceae families [1–61]. **Table 1** shows the alkaloids of these two classes and their natural sources. These alkaloids have been obtained mainly in the studies of isolation of chemical constituents from a natural source, chromatographic LC-HRMS analyses and biological studies.

Alkaloid	Species	Refs.
Annomontine	Annonaceae: <i>Annona foetida</i> Mart., <i>A. montana</i> Macf., <i>A. purpurea</i> Moc & Sessé ex Dunal, <i>A. reticulata</i> L.	[1–3]
Brunneins A–C	Cortinariaceae: <i>Cortinarius brunneus</i> (Pers.) Fr.	[4]
Canthin-2,6-dione	Simaroubaceae: <i>Simaba multiflora</i> A. Juss., <i>S. polyphylla</i> (Cavalcante) W.W. Thomas	[5, 6]
Canthin-6-one	Amaranthaceae: <i>Aerva lanata</i> (L.) A.L. Juss. ex Schultes; Rutaceae: <i>Fagara mayu</i> (Bert.) Engl., <i>F. viridis</i> A. Chev., <i>F. zanthoxyloides</i> Lam., <i>Pentaceras australis</i> Hook. F., <i>Phellodendron amurense</i> Rup., <i>Zanthoxylum belizense</i> Lundell, <i>Z. chiloperone</i> var. <i>angustifolium</i> (Engl.), <i>Z. coreanum</i> Nakai, <i>Z. dipetalum</i> H. Mann, <i>Z. elephantiasis</i> Macfad., <i>Z. flavum</i> Vahl, <i>Z. ovalifolium</i> Tutcher, <i>Z. suberosum</i> C.T. White; Simaroubaceae: <i>Ailanthus altissima</i> Swingle, <i>A. excelsa</i> Roxb., <i>Brucea antidysenterica</i> J.F. Mill., <i>Eurycoma harmandiana</i> Pierre, <i>E. longifolia</i> Jack, <i>Hannoa chlorantha</i> Engl. & Gilg., <i>H. klaineana</i> Pierre & Engl., <i>Odyndea gaboniensis</i> (Pierre) Engler, <i>Picrasma crenata</i> Engl. in Engl. & Prantl	[6–24]
Canthin-6-one-3-N-oxide	Rutaceae: <i>Zanthoxylum chiloperone</i> var. <i>angustifolium</i> (Engl.); Simaroubaceae: <i>Ailanthus altissima</i> Swingle, <i>Eurycoma harmandiana</i> Pierre, <i>Hannoa chlorantha</i> Engl. & Gilg., <i>Simarouba berteriana</i> Krug & Urban	[6, 8, 11, 13, 14, 17, 25]
Canthin-6-one-9-methoxy-5-O- $\beta$ -D-glucopyranoside	Simaroubaceae: <i>Simarouba berteriana</i> Krug & Urban	[25]
$\beta$ -Carboline-1-propionic acid	Amaranthaceae: <i>Aerva lanata</i> (L.) A.L. Juss. ex Schultes; Rutaceae: <i>Zanthoxylum chiloperone</i> var. <i>angustifolium</i> (Engl.); Simaroubaceae: <i>Eurycoma harmandiana</i> Pierre, <i>Simarouba berteriana</i> Krug & Urban	[9, 17, 25]
(E)-O-(6'-Cinnamoyl-4''-hydroxy-3'', 5''-dimethoxy-lyaloside	Rubiaceae: <i>Psychotria suterella</i> Müll. Arg., <i>P. laciniata</i> Vell.	[26–28]
Deppeanol	Rubiaceae: <i>Deppea blumenaviensis</i> (K. Schum.) Lorence	[29]
4,5-Dihydrocanthin-6-one	Simaroubaceae: <i>Ailanthus altissima</i> Swingle	[21]

Alkaloid	Species	Refs.
1,11-Dimethoxycanthin-6-one	Simaroubaceae: <i>Brucea antidysenterica</i> J.F. Mill., <i>Picrasma quassioides</i> (D. Don) Benn., <i>Soulamea pancheri</i> Brongn. & Gris	[21]
4,5-Dimethoxycanthin-6-one	Simaroubaceae: <i>Odyndea gabonensis</i> (Pierre) Engler; <i>Picrasma quassioides</i> (D. Don) Benn., <i>Picrolemma granatensis</i> , <i>Quassia africana</i> (Baill.) Baill.	[21, 22, 30–33]
5,9-Dimethoxycanthin-6-one	Simaroubaceae: <i>Eurycoma longifolia</i> Jack	[24]
9,10-Dimethoxycanthin-6-one	Simaroubaceae: <i>Eurycoma harmandiana</i> Pierre	[17]
Eudistomin G, H, I, P, R, S, T	Polycitoridae: <i>Eudistoma olivaceum</i> Van Name	[34]
Eurycomine E	Simaroubaceae: <i>Picrasma quassioides</i> (D. Don) Benn.,	[35]
11-O- $\beta$ -D-Glucopyranosylcanthin-6-one	Simaroubaceae: <i>Eurycoma longifolia</i> Jack	[24]
10-O- $\beta$ -D-Glucopyranosylxycanthin-6-one	Amaranthaceae: <i>Aerva lanata</i> (L.) A.L. Juss. ex Schultes	[9]
1-(2-Guanidinoethyl)-1,2,3,4-tetrahydro-3-(hydroxymethyl)- $\beta$ -carboline	Nephilidae: <i>Nephila clavipes</i> L.	[23]
Harmaline	Malvaceae: <i>Grewia bicolor</i> Juss.; Passifloraceae: <i>Passiflora edulis</i> f. <i>flavicarpa</i> O. Deg., <i>P. incarnata</i> L.; Zygophyllaceae: <i>Peganum harmala</i> L. <i>Tribulus terrestris</i> L.	[36–39]
Harmalol	Zygophyllaceae: <i>Peganum harmala</i> L.	[36]
Harmane	Ciidae: <i>Coriolus maximus</i> (Mont.) Murrill Malvaceae: <i>Grewia bicolor</i> Juss; Passifloraceae: <i>Passiflora edulis</i> f. <i>flavicarpa</i> O. Deg., <i>P. incarnata</i> L.; Tricholomataceae: <i>Hygrophorus eburneus</i> (Bull.) Fr.; Zygophyllaceae: <i>Tribulus terrestris</i> L.	[4, 37–39]
Harmicine	Apocynaceae: <i>Kopsia griffithii</i> King & Gamble	[40, 41]
Harmine	Malpighiaceae: <i>Banisteriopsis caapi</i> (Spruce ex Griseb.) Morton; Malvaceae: <i>Grewia bicolor</i> Juss.; Passifloraceae: <i>Passiflora edulis</i> f. <i>flavicarpa</i> O. Deg., <i>P. incarnata</i> L.; Zygophyllaceae: <i>Tribulus terrestris</i> L.; <i>Peganum harmala</i> L.	[36–39]
Harmol	Passifloraceae: <i>Passiflora edulis</i> f. <i>flavicarpa</i> O. Deg.; Zygophyllaceae: <i>Peganum harmala</i> L.	[36, 37]
N-Hydroxyannomontine	Annonaceae: <i>Annona foetida</i> Mart.	[1, 2]
10-Hydroxy-antirrhine	Apocynaceae: <i>Ochrosia alyxioidis</i> Guillaumin; Rubiaceae: <i>Psychotria prunifolia</i> (Kunth) Steyerl.	[29]
10-Hydroxy-antirrhine N-oxide	Rubiaceae: <i>Psychotria prunifolia</i> (Kunth) Steyerl.	[29]
1-Hydroxycanthin-6-one	Simaroubaceae: <i>Ailanthus altissima</i> Swingle, <i>Hannoa chlorantha</i> Engl. & Gilg.	[8, 11]
11-Hydroxycanthin-6-one	Simaroubaceae: <i>Ailanthus altissima</i> Swingle	[18]
8-Hydroxycanthin-6-one	Simaroubaceae: <i>Hannoa chlorantha</i> Engl. & Gilg., <i>Odyndea gabonensis</i> (Pierre) Engler	[11, 22]
9-Hydroxycanthin-6-one	Simaroubaceae: <i>Ailanthus altissima</i> Swingle, <i>Eurycoma harmandiana</i> Pierre, <i>Picrolemma granatensis</i> , <i>Simarouba berteriana</i> Krug & Urban	[17, 18, 25, 31]

Alkaloid	Species	Refs.
10-Hydroxycanthin-6-one (aervine)	Amaranthaceae: <i>Aerva lanata</i> (L.) A.L. Juss. ex Schultes; Simaroubaceae: <i>Ailanthus altissima</i> Swingle; <i>Hannoa chlorantha</i> Engl. & Gilg.	[9, 11, 18]
11-Hydroxycanthin-6-one-N-oxide	Simaroubaceae: <i>Simarouba berteriana</i> Krug & Urban	[25]
9-Hydroxycanthin-6-one-N-oxide	Simaroubaceae: <i>S. berteriana</i>	[25]
(R)-5-(1-Hydroxyethyl)-canthine-6-one	Simaroubaceae: <i>Ailanthus altissima</i> Swingle	[18]
10-hydroxy-iso-deppeaninol	Rubiaceae: <i>Psychotria prunifolia</i> (Kunth) Steyerm.	[29]
1-Hydroxy-11-methoxycanthin-6-one	Simaroubaceae: <i>Eurycoma longifolia</i> Jack	[24]
10-Hydroxy-9-methoxycanthin-6-one	Simaroubaceae: <i>E. longifolia</i>	[21, 24]
11-Hydroxy-1-methoxycanthin-6-one	Simaroubaceae: <i>E. longifolia</i>	[21]
11-Hydroxy-10-methoxycanthin-6-one	Simaroubaceae: <i>E. longifolia</i>	[24]
5-Hydroxy-4-methoxycanthin-6-one (nigakinone)	Simaroubaceae: <i>Picrasma excelsa</i> (SW.) Planch. <i>Picrasma quassioides</i> (D. Don) Benn.	[21, 30, 32, 33, 42, 43]
8-Hydroxy-9-methoxycanthin-6-one	Simaroubaceae: <i>Picrolemma granatensis</i> , <i>Simarouba berteriana</i> Krug & Urban	[25, 31]
8-Hydroxymanzamine A	Petrosiidae: <i>Acanthostrongylophora ingens</i> (Thiele); Phloeodictyidae: <i>Pachypellina</i> sp.	[44, 45]
6-Hydroxymetatacarbolines A, B, C, D, E, F, G, H, I	Mycenaceae: <i>Mycena metata</i> (Fr.) Kumm.	[4]
1-(Hydroxymethyl)-3-(2-hydroxypropan-2-yl)-2-(5-methoxy-9H- $\beta$ -carbolin-1-yl) cyclopentanol	Rubiaceae: <i>Galianthe thalictroides</i> (K. Schum.) E.L. Cabral	[46]
Isovallesiachotamine	Rubiaceae: <i>Chimarrhis turbinata</i> DC., <i>Palicourea rigida</i> Kunth, <i>Psychotria bahiensis</i> DC., <i>P. suterella</i> Müll. Arg., <i>P. laciniata</i> Vell.	[27]
Lyaloside	Rubiaceae: <i>Ophiorrhiza japonica</i> Blume, <i>Psychotria suterella</i> Müll. Arg., <i>P. laciniata</i> Vell., <i>Pauridiantha lyalli</i> (Baker) Bremek., <i>Uncaria tomentosa</i> (Willd. ex Schult.) DC., <i>Palicourea adusta</i> Standley	[26–28]
Manzamine A	Petrosiidae: <i>Acanthostrongylophora ingens</i> (Thiele)	[44]
Metatacarbolines A, B, C, D, E, F, G	Mycenaceae: <i>Mycena metata</i> (Fr.) Kumm.	[4]
Methoxyannomontine	Annonaceae: <i>Annona impressivenia</i> Safford, <i>A. Montana</i> Macf., <i>A. reticulata</i> L.; Lauraceae: <i>Neolitsea Konishii</i> (H.) Kan & Sas	[2]
3-Methoxycanthin-2,6-dione	Simaroubaceae: <i>Simaba cuspidata</i> Spruce ex Engl., <i>S. multiflora</i> A. Juss.	[21, 47]
1-Methoxycanthin-6-one	Simaroubaceae: <i>Ailanthus altissima</i> Swingle, <i>Hannoa chlorantha</i> Engl. & Gilg.	[8, 11]
10-Methoxycanthin-6-one (methylaervine)	Amaranthaceae: <i>Aerva lanata</i> (L.) A.L. Juss. ex Schultes	[9]

Alkaloid	Species	Refs.
4-Methoxycanthin-6-one	Amaranthaceae: <i>Charpentiera obovata</i> Gaudich.	[6, 48]
5-Methoxycanthin-6-one	Rutaceae: <i>Zanthoxylum caribaeum</i> Lam., <i>Z. chiloperone</i> var. <i>angustifolium</i> (Engl.), Simaroubaceae: <i>Leitneria floridana</i> Chapm., <i>Odyndea gabonensis</i> (Pierre) Engler	[6, 10, 12–14, 22, 49]
9-Methoxycanthin-6-one	Simaroubaceae: <i>Eurycoma longifolia</i> Jack, <i>Picrolemma granatensis</i> , <i>Simaba polyphylla</i> (Cavalcante) W.W. Thomas, <i>Simarouba berteriana</i> Krug & Urban	[5, 17, 25, 31, 50]
9-Methoxycanthin-6-one-3-N-oxide	Simaroubaceae: <i>Picrolemma granatensis</i>	[31]
7-Methoxy- $\beta$ -carboline-1-propionic acid	Simaroubaceae: <i>Eurycoma harmandiana</i> Pierre	[17]
1-Methoxycarbonyl- $\beta$ -carboline	Simaroubaceae: <i>Picrasma quassioides</i> (D. Don) Benn.	[42]
9-Methoxy-3-methylcanthin-5,6-dione	Simaroubaceae: <i>Eurycoma longifolia</i> Jack	[50]
1-Methoxymethyl- $\beta$ -carboline	Simaroubaceae: <i>E. longifolia</i>	[24]
3-Methylcanthin-2,6-dione	Simaroubaceae: <i>Picrasma quassioides</i> (D. Don) Benn.	[30, 42]
N-Methyltetrahydro- $\beta$ -carboline	Amaranthaceae: <i>Arthropodium leptocladum</i> M. Pop. ex Iljin, <i>Cyathobasis fruticulosa</i> (Bunge) Aellen, <i>Hammada leptoclada</i> Iljin; Elaeagnaceae: <i>Elaeagnus angustifolia</i> L.; Leguminosae: <i>Acacia simplicifolia</i> (L.f.) Schinz & Guillaumin, <i>Anadenanthera peregrina</i> (L.) Speg.; Malpighiaceae: <i>Banisteriopsis rusbyana</i> (Nied.) Morton; Myristicaceae: <i>Gymnacranthera paniculata</i> (A.DC.) Warb., <i>Viola sebifera</i> Aubl., <i>Viola theiodora</i> (Spruce) ex Benth. Warb.; Phyllanthaceae: <i>Flueggea microcarpa</i> Blume; Poaceae: <i>Phalaris aquatica</i> L.; Rubiaceae: <i>Psychotria carthagenensis</i> Jacq.; <i>Psychotria viridis</i> Ruiz & Pav.; Ochnaceae: <i>Testulea gabonensis</i> Pellegr.	[8, 51, 52]
Mitragynine	Rubiaceae: <i>Mitragyna speciosa</i> Korth	[53]
Norharmane	Tricholomataceae: <i>Hygrophorus eburneus</i> (Bull.) Fr.	[54]
14-Oxoprunifoleine	Rubiaceae: <i>Psychotria prunifolia</i> (Kunth) Steyerl.	[29, 55]
Paymantheine	Rubiaceae: <i>Mitragyna speciosa</i> Korth	[53]
Picrasidine L (3-methylcanthin-5,6-dione)	Simaroubaceae: <i>Eurycoma longifolia</i> Jack, <i>Picrasma quassioides</i> (D. Don) Benn., <i>Quassia amara</i> L.	[21, 50]
Picrasidine N, M, U, W, X, Y	Simaroubaceae: <i>Picrasma quassioides</i> (D. Don) Benn.	[21]
Picrasidine O	Simaroubaceae: <i>Eurycoma longifolia</i> Jack, <i>Picrasma quassioides</i> (D. Don) Benn.	[21, 35]
Picrasidine P, V	Simaroubaceae: <i>P. quassioides</i>	[56]
Picrasidine Q (4-hydroxy-5-methoxycanthin-6-one)	Simaroubaceae: <i>P. quassioides</i>	[33]
Psychollatine	Rubiaceae: <i>Psychotria umbellata</i> Thonn.	[27]
Reserpine	Apocynaceae: <i>Rauvolfia hookeri</i> S.R. Sriniv. & Chithra, <i>R. micrantha</i> Hook. f., <i>R. serpentina</i> (L.) Benth. ex Kurz, <i>R. tetraphylla</i> L., <i>R. verticillata</i> (Lour.) Baill., <i>R. vomitoria</i> Afzel	[57]
Speciogynine	Rubiaceae: <i>M. speciosa</i>	[53]
Strictosamide	Rubiaceae: <i>Psychotria nuda</i> (Cham. et Schltdl) Wawra, <i>P. suterella</i> Müll. Arg., <i>P. laciniata</i> Vell., <i>P. prunifolia</i> (Kunth) Steyerl.	[27, 29, 55, 58]

Alkaloid	Species	Refs.
Strictosidinic acid	Rubiaceae: <i>Psychotria umbellate</i> Thonn.	[27]
1,2,3,4-Tetrahydro- $\beta$ -carboline-3-carboxylic acid	Asteraceae: <i>Cichorium endivia</i> L.	[60]
Tetrahydroharmine	Malpighiaceae: <i>Banisteriopsis caapi</i> (Spruce ex Griseb.) Morton; Zygophyllaceae: <i>Peganum harmala</i> L.	[36, 59]
Vallesiachotamine	Rubiaceae: <i>Chimarrhis turbinata</i> DC., <i>Palicourea rigida</i> Kunth, <i>Psychotria bahiensis</i> DC., <i>P. suterella</i> Müll. Arg., <i>P. laciniata</i> Vell.	[27]
Yohimbine	Apocynaceae: <i>Aspidosperma discolor</i> A. DC., <i>A. excelsum</i> Benth, <i>A. eburneum</i> F. Allem, <i>A. marcgravianum</i> Woodson, <i>A. oblongum</i> A. DC.	[61]

**Table 1.** Natural sources of some  $\beta$ -carboline and canthinone alkaloids.

### 3. Alkaloids and biological activity

Many pharmacological properties attributed to  $\beta$ -carboline alkaloids have been described in the literature, which makes it an important class of natural products. Among them, anti-malarial, antileishmanial, trypanocidal, antibacterial and antitumor activities are described [38, 44, 62]. The alkaloids described below have studies of LC-HRMS.

A search for antimalarial drugs describes the activity of the alkaloids (+)-8-hydroxymanzamine A and (+)-manzamine A against chloroquine-sensitive D6 and chloroquine-resistant W2 strains of *Plasmodium falciparum*, with half maximal inhibitory concentration ( $IC_{50}$ ) of 19.5 and 22.0 ng/mL for (+)-8-hydroxymanzamine A, and selectivity index (SI) of 40 and 35, respectively. For (+)-manzamine A, the  $IC_{50}$  values are 20.8 and 25.8 ng/mL, with SI of 47 and 38, respectively [44]. Canthin-6-one and 5-methoxycanthin-6-one, isolated from stem bark of *Zanthoxylum chiloperone* var. *angustifolium*, have  $IC_{50}$  values on chloroquine/mefloquine-resistant and sensitive strains of *P. falciparum* of 2.0–5.3 and 5.1–10.4  $\mu$ g/mL, respectively [10].

The  $\beta$ -carboline alkaloids harmane, harmine and harmaline have been reported to possess antileishmanial activity. Harmane, harmine and harmaline have activity against the amastigote forms of *Leishmania infantum*, with  $IC_{50}$  values of 0.27, 0.23 and 1.16  $\mu$ M, respectively. The harmane and harmaline activities against promastigote forms are less pronounced, with  $IC_{50}$  values of 19.2 and 116.8  $\mu$ M, respectively. Harmine inhibits promastigotes with  $IC_{50}$  of 3.7  $\mu$ M [39]. Strictosamide, alkaloid glycoside isolated from the crude ethanol extracts of roots and branches of *Psychotria prunifolia*, has *in vitro* antiprotozoal activity, especially against promastigotes of *Leishmania amazonensis*, with  $IC_{50}$  values of 40.7  $\mu$ g/mL [29]. The alkaloid (+)-8-hydroxymanzamine A has activity against *Leishmania donovani* with  $IC_{50}$  of 2.5 mg/mL and  $IC_{90}$  of 6.1 mg/mL, whereas (+)-manzamine A is less active, with  $IC_{50}$  of 11.15 mg/mL and  $IC_{90}$  of 31.05 mg/mL [44]. Canthin-6-one, isolated from dichloromethane extract of *Z. chiloperone* stem bark, has antileishmanial activity in BALB/c mice infected with *L. amazonensis*. The intralesional treatment with canthin-6-one is able to decrease by 15.0% a lesion weight and the parasite load by 77.6% when compared with the group of untreated mice [12].

Canthin-6-one also has trypanocidal activity. The alkaloid can provoke 90% of anti-amastigote activity and 79% of trypanomastigotes lysis in assays using *Trypanosoma cruzi*. The alkaloid

5-methoxy-canthin-6-one, isolated from the leaves of the same species, is able to cause 66.4% of anti-amastigote activity and 75% of trypomastigotes lysis [14]. Harmine also has trypanocidal effect against *Trypanosoma brucei*, with  $IC_{50}$  of 74  $\mu$ M [13].

The  $\beta$ -carboline alkaloids have antiproliferative effects against many tumor cell lines. The mechanism of action is probably associated with DNA intercalation, inhibition of topoisomerase I and II, cyclin-dependent kinase (CDK), and I $\kappa$ B kinase complex [40, 62]. In cytotoxicity assays with (+)-8-hydroxymanzamine A and (+)-manzamine A, the  $IC_{50}$  are, respectively, 0.47 and 1.0  $\mu$ g/mL against SK-MEL (human malignant melanoma); 0.78 and 1.0  $\mu$ g/mL against KB (human epidermoid carcinoma); 0.75 and 1.1  $\mu$ g/mL against BT-549 (human breast ductal carcinoma); 0.51 and 4.40  $\mu$ g/mL against HepG<sub>2</sub> (human hepatocellular carcinoma); and 1.25 and 2.15  $\mu$ g/mL against LLC-PK<sub>11</sub> (pig kidney epithelial cells) [44]. Canthin-6-one has in vitro cytotoxicity against many cell lines, such as CHO ( $IC_{50}$  = 7.529  $\mu$ M/mL), HepG2 ( $IC_{50}$  = 4.551  $\mu$ M/mL), HeLa ( $IC_{50}$  = 14.9  $\mu$ M/mL), the human epidermoid carcinoma cell line A-431 ( $IC_{50}$  = 8.393  $\mu$ M/mL), the human breast cancer cell line MCF-7 ( $IC_{50}$  = 5.541  $\mu$ M/mL) [9] and MRC5 (fibroblasts) ( $IC_{50}$  = 12.1  $\mu$ g/mL) [10]. The alkaloid 9-methoxy-canthin-6-one has high in vitro cytotoxicity in MCF-7 and A-549 cells (adenocarcinomic human alveolar basal epithelial cells), with  $IC_{50}$  of 4.5 and <2.5  $\mu$ g/mL, respectively [63].

Antimicrobial activity has also been related to this class of compounds. The alkaloids (+)-8-hydroxymanzamine A and (+)-manzamine A are more potent as antimycobacterial than the control ciprofloxacin, with  $IC_{50}$  values of 0.13 and 0.36  $\mu$ g/mL against *Mycobacterium intracellulare* vs. 0.48  $\mu$ g/mL of ciprofloxacin. However, both substances were inactive against the filamentous fungus *Aspergillus fumigatus* and the Gram-negative bacteria *Escherichia coli* and *Pseudomonas aeruginosa* [44].

Canthin-6-one, 9-hydroxycanthin-6-one and 10-hydroxycanthin-6-one show active in the anti-inflammatory assays involving LPS-induced nitric oxide (NO), a proinflammatory mediator, in RAW 264.7 cells (murine macrophage from blood) with  $IC_{50}$  values ranging from 7.73 to 15.09  $\mu$ M [64].

#### 4. Ionization source and mass analyzers

An analysis of a sample comprises ionization where the ion beam is accelerated by an electric field and then a mass analyzer, a region of the mass spectrometer where the ions are separated according to their mass/charge ratio ( $m/z$ ) [65].

There are many different ionization methods, such as ESI, APCI, FAB, suitable for different applications. Many types of mass analyzers are used according to the type and objectives of the analysis: e.g., dual focus, quadrupole, ion trap, time-of-flight (TOF), Orbitrap and Fourier transform ion cyclotron resonance (FT-ICR) mass analyzers are the magnetic sectors [66]. According to this review, the most used mass analyzers for the analysis of  $\beta$ -carbonyl and canthinone alkaloids are the quadrupole, ion trap, TOF and Orbitrap. The most articles reported TOF as the most used analyzer followed by Orbitrap. TOF is based on the simple idea that the speed of two ions created at the same instant with the same kinetic energy will vary according to the mass of the ion (the lighter ion will be faster), when traveling against



the mass spectrometer detector. The main characteristics are as follows: simultaneous analysis of all produced ions, high sensitivity and high mass resolution, which requires very fast data acquisition and detection systems. An Orbitrap mass analyzer is an ion trap comprising a barrel type electrode and an inner coaxial electrode similar to a reel holding the ions in an orbital motion inside the trap [66].

**Table 2** presents some LC-HRMS data analysis used to identify  $\beta$ -carboline and canthinone alkaloids. These alkaloids are listed in **Table 1** and have publications demonstrating analyses by LC-HRMS.

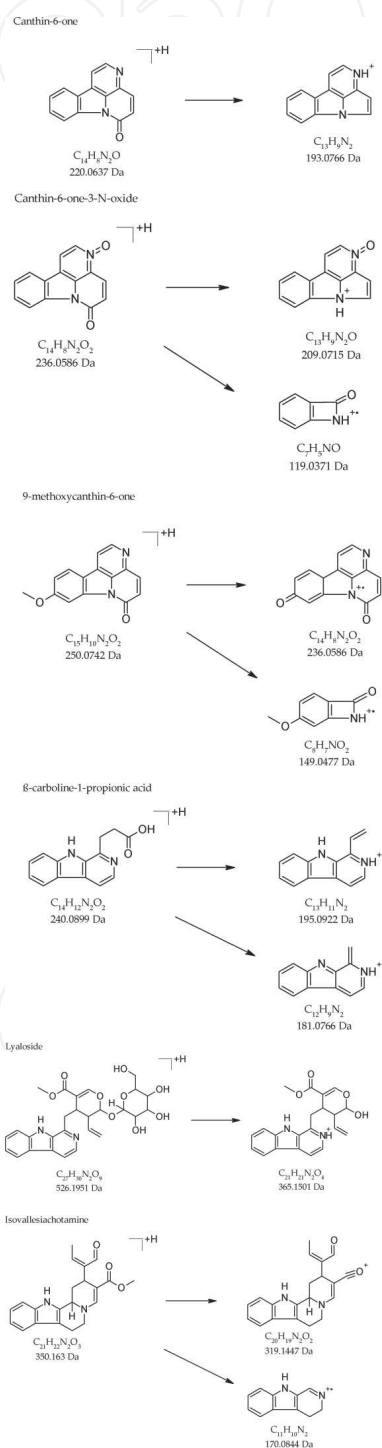
Name	Ionization source and mode	Mass analyzer	Found mass [M+H] <sup>+</sup>	Refs.
Brunnein A	ESI+	FT-ICR	245.0919	[54]
Canthin-6-one	ESI+	Triple QTOF	221.0707	[24]
Canthin-6-one-3N-oxide	ESI+	Triple QTOF	237.0658	[24]
$\beta$ -Carboline-1-propionic acid	ESI+	Triple QTOF	241.0973	[24]
5,9-Dimethoxycanthin-6-one	ESI+	Triple QTOF	281.0913	[24]
9,10-Dimethoxycanthin-6-one	ESI+	Triple QTOF	281.0913	[24]
11-O- $\beta$ -D-Glucopyranosylcanthin-6-one	ESI+	Triple QTOF	399.1202	[24]
1-(2-Guanidinoethyl)-1,2,3,4-tetrahydro-3-(hydroxymethyl)- $\beta$ -carboline	ESI+	Triple QTOF	288.1824	[23]
Harmane	ESI+	FT-ICR	183.09152	[54]
11-Hydroxy-10-methoxycanthin-6-one	ESI+	Triple QTOF	267.0752	[24]
1-Hydroxy-11-methoxycanthin-6-one	ESI+	Triple QTOF	267.0752	[24]
5-Hydroxy-4-methoxycanthin-6-one	ESI+	QTOF	267.0758	[43]
10-Hydroxy-9-methoxycanthin-6-one	ESI+	Triple QTOF	267.0752	[24]
10-Hydroxy-antirrhine	ESI+	Synapt HDMS	313.1920	[29]
10-Hydroxyantirrhine N-oxide derivative	ESI-	Synapt HDMS	327.1712	[29]
11-Hydroxycanthin-6-one	ESI+	Triple QTOF	237.0658	[24]
(R)-5-(1-Hydroxyethyl)-canthine-6-one	DART-SVP+	AccuTOF-TLC	265.1006	[18]
10-Hydroxy-iso-deppeaninol	ESI+	Synapt HDMS	327.1693	[29]
(+)-8-Hydroxymanzamine A	ESI+	FT	565.3608	[44]
(+)-8-Hydroxymanzamine A hydrochloride	ESI+	FT	565.3560	[44]

Name	Ionization source and mode	Mass analyzer	Found mass [M+H] <sup>+</sup>	Refs.
6-Hydroxymetatacarboline A	ESI+	Orbitrap	398.1348	[4]
6-Hydroxymetatacarboline B	ESI+	Orbitrap	526.1934	[4]
6-Hydroxymetatacarboline C	ESI+	Orbitrap	485.1668	[4]
6-Hydroxymetatacarboline D	ESI/MALDI+	Orbitrap	499.1828	[4]
6-Hydroxymetatacarboline E	ESI+	Orbitrap	469.1721	[4]
6-Hydroxymetatacarboline F	ESI+	Orbitrap	497.2032	[4]
6-Hydroxymetatacarboline G	ESI+	Orbitrap	511.2192	[4]
6-Hydroxymetatacarboline H	ESI+	Orbitrap	545.2031	[4]
6-Hydroxymetatacarboline I	ESI+	Orbitrap	511.2192	[4]
7-Hydroxy- $\beta$ -carboline-1-propionic acid	ESI+	Triple QTOF	257.0915	[24]
Isovallesiachotamine	ESI+	TOF	351.1696	[27]
Lyaloside	ESI+	TOF	527.1982	[27]
(+)-8-Manzamine A	ESI+	FT	549.3592	[44]
(+)-Manzamine A hydrochloride	ESI+	FT	549.3550	[44]
Metatacarboline A	ESI+	Orbitrap	382.1398	[4]
Metatacarboline B	ESI+	Orbitrap	510.1987	[4]
Metatacarboline C	ESI+	Orbitrap	469.1721	[4]
Metatacarboline D	ESI+	Orbitrap	483.1879	[4]
Metatacarboline E	ESI+	Orbitrap	453.1770	[4]
Metatacarboline F	ESI+	Orbitrap	481.2084	[4]
Metatacarboline G	ESI+	Orbitrap	495.2241	[4]
9-Methoxy-3-methylcanthin-5,6-dione	ESI+	Triple QTOF	281.0913	[24]
9-Methoxycanthin-6-one	ESI+	Triple QTOF	251.0817	[24]
9-Methoxycanthin-6-one-3N-oxide	ESI+	Triple QTOF	269.0811	[24]
1-Methoxymethyl- $\beta$ -carboline	ESI+	Triple QTOF	213.0990	[24]
Norharmane	ESI+	FT-ICR	169.0760	[54]
Speciogynine	ESI+	Orbitrap	399.22766	[53]
Strictosamide	ESI+	TOF	499.2083	[27]
1,2,3,4-Tetrahydro- $\beta$ -carboline-3-carboxylic acid	ESI+	QTOF	217.0963	[67]
Vallesiachotamine	ESI+	TOF	351.1696	[27]
Yohimbine	ESI+	Quadrupole-Orbitrap	355.2016	[68]

**Table 2.** LC-HRMS data of  $\beta$ -carboline and canthinone alkaloids.

## 5. Mass fragmentograms

The observed masses of the fragments in LC-HRMS of the main cited  $\beta$ -carboline and canthinone alkaloids are shown below (**Figure 1**). The principal peaks are shown in the fragmentograms below. The fragments are based on characteristic alkaloid breaks and/or proposals based on mass spectrometry theory.



**Figure 1.** Fragmentogram of  $\beta$ -carboline and canthinone alkaloids.

## 6. Advantages and disadvantages of the LC-HRMS as analytical tool

The natural product research requires the development of fast and robust techniques for the difficult identification of substances in samples of plant extracts. Actually, GC-MS and LC-MS/MS are more used techniques than LC-HRMS for the identification of plant metabolites. However, the advantages of LC-HRMS and the chemical complexity of plant extracts can justify the investment in that newer technique.

Compared with gas chromatography (GC), techniques involving liquid chromatography (LC) have the advantage of being applicable to a wider variety of chemical classes of compounds. In GC, the analytes must be in gaseous form, and some substances must be hydrolyzed or derivatized to lower polarity and increase volatility to be analyzed. In LC, the analytes must be soluble in the liquid mobile phase and works well with polar substances. LC-MS/MS also has higher sensitivity than GC-MS [65, 69].

GC-MS has a single quadrupole mass detector, whereas LC-MS/MS has two quadrupole detectors in tandem. In MS/MS, only one ion from the first detector, frequently the molecular ion is fragmented in the second detector. The selected-ion monitoring (SIM) mode can be applied for GC-MS to increase sensibility and consists of the selection of three of the more abundant ions from the mass spectrum to be measured by the spectrometer and the comparison between the abundance relative ratio of these ions with the predetermined ratio for the suspect substance. The presence of contaminants affects the ion ratio hinders the identification. The selected reaction monitoring (SRM) mode is applied for LC-MS/MS and consists of the selection of some ions fragmented in the second detector. Thus, LC-MS/MS has more specificity than GC-MS, because two substances with the same nominal mass will exhibit different fragmentations in the second detector. Therefore, SIM or SRM is suitable only for targeted substances. GC-MS and LC-MS/MS can also be employed for the analyses of unknown compounds, but only in the full-scan MS mode and with lower sensitivity. Both GC-MS and LC-MS/MS have resolution of 1 atomic mass unit (amu) [65].

The LC-HRMS has the characteristics of the accurate mass measurement of the analytes, which confers many advantages as compared to other techniques of analysis traditionally used. The mass resolution is about 2 ppm, which represents an error of 0.0006 amu for substances of 300 amu [65]. The exact molecular ion mass is associated with an exact molecular formula of the analyte, a valuable structural information. The exact mass is a calculated parameter, while other techniques depend on experimental results for comparison. Therefore, the main advantage of LC-HRMS is that it allows the identification of a wider number of analytes, including unexpected substances in the sample, and does not require reference standards or preexisting MS libraries for comparison [70]. Additionally, LC-TOF/MS can be applied to a larger range of molecular masses (up about 20,000 amu), while LC-MS/MS is indicated for substances up to about 3000 amu [65].

Besides the high mass resolution, the LC-HR/MS has other important advantages. A previous chromatographic treatment of the sample is not required, and a robust method for qualitative analysis can be applied for different and unknown samples, even for the identification of minority substances. Thus, analyses are faster than in LC-MS/MS, because the time in the development of the method is saved. It is especially interesting in natural product studies

which frequently are related to complex mixtures, as in metabolomics, extract authentication and screening studies [70–74].

However, given the high complexity of many substances of plant origin, it is important to carry out analyzes using different ionization modes and both polarities. Most alkaloids are detectable in positive mode, either for ESI or APCI, but the matrix interference is more pronounced. The formation of adducts is possible, more specifically, cationization in positive mode may lead to the formation of alkali adducts, with the formation of multimers that add ions to the mass spectrum [73].

The LC-QTOF/MS adds the high mass resolution to mass fragmentation, which provides higher confidence in identification, although with higher cost. Comparing LC-QTOF/MS to LC triple quadrupole linear ion trap (QqLiT), the first leads to fewer false positives, but the latter has slightly lower detection limits in most situations [74].

Besides the high cost, LC-HRMS has the disadvantage of not differentiating structural isomers, which is important in phytochemistry since substances with more than one stereocenter are common. In those cases, it is necessary to complement with other information, such as retention time and spectroscopic data [73]. Another disadvantage is the rapid saturation of the detector, which requires work with more diluted samples [65]. It is expected that these equipments will become less costly, so that the technique will gain wide use.

## 7. Analysis of alkaloids in body samples by LC-HRMS

Plant species that contain  $\beta$ -carboline alkaloids, including canthinone alkaloids, are widely employed therapeutically or even as a drug of abuse. Given the diversity of the biological activities already described for these alkaloids, including neurological effects, it is necessary to develop techniques for the detection and quantification of these alkaloids and their metabolites in biological fluids and tissues, as a tool for toxicological analysis and pharmacokinetic studies. This knowledge may also be the starting point for the development of new drugs with potential commercialization.

LC-HRMS is promising in toxicological and analytical studies of metabolism, where substances are often unexpected, and the sample is available in small amount. In addition, it provides rapid analysis and the possibility of using a general method for a wide variety of substances [65, 74–76]. To date, there are few studies using LC-HRMS for the analysis of alkaloids, including  $\beta$ -carboline alkaloids in biological samples, possibly because of the still very high equipment prices. Frequently, LC-MS/MS or GC/MS is used previously, and only after the high-resolution mass is obtained for confirmation.

Biological samples, such as blood, bile, urine, milk, feces and pineal dialysates, consist of a complex matrix, which may cause interference in LC-MS analyses of low or high resolution. Therefore, it is common to submit samples to a pretreatment by solid phase extraction (SPE), using HCX cartridge [53, 76] or C18 cartridges [43, 53, 68]. However, in some cases,

the sample is simply extracted with an organic solvent, such as the procedure described by Shi et al. [32] for the analysis of 5-hydroxy-4-methoxycanthin-6-one and its metabolites, that uses ethyl acetate to extract the analytes from plasma and methanol for feces collected from male Sprague-Dawley rats. There are cases that no pretreatment is required, such as in the analysis of  $\beta$ -carbolines (1,2,3,4-tetrahydro- $\beta$ -carboline, 2-methyl-1,2,3,4-tetrahydro- $\beta$ -carboline, 6-hydroxy-tetrahydro- $\beta$ -carboline, and 6-methoxy-tetrahydro- $\beta$ -carboline), metabolites of dimethyltryptamine and derivatives, in pineal gland microdialysate collected from male Wistar rats [68].

A large variety of phase I metabolites of  $\beta$ -carboline alkaloids, formed by N-decarbonylation, oxidation and methylation, and phase II metabolites, formed by conjugation, such as glucuronides, sulfates and N-acetylcysteine derivatives, are present in body samples. For analysis of phase I metabolites,  $\beta$ -glucuronidase and/or arylsulfatase enzymes can be added to the sample for cleavage of conjugates and to avoid interferences of phase II metabolites [43, 53, 68, 76].

The liquid chromatography step is similar for low and high mass resolution. The separation can occur in TF Hypersil Gold C18 column, 100 mm  $\times$  2.1 mm, 1.9  $\mu$ m [53]; Heder ODS-2 C18 column, 250 mm  $\times$  4.6 mm, 5  $\mu$ m [43]; C18 BEH column, 100 mm  $\times$  2.1 mm, 1.7  $\mu$ m [67]; Zorbax Eclipse Plus C18, 100 mm  $\times$  3.0 mm, 3.5  $\mu$ m [68]; Superspher 60 RP-8 column, 125 mm  $\times$  2 mm, 5  $\mu$ m [76]; Zorbax Eclipse Plus rapid resolution HT C18 column, 50 mm  $\times$  2.1 mm, 1.8  $\mu$ m [75]. The oven temperature is set at 30°C [43], 35°C [53] or 40°C [75]. After pretreatment, samples are frequently diluted in methanol or in mobile phase before injection in LC systems. The mobile phase is frequently a gradient from formic acid (0.05 or 0.1%) in water to acetonitrile, with or without formic acid [43, 68, 76]. This aqueous phase may be replaced by an aqueous solution containing ammonium formate buffer (2.5 or 10 mM) with 0.1% (v/v) formic acid [53, 75]. The organic phase may be 0.1% formic acid in acetone:acetonitrile 20:80 [67]. The solution B of the method developed by Kolmonen et al. [75] consists of 2.5 mM ammonium formate and 0.1% formic acid in 90% acetonitrile. The flow rate varies from 300  $\mu$ L/min [68] to 1 mL/min [43]. The total run time varies from 8 min [75] to 67 min [53].

In general, the MS analyzer, TOF or Orbitrap, employs electrospray ion source. For this class of substances, the positive ionization mode is the most applied (ESI+) [43, 53, 67], although it is more appropriate to use both positive and negative ionization modes in screening analyses to cover more substances [68, 75]. Capillary voltage varied from 3 to 4.5 kV, [43, 53, 67, 68, 75] and resolution varies from 7500 to 60,000 [46, 53]. After the analysis, data processing is necessary with suitable software to help in the identification of metabolites.

Although LC-HRMS has been more used to confirm identification, the technique can be used alone successfully in screening, as the methodology proposed by Kolmonen et al. [75]. The methodology uses LCTOFMS for the search of doping agents in human urine. The method is applicable to at least 207 analytes, including the indole alkaloid strychnine, and may even be used for quantitative analyzes for many of this substances. After an SPE sample pretreatment, the analysis run time is 8 min for each ionization mode, with a total time of 16 min.

Some  $\beta$ -carboline alkaloids and their metabolites have been identified in biological tissues and fluids, such as tetrahydro- $\beta$ -carboline derivatives – present in plant species and also considered

an endogenous alkaloid; [43, 67, 68] speciogynine—isolated from *Mitragyna speciosa*, a plant species used as drug of abuse [53]; 1-methyl-3-carboxy- $\beta$ -carboline—found in cow milk probably derived from the diet and metabolism [76]. The technique is still expanding, and the few works found in the literature indicate a great potential not yet explored.

## 8. Summary

An important class of natural products found in Angiosperms,  $\beta$ -carboline and canthinone alkaloids, has various pharmacological properties and toxic effects. Coupled chromatographic and mass spectrometric techniques can be used to identification of these alkaloids. In this chapter, an approach overview of LC-HRMS applied to chemical complexity of plant extracts and forensic samples containing  $\beta$ -carboline and canthinone alkaloids can be a good choice technique to analyze and elucidate this kind of compounds. In addition, the HRMS/MS fragments of some important  $\beta$ -carboline and canthinone alkaloid are shown in mass fragmentograms schemes. Among important advantages of LC-HRMS, the main one is that it allows the identification of a wider number of analytes, including unexpected substances in the sample, and does not require reference standards or preexisting MS libraries for comparison. This technique can be used alone successfully in screening since it provides rapid analysis and the possibility of using a general method for wide variety of substances.

## Author details

Ana Claudia F. Amaral<sup>1\*</sup>, Aline de S. Ramos<sup>1</sup>, José Luiz P. Ferreira<sup>1</sup>, Arith R. dos Santos<sup>1</sup>, Jefferson D. da Cruz<sup>1</sup>, Adélia Viviane M. De Luna<sup>1</sup>, Vinicius Vaz C. Nery<sup>2</sup>, Iasmim C. de Lima<sup>1</sup>, Marcelo Henrique da C. Chaves<sup>3</sup> and Jefferson Rocha de A. Silva<sup>4</sup>

\*Address all correspondence to: [acamaral@fiocruz.br](mailto:acamaral@fiocruz.br)

1 Laboratório de Plantas Medicinais e Derivados-PN1, Depto de Produtos Naturais, Farmanguinhos – FIOCRUZ, Manguinhos, Brazil

2 Serviços de Métodos Analíticos, Farmanguinhos – FIOCRUZ, Manguinhos, Brazil

3 Divisão de Controle de Qualidade, Farmanguinhos – FIOCRUZ, Brazil

4 Laboratório de Cromatografia – Depto. de Química – UFAM, Japiim, Manaus, Brazil

## References

- [1] Costa EV, Pinheiro MLB, Xavier CM, Silva JRA, Amaral ACF, Souza ADL, Barison A, Campos FR, Ferreira AG, Machado GMC, Leon LLP. A pyrimidine- $\beta$ -carboline and other alkaloids from *Annona foetida* with antileishmanial activity. *Journal of Natural Products*. 2006;**69**:292-294. DOI: 10.1021/np050422s

- [2] Costa EV, Pinheiro MLB, Souza ADL, Santos AG, Campos FR, Ferreira AG, Barison A. Mint: Full NMR analysis of annomontine, methoxyannomontine and N-hydroxyannomontine pyrimidine- $\beta$ -carboline alkaloids. *Magnetic Resonance in Chemistry*. 2008;**46**:69-74. DOI: 10.1002/mrc.2134
- [3] Rejón-Orantes JC, González-Esquinca, La Mora MP, Roldan GR, Cortes D. Mint: Annomontine, an alkaloid isolated from *Annona purpurea*, has anxiolytic-like effects in the elevated plus-maze. *Planta Medica*. 2011;**77**:322-327. DOI: 10.1055/s-0030-1250406
- [4] Jaeger RJR, Lamshöft M, Gottfried S, Spiteller M, Spiteller P. Mint: HR-MALDI MS imaging assisted screening of  $\beta$ -carboline alkaloids discovered from *Mycena metata*. *Journal of Natural Products*. 2013;**76**:127-134. DOI: 10.1021/np300455a
- [5] Saraiva RCG, Pinto AC, Nunomura SM, Pohlit AM. Mint: Triterpenos e alcalóide tipo canthinona dos galhos de *Simaba polyphylla* (Cavalcante) W.W. Thomas (Simaroubaceae). *Química Nova*. 2006;**29**:264-268. DOI: 10.1590/S0100-40422006000200017
- [6] Ohmoto T, Tanaka R, Tamotsu N. Mint: Studies on the constituents of *Ailanthus altissima* Swingle on the alkaloidal constituents. *Chemical & Pharmaceutical Bulletin*. 1976;**24**:1532-1536. DOI: 10.1248/cpb.24.1532
- [7] Galih PR, Esyanti RR. Mint: Effect of immobilization on cell growth and alkaloid contents in cell-aggregate culture of *Eurycoma longifolia* Jack. *International Journal of Chemical, Environmental and Biological Sciences*. 2014;**2**:90-93.
- [8] Anderson L, Harris A, Phillipson JD. Mint: Production of cytotoxic canthin-6-one alkaloids by *Ailanthus altissima* plant cell cultures. *Journal of Natural Products*. 1983;**46**:374-378. DOI: 10.1021/np50027a014
- [9] Bharitkar YP, Hazra A, Poduri NSA, Ash A, Maulik PR, Mondal NB. Mint: Isolation, structural elucidation and cytotoxicity evaluation of a new pentahydroxy-pimarane diterpenoid along with other chemical constituents from *Aerva lanata*. *Natural Product Research*. 2015;**29**:253-261. DOI: 10.1080/14786419.2014.971794
- [10] Cebrián-Torrejón G, Spelman K, Leblanc K, Muñoz-Durango K, Torijano S, Gutiérrez ST, Ferreira ME, Arias AR, Figadère B, Alain Fournet A, Maciuk A, Philippe Grellier P, Nadja B, Cech PN, Poupon E. Mint: The antiplasmodium effects of a traditional South American remedy: *Zanthoxylum chiloperone* var. *angustifolium* against chloroquine resistant and chloroquine sensitive strains of *Plasmodium falciparum*. *The Brazilian Journal of Pharmacognosy*. 2011;**21**:652-661. DOI: 10.1590/S0102-695X2011005000104
- [11] Diakanamwa C, Diallo B, Vanhaelen-Fastré R, Vanhaelen M, Jaziri M, Homès J. Mint: Gas liquid chromatographic determination of canthinone alkaloids in *Hannoa chlorantha* root bark and tissue cultures. *Phytochemical Analysis*. 1995;**6**:193-195. DOI: 10.1002/pca.2800060403
- [12] Ferreira ME, Arias AR, Ortiz ST, Inchausti A, Nakayama H, Thouvenel C, Hocquemiller R, A. Fournet A. Mint: Leishmanicidal activity of two canthin-6-one alkaloids, two major constituents of *Zanthoxylum chiloperone* var. *angustifolium*. *Journal of Ethnopharmacology*. 2002;**80**:199-202.



- [13] Ferreira ME, Nakayama H, de Arias AR, Schinini A, de Bilbao NV, Serna E, Lagoutte D, Soriano-Agatón F, Poupon E, Hocquemiller R, Fournet A. Mint: Effects of canthin-6-one alkaloids from *Zanthoxylum chiloperone* on *Trypanosoma cruzi*-infected mice. *Journal of Ethnopharmacology*. 2007;**109**:258-263. DOI: 10.1016/j.jep.2006.07.028
- [14] Ferreira ME, Cebrián-Torrejón G, Corrales AS, de Bilbao NV, Rolón M, Gomez CV, Leblanc K, Yaluf G, Schinini A, Torresa S, Serna E, de Arias AR, Poupon E, Fournet A. Mint: *Zanthoxylum chiloperone* leaves extract: First sustainable Chagas disease treatment. *Journal of Ethnopharmacology*. 2011;**133**:986-993. DOI: 10.1016/j.jep.2010.11.032
- [15] Haynes HF, Nelson ER, Price JR. Alkaloids of the Australian Rutaceae: *Pentaceras australis* Hook. f. I. Mint: Isolation of the alkaloids and identification of canthin-6-one. *Australian Journal of Scientific Research*. 1952;**5**:387-400. DOI: 10.1071/CH9520387
- [16] Ikuta A, Nakamura T, Urabe H. Mint: Indolopyridoquinazoline, furoquinoline and canthinone type alkaloids from *Phellodendron amurense* callus tissues. *Phytochemistry*. 1998;**48**:285-291. DOI: 10.1016/S0031-9422(97)01130-8
- [17] Kanchanapoom T, Kasai R, Chumsri P, Hiraga Y, Yamasaki K. Mint: Canthin-6-one and b-carboline alkaloids from *Eurycoma harmandiana*. *Phytochemistry*. 2001;**56**:383-386. DOI: 10.1016/S00319422(00)00363-0
- [18] Kim HM, Lee JS, Szirahiga J, Kwon J, Jeong M, Lee D, Choi J-H, Jang DS. Mint: A new canthinone-type alkaloid isolated from *Ailanthus altissima* Swingle. *Molecules*. 2016;**21**:642. DOI: 10.3390/molecules21050642
- [19] Talaprata SK, Dutta S, Talaprata B. Mint: Alkaloids and terpenoids of *Zanthoxylum ovalifolium*. *Phytochemistry*. 1973;**12**:729-730. DOI: 10.1016/s0031-9422(00)84478-7
- [20] Mitscher LA, Showalter HD, Shipchandler MT, Leu RP, Beal JL. Mint: Antimicrobial agents from higher plants. IV. *Zanthoxylum elephantiasis*. Isolation and identification of canthin-6-one. *Lloydia*. 1972;**35**:177-180.
- [21] Ohmoto T, Koike K. Chapter 3: Canthin-6-one alkaloids. In: Brossi A, editor. *The Alkaloids: Chemistry and Pharmacology*. 1st ed. Elsevier; 1990;**36**:135-170. DOI: 10.1016/s0099-9598(08)60082-6
- [22] Forgacs P, Provost J, Touché A. Mint: Alcaloïdes indoliques d'*Odyndea gabonensis*. *Planta Medica*. 1982;**46**:187-189. DOI: 10.1055/s-2007-971212
- [23] Marques MR, Mendes MA, Tormena CF, Souza BM, Cesar LMM, Rittner R, Palma MS. Mint: Structure determination of a tetrahydro- $\beta$ -carboline of arthropod origin: A novel alkaloid-toxin subclass from the web of spider *Nephila clavipes*. *Chemistry & Biodiversity*. 2005;**2**:525-534. DOI: 10.1002/cbdv.200590034
- [24] Chua LS, Amin NAM, Neo JCH, Lee TH, Lee CT, Sarmidi MR, Aziz RA. Mint: LC-MS/MS based metabolites of *Eurycoma longifolia* (Tongkat Ali) in Malaysia (Perak and Pahang). *Journal of Chromatography B*. 2011;**879**:3909-3919. DOI: 10.1016/j.jchromb.2011.11.002

- [25] Devkota KP, Wilson JA, Henrich CJ, McMahon JB, Reilly KM, Beutler JA. Mint: Compounds from *Simarouba berteriana* which inhibit proliferation of NF1-defective cancer cells. *Phytochemistry Letters*. 2014;**7**:42-45. DOI: 10.1016/j.phytol.2013.09.007
- [26] Aimi N, Seiki H, Sakai S. Mint: Synthesis of lyaloside a prototypal  $\beta$ -carboline glucoindole alkaloid in rubiaceae plants. *Mint: Chemical & Pharmaceutical Bulletin*. 1992;**40**:2588-2590. DOI: 10.1248/cpb.40.2588
- [27] Passos CS, Soldi TC, Abib RT, Apel MA, Simões-Pires C, Marcourt L, Gottfried C, Henriques AT. Mint: Monoamine oxidase inhibition by monoterpene indole alkaloids and fractions obtained from *Psychotria suterella* and *Psychotria laciniata*. *Journal of Enzyme Inhibition and Medicinal Chemistry*. 2013;**28**:611-618. DOI: 10.3109/14756366.2012.666536
- [28] Valverde J, Tamayo G, Hesse M. Mint:  $\beta$ -carboline monoterpene glucosides from *Palicourea adusta*. *Phytochemistry*. 1999;**52**:1485-1489. DOI: 10.1016/S0031-9422(99)00215-0
- [29] Kato L, Oliveira CMA, Faria EO, Ribeiro LC, Carvalho BG, Silva CC, Schuquel ITA, Santin SMO, Nakamura CV, Britta EA, Miranda N, Iglesias AH, Drelprete PG. Mint: Antiprotozoal alkaloids from *Psychotria prunifolia* (Kunth) Steyerf. *Journal of the Brazilian Chemical Society*. 2012;**23**:355-360. DOI: 10.1590/S0103-50532012000200024
- [30] Chen L, Miao X, Peng Z, Wang J, Chen Y. Mint: The pharmacokinetics and bioavailability of three canthinone alkaloids after administration of Kumu injection to rats. *Journal of Ethnopharmacology*. 2016;**182**:235-241. DOI: 10.1016/j.jep.2016.01.019
- [31] Fo EF, Fernandes JB, Vieira PC, da Silva MFGF. Mint: Canthin-6-one alkaloids from *Picrolemma granatensis*. *Phytochemistry*, 1992;**31**:2499-2501.
- [32] Shi Y, Hong C, Xub J, Yang X, Xie N, Feng F, Liu W. Mint: Simultaneous quantification of two canthinone alkaloids of *Picrasma quassioides* in rat plasma by liquid chromatography–tandem mass spectrometry and its application to a rat pharmacokinetic study. *Journal of Chromatography B*. 2015;**986-987**:100-107. DOI: 10.1016/j.jchromb.2015.02.008
- [33] Li H-Y, Koike K, Ohmoto T. Mint: New alkaloids, picrasidines W, X and Y, from *Picrasma quassioides* and X-ray crystallographic analysis of picrasidine Q. *Chemical & Pharmaceutical Bulletin*. 1993;**41**:1807-1811. DOI: 10.1248/cpb.41.1807
- [34] Kinzer KF, Cardellina JH. Mint: Three new  $\beta$ -carbolines from the bermudian tunicate *Eudistoma olivaceum*. *Tetrahedron Letters*. 1987;**28**:925-926. DOI: 10.1016/S0040-4039(00)95875-1
- [35] Sasaki T, Li H, Ohmoto T, Koike K. Mint: Evaluation of canthinone alkaloids as cerebral protective agents. *Bioorganic & Medicinal Chemistry Letters*. 2016;**26**:4992-4995. DOI: 10.1016/j.bmcl.2016.09.006
- [36] Herraiz T, González D, Ancín-Azpilicueta, Arán VJ, Guillén H. Mint:  $\beta$ -carboline alkaloids in *Peganum harmala* and inhibition of human monoamine oxidase (MAO). *Food and Chemical Toxicology*. 2010;**48**:839-845. DOI: 10.1016/j.fct.2009.12.019

- [37] Pereira CAM, Rodrigues TR, Yariwake JH. Mint: Quantification of harman alkaloid in sour passion fruit pulp and seed by a novel dual SBSE-LC/Flu (stir bar sorptive extraction-liquid chromatography with fluorescence detector) method. *Journal of the Brazilian Chemical Society*. 2014;**25**:1472-1483. DOI: 10.5935/0103-5053.20140130
- [38] Manda S, Khan SI, Jain SK, Mohammeda S, Tekwani, BL, Khan IA, Vishwakarma RA, Bharate SB. Mint: Synthesis, antileishmanial and antitrypanosomal activities of N-substituted tetrahydro- $\beta$ -carbolines. *Bioorganic & Medicinal Chemistry Letters*. 2014;**24**:3247-3250. DOI: 10.1016/j.bmcl.2014.06.030
- [39] Di Giorgio C, Delmas F, Ollivier E, Elias R, Balansard G, Timon-David P. Mint: In vitro activity of the  $\beta$ -carboline alkaloids harmine, harmine, and harmaline towards parasites of the species *Leishmania infantum*. *Experimental Parasitology*. 2004;**106**:67-74. DOI: 10.1016/j.exppara.2004.04.002
- [40] Dighe SU, Khan S, Soni I, Jain P, Shukla S, Yadav R, Sen P, Meeran SM, Batra S. Mint: Synthesis of  $\beta$ -carboline-based N-heterocyclic carbenes and their antiproliferative and antimetastatic activities against human breast cancer cells. *Journal of Medicinal Chemistry*. 2015;**58**:3485-3499. DOI: 10.1021/acs.jmedchem.5b00016
- [41] Lood CS, Koskinen AMP. Mint: Harmicine, a tetracyclic tetrahydro- $\beta$ -carboline: From the first synthetic precedent to isolation from natural sources to target-oriented synthesis (review). *Chemistry of Heterocyclic Compounds*. 2015;**50**:1488-1509. DOI: 10.1007/s10593-014-1602-4
- [42] Zhao W, He J, Zhang Y, Ito Y, Su Q, Sun W. Mint: Preparative isolation and purification of alkaloids from *Picrasma quassioides* (D. Don) Benn. by high-speed countercurrent chromatography. *Journal of Liquid Chromatography & Related Technologies*. 2012;**35**:1597-1606. DOI: 10.1080/10826076.2011.621150
- [43] Shi Y, Xia Y, Wang J, He J, Feng F, Liu W. Mint: Metabolic profile of 5-hydroxy-4-methoxycanthin-6-one, a typical canthinone alkaloid, in rats determined by liquid chromatography-quadrupole time-of-flight tandem mass spectrometry together with multiple data processing techniques. *Journal of Pharmaceutical and Biomedical Analysis*. 2016;**129**:60-69. DOI: 10.1016/j.jpba.2016.06.047
- [44] Samoylenko V, Khan SI, Jacob MR, Tekwani BL, Walker LA, Hufford CD, Muhammad I. Mint: Bioactive (+)-manzamine A and (+)-8-hydroxymanzamine A tertiary bases and salts from *Acanthostrongylophora ingens* and their preparations. *Natural Product Communications*. 2009;**4**:185-192.
- [45] Ichiba T, Corgiat JM, Scheuer PJ, Kelly-Borges M. Mint: 8-Hydroxymanzamine A, a beta-carboline alkaloid from a sponge, *Pachypellina* sp. 1994;**57**:168-170. DOI: 10.1021/np50103a027
- [46] Figueiredo PO, Garcez FR, Matos MFC, Perdomo RT, Queiroz LMM, Pott A, Garcez AJS, Garcez WS. Mint: A new cytotoxic  $\beta$ -carboline alkaloid from *Galianthe thalictroides*. *Planta Medica*. 2011;**77**:1852-1854. DOI: 10.1055/s-0030-1271154

- [47] Giesbrecht AM, Gottlieb HE, Gottlieb OR, Goulart MOF, De Lima RA, Sant'Ana AEG. Mint: Canthinones from *Simaba cuspidata*. *Phytochemistry*. 1980;**19**:313-315. DOI: 10.1016/S0031-9422(00)81981-0
- [48] Scheuer PJ, Pattabhiraman TR. Mint: Hawaiian plants studies XIII isolation of a canthinone from a member of family Amaranthaceae. *Lloydia*. 1965;**28**:95-100.
- [49] Readell KE, Seigler DS, Young DA. Mint: 5-methoxycanthin-6-one from *Leitneria floridana* (Simaroubaceae). *Biochemical Systematics and Ecology*. 2003;**31**:167-170. DOI: 10.1016/S0305-1978(02)00074-1
- [50] Choo C-Y, Cham K-L. Mint: High performance liquid chromatography analysis of canthinone alkaloids from *Eurycoma longifolia*. *Planta Medica*. 2002;**68**:382-384. DOI: 10.1055/s-2002-26745
- [51] Poupat C, Ahondt A, Séveneti T. Mint: Alcaloides de *Acacia simplifolia*. *Phytochemistry*. 1976;**15**:2019-2020. DOI: 10.1016/S0031-9422(00)88891-3
- [52] Bahçeevli AK, Kurucu S, Kolak U, Topçüü G, Adou E, Kingston DGI. Mint: Alkaloids and aromatics of *Cyathobasis fruticulosa* (Bunge) Aellen. *Journal of Natural Products*. 2005;**68**:956-958. DOI: 10.1021/np0580006
- [53] Philipp AA, Wissenbach DK, Weber AA, Zapp J, Maurer HH. Mint: Phase I and II metabolites of speciogynine, a diastereomer of the main *Kratom* alkaloid mitragynine, identified in rat and human urine by liquid chromatography coupled to low and high-resolution linear ion trap mass spectrometry. *Journal of Mass Spectrometry*. 2010;**45**:1344-1357. DOI: 10.1002/jms.1848
- [54] Teichert A, Lübken T, Schmidt J, Kuhnt C, Huth M, Porzel A, Wessjohann L, Arnold N. Mint: Determination of  $\beta$ -carboline alkaloids in fruiting bodies of *Hygrophorus* spp. by liquid chromatography/electrospray ionization tandem mass spectrometry. *Phytochemical Analysis*. 2008;**19**:335-341. DOI: 10.1002.pca
- [55] Faria EO, Kato L, Oliveira CMA, Carvalho BG, Silva CC, Sales LS, Schuquel ITA, Silveira-Lacerda EP, Delprete PG. Mint: Quaternary  $\beta$ -carboline alkaloids from *Psychotria prunifolia* (Kunth) Steyerf. *Phytochemistry Letters*. 2010;**3**:113-116. DOI: 10.1016/j.phytol.2010.02.008
- [56] Koike K, Ohmoto T, Ikeda K. Mint:  $\beta$ -Carboline alkaloids from *Picrasma quassioides*. *Phytochemistry*, 1990;**29**:3060-3061. DOI: 10.1016/0031-9422(90)87144-J
- [57] Bindu S, Rameshkumar KB, Kumar B, Singh A, Anilkumar C. Mint: Distribution of reserpine in *Rauvolfia* species from India – HPTLC and LC-MS studies. *Industrial Crops and Products*. 2014;**62**:430-436. DOI: 10.1016/j.indcrop.2014.09.018
- [58] Farias FM, Konrath EL, Zuanazzi JA, Henriques AT. Strictosamide from *Psychotria nuda* (Cham. et Schltdl) Wawra (Rubiaceae). *Biochemical Systematics and Ecology*. 2008;**36**: 919-920. DOI: 10.1016/j.bse.2008.10.002

- [59] Milen M, Hazai L, Kolonits P, Gomory Á, Szántay CS, Fekete J. Mint: Preparation and separation of 1-methyl-1,2,3,4-tetrahydro- $\beta$ -carboline enantiomers by HPLC using a new derivatization reagent. *Journal of Liquid Chromatography & Related Technologies*. 2004;**27**:2921-2933. DOI: 10.1002/hlca.201400035
- [60] Wang F-X, Deng A-J, Li M, Wei J-F, Qin H-L, Wang A-P. Mint: (3S)-1,2,3,4-tetrahydro- $\beta$ -carboline-3-carboxylic acid from *Cichorium endivia*. L. induces apoptosis of human colorectal cancer HCT-8 cells. *Molecules*. 2013;**18**:418-429. DOI: 10.3390/molecules18010418
- [61] Pereira MM, Jácome RLRP, Alcântara AFC, Alves RB, Raslan DS. Mint: Alcalóides indólicos isolados de espécies do gênero *Aspidospema* (Apocynaceae). *Química Nova*. 2007;**30**:970-983. DOI: 10.1590/S0100-40422007000400037
- [62] Kuo PC, Shi LS, Damu AG, Su CR, Huang, CH, Ke CH, Wu JB, Lin AJ, Bastow KF, Lee KH, Wu TS. Cytotoxic and antimalarial  $\beta$ -carboline alkaloids from the roots of *Eurycoma longifolia*. *Journal of Natural Products*. 2003;**66**:1324-1327. DOI: 10.1021/np030277n
- [63] Kim HM, Kim, SJ, Kim HY, Ryu B, Kwak H, Hur J, Choi J-H, Jang DS. Mint: Constituents of the stem barks of *Ailanthus altissima* and their potential to inhibit LPS-induced nitric oxide production. *Bioorganic & Medicinal Chemistry Letters*. 2015;**25**:1017-1020. DOI: 10.1016/j.bmcl.2015.01.034
- [64] Sasaki T, Li W, Higai K, Koike K. Mint: Canthinone alkaloids are novel protein tyrosine phosphatase 1B inhibitors. *Bioorganic & Medicinal Chemistry Letters*. 2015;**25**:1979-1981. DOI: 10.1016/j.bmcl.2015.03.014
- [65] Wu AHB, Gerona R, Armenian P, French D, Petrie M, Lynch KL. Mint: Role of liquid chromatography-high-resolution mass spectrometry (LC-HR/MS) in clinical toxicology. *Clinical Toxicology*. 2012;**50**:733-742. DOI: 10.3109/15563650.2012.713108
- [66] Pavia DL, Lampman GM, Kriz GS, Vyvyan JR. *Introdução à Espectroscopia*. Translation of 4th ed. São Paulo: Cengage Learning; 2010. 716 p.
- [67] Stanstrup J, Schou SS, Holmer-Jensen J, Kjeld H, Dragsted LO. Mint: Whey protein delays gastric emptying and suppresses plasma fatty acids and their metabolites compared to casein, gluten, and fish protein. *Journal of Proteome Research*. 2014;**13**:2396-2408. DOI: 10.1021/pr401214w
- [68] Steinmann D, Ganzera M. Mint: Recent advances on HPLC/MS in medicinal plant analysis. *Journal of Pharmaceutical and Biomedical Analysis*. 2011;**55**:744-757. DOI: 10.1016/j.jpba.2010.11.015
- [69] Vergeynst L, Van Langenhove H, Demeestere K. Mint: Balancing the false negative and positive rates in suspect screening with high-resolution orbitrap mass spectrometry using multivariate statistics. *Analytical Chemistry*. 2015;**87**:2170-2177. DOI: 10.1021/ac503426k
- [70] Kumari T, Sharma C, Bajpai V, Kumar B, Srivastava M, Arya KR. Mint: Qualitative determination of bioactive metabolites through Q-TOF LC/MS in different parts and undifferentiated cultures of *Ulmus wallichiana* Planchon. *Plant Growth Regulation*. 2015;**75**:331-340. DOI: 10.1007/s10725-014-9956-2

- [71] Kluger B, Bueschl C, Neumann N, Stückler R, Doppler M, Chassy AW, Waterhouse AL, Rechthaler J, Kamleitner N, Thallinger GG, Adam G, Krska R, Schuhmacher R. Mint: Untargeted profiling of tracer-derived metabolites using stable isotopic labeling and fast polarity-switching LC-ESI-HRMS. *Analytical Chemistry*. 2014;**86**:11533-11537. DOI: 10.1021/ac503290j
- [72] Cuthbertson DJ, Johnson SR, Piljac-Žegarac J, Kappel J, Schäfer S, Wüst M, Ketchum REB, Croteau RB, Marques JV, Davin LB, Norman G, Lewis NG, Rolf M, Kutchan TM, Soejarto DD, Lange BM. Mint: Accurate mass - time tag library for LC/MS-based metabolite profiling of medicinal plants. *Phytochemistry*. 2013;**91**:187-197. DOI: 10.1016/j.phytochem.2013.02.018
- [73] Thoren KL, Colby JM, Shugarts SB, Wu AHB, Lynch KL. Mint: Comparison of information-dependent acquisition on a tandem quadrupole TOF vs a triple quadrupole linear ion trap mass spectrometer for broad-spectrum drug screening. *Clinical Chemistry*. 2016;**62**:170-178. DOI: 10.1373/clinchem.2015.241315
- [74] Kolmonen M, Leinonen A, Kuuranne T, Pelander A, Ojanperä I. Mint: Generic sample preparation and dual polarity liquid chromatography – time-of-flight mass spectrometry for high-throughput screening in doping analysis. *Drug Testing and Analysis*. 2009;**1**: 250-266. DOI: 10.1002/dta.50
- [75] Rouge P, Cornu A, Biesse-Martin A-S, Lyan B, Rochut N, Graulet B. Mint: Identification of quinolone, carboline and glycinamide compounds in cow milk using HRMS and NMR. *Food Chemistry*. 2013;**141**:1888-1894. DOI: 10.1016/j.foodchem.2013.04.072
- [76] Barker SA, Borjigin J, Lomnicka I, Strassman R. Mint: LC/MS/MS analysis of the endogenous dimethyltryptamine hallucinogens, their precursors, and major metabolites in rat pineal gland microdialysate. *Biomedical Chromatography*. 2013;**27**:1690-1700. DOI: 10.1002/bmc.2981

IntechOpen

