Chapter 1

Introduction

1.1 General introduction

Natural products have shown an important role in the drug discovery process since ancient time as human civilization (Rates, 2001; Strohl, 2000). Higher plants, animals, microbials and minerals with such natural products have provided lead compound for semisynthetic manipulations and templates for total synthetic modification of the novel drugs (Kinghorn, 2001). When compared with libraries of synthetic substances, natural products offer the prospects of discovering a greater number of compounds with sterically more complex structures, particularly in the areas where good synthetic leads do not exist (Raskin *et al*., 2002; Kinghorn, 2001; Mans *et al*., 2000). About 25% of the drugs prescribed worldwide come from plants, 121 such active compounds being in current use. Of the 252 drugs considered as basic and essential by the World Health Organization (WHO), 11% were exclusively of plant origin and a significant number were synthetic drugs obtained from natural precursors (Rates, 2001). Plant-based medicines have been developed through out the human culture. The traditional systems of medicine including the traditional Chinese, the native American and the

Ayurvedic, which have sustained their communication for thousands of years, have provide 74% of the plants-pharmaceutical agents (Cordell *et al*., 2001). However, the potential uses of higher plants as a source of new drugs is still poorly explored. Only a small percentage of 250,000 to 500,000 plant species has been investigated phytochemically and even a smaller percentage has been properly studied for their pharmacological properties. It was estimated that only 5,000 species have been carried out for medical uses (Kinghorn, 2001; Rates, 2001).

Alkaloids were a fascinating group of natural products which belonged to one of the largest and remarkable complicated groups of secondary metabolites comprising 26,900 known structures out of about 150,000 characterized natural products. Sources of alkaloids were plants, fungi, bacteria, marine animals, microorganisms and also in many new sources such as mammals, vertebrates, parasitic organisms and insects. Within the plant kingdom, alkaloids were distributed in 7,231 species of higher plants in 1,730 genera within 186 plant families. The total number of plants-derived alkaloids was 21,120 structures from 1,872 skeletons (Stockigt *et al*., 2002; Cordell *et al.,* 2001). Indole alkaloids were recognized as the important group of alkaloids from plants, comprising 3,767 structures out of 14,658 (Cordell *et al.,* 2001). They were found mainly in eight plant families of which the Apocynaceae, the Loganiaeceae and the Rubiaceae provided the best sources. Many indole alkaloids exhibit important biological activities and have a therapeutical application in the treatment of various diseases. Three main structural types of indole alkaloids were 1) the corynan type, as in

ajamalicine which employed as an antihypertensive 2) the aspidosperma type, as in vindoline which found as single unit of bis-indole alkaloids used in cancer therapy, vinblastine 3) the iboga type, exemplified by catharanthine, the another parts of vinblastine (Dewick, 1998).

The genus *Tabernaemontana* belonging to the family Apocynaceae, incorporates a large number of species distributed mainly over tropical America, Africa and Asia. They were notable for producing a wide variety of indole alkaloids, including many with intriguing carbon skeletons as well as novel biological activities (Kam and Sim, 2003; Prakash *et al*., 2003; Taesotikul, 1997). The plants in this genus are shrubs or small trees with glabrous or sparsely pubescent branches. Leaves are opposite; often have a pair unequal in size and distinct stipule-like ochrea in the axils. Inflorescence is a cyme or solitary flower. Two inflorescences are found at each ramification, occasionally with one missing. Flowers are usually fragrant. Sepals often have collectors inside. Corollas are in mature bud with a narrow tube and globose to ovoid head. Corolla lobes are overlapping to the left (to the right in some extra-Thai species) and the mature corollas are salverform. Stamens are subsessile or with short filaments and completely included in tube. Anthers are narrowly triangular to oblong with the cordate bases. In addition, the anther's apexes are acute and fertile entire length and also free form the pistil. Ovaries of two separate carpels are united into a common style, filiform. Pistil heads are short. Fruits are pair follicles with obliquely ellipsoid to somewhat elongated and ridged or smooth and sometimes

also torulose. Seeds are covered in a fleshy aril, obliquely ellipsoid (Endress and Bruyns, 2000; The Forest Herbarium, 1999).

According to Flora of Thailand (1999) the species of genus *Tabernaemontana* found in Thailand are as follows.

Tabernaemontana bovina Lour.	พุดสูง phut sung (General)
T. bufalina Lour.	เข็มดง khem dong (Tak)
T. corymbosa Wall.	สั่งลา sang la (Ranong)
T. divaricata (L.) R.Br. ex Roem.&Shuilt.	พุดซ้อน phut son (Central)
T. macrocarpa Jack	พุดใต้ phut tai (Penisular)
T. pandacaqui Poir.	พุดฝรั่ง phut farang (Bangkok)
T. pauciflora Blume	พริกป่า prik pa (Rayong)
T. peduncularis Wall.	พุดก้านยาว phut khan yao (General)
T. rostrata Roxb. Ex Wall.	พุดเวียน phut wian (General)

(Smitinan, 2001; The Forest Herbarium, 1999)

(The Forest Herbarium, 1999)

Figure 1.1 *Tabernaemontana peduncularis* Wall.

A. habit B. flower C. dissected flower D. fruit

Figure 1.2 *Tabernaemontana peduncularis* Wall. (habit)

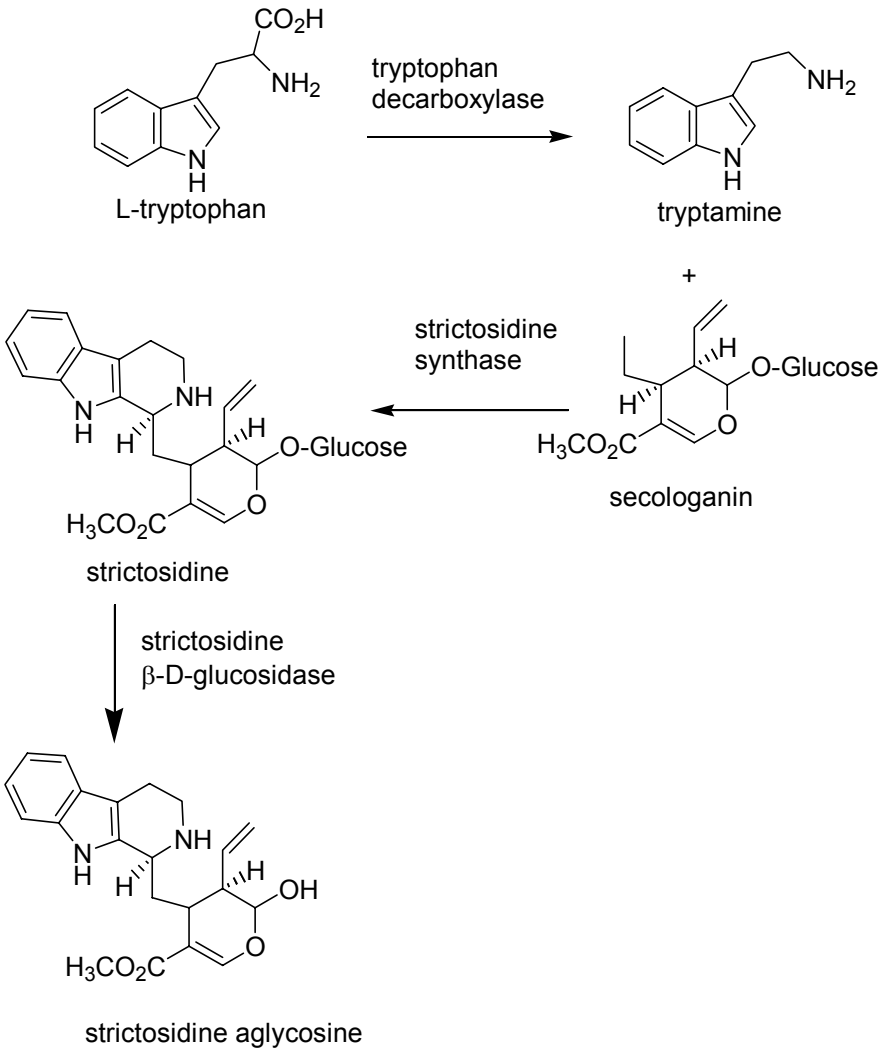
Figure 1.3 *Tabernaemontana peduncularis* Wall. (flowers)

Tabernaemontana peduncularis Wall. has the synonyms as *Ervatamia peduncularis* (Wall.) King & Gamble, *T. graciliflora* Wall. and *E. graciliflora* (Wall.) Lace. The plants are shrubs or small trees to 5 m high. Branchlets are glabrous. Leaves have a petiole which is 4-15 mm long and will be blade papery when dry. Their shapes are elliptic to obovate and the size is 6-33.5 × 1.8-9 cm. In addition, their apex is acuminate or caudate and base is cuneate or decurrent onto petiole and glabrous. Inflorescence is delicate with 7-17(-25) cm long and has a few to many flowers. Sepals are 0.9-1.5 mm long or 1-1.5 × as long as broad. Moreover, their sepals shape is ovate with the apexes are usually obtuse, occasionally acute and ciliate. Corollas are in mature bud outside with pubescent around stamen insertion inside. The stamen tubes are 1-2.6 cm long and not twisted. Furthermore their lobes are 5-9 mm long. Stamens are inserted in the upper half of the corolla tube with the anther apex just beneath the mouth. Anthers are 1.2-2.4 × 0.3-0.7 mm. Ovaries are 0.5-2.0 mm long with style and pistil head in 7.2-19 mm long. Fruits are stipitate and often having a twisted appearance. The fruits apex are caudate, usually strongly reflexed with 0.7-2.2 × 0.3-1.2 cm and has 1-2 seeds. Seeds are obliquely ellipsoid with longitudinal grooves and the normal size is 8-12 × 4-8 mm. (The Forest Herbarium, 1999).

1.2 Biosynthesis and classification of the indole alkaloids in the genus *Tabernaemontana*

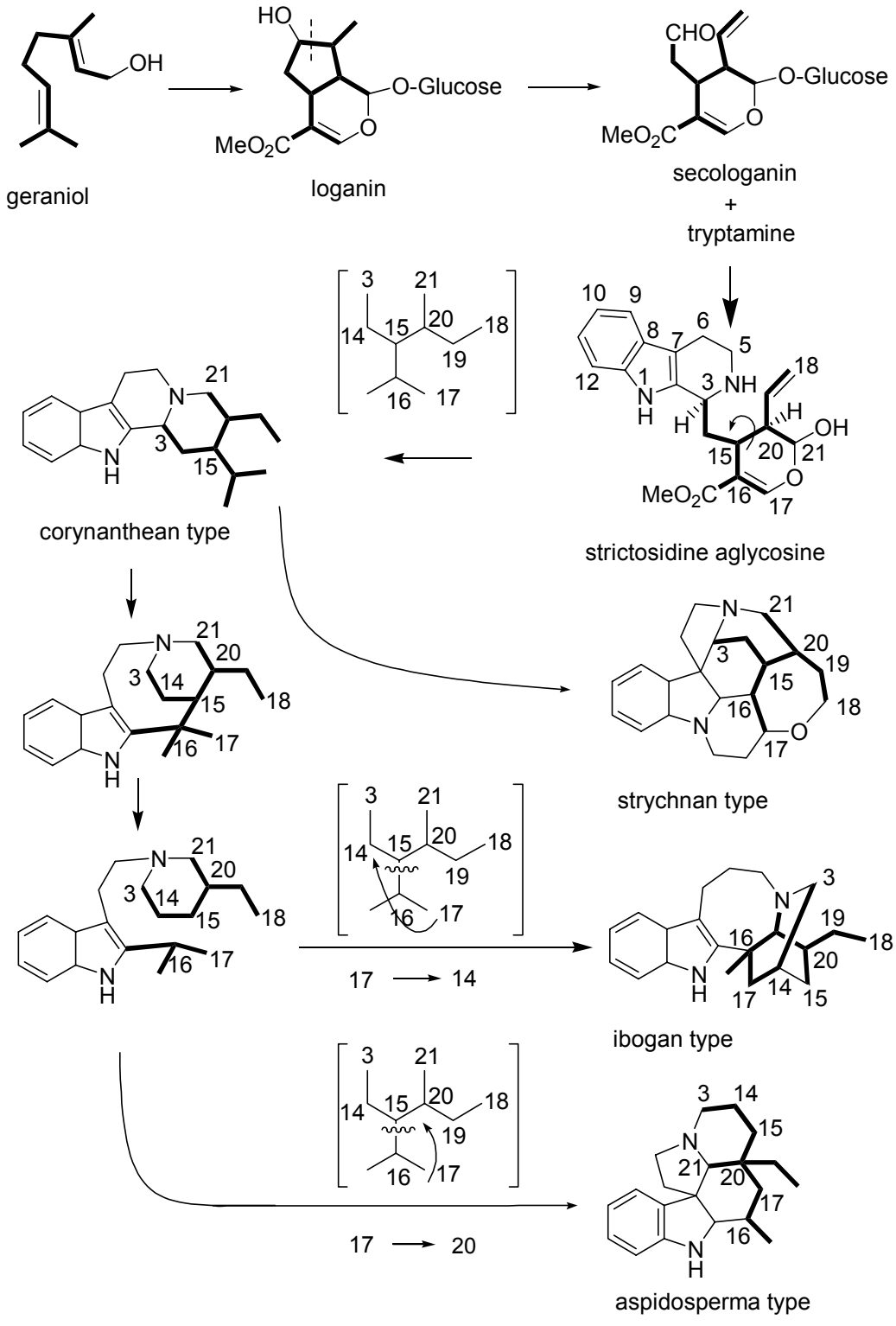
The biosynthesis pathways leading to the monoterpenoid indole alkaloids were established through feeding radiolabeled precursors and followed by extensive product analysis. Therefore, secologanin was found to be a central intermediate in monoterpenoid indole alkaloids biosynthesis as it was incorporated in strictosidine that subsequently rearranged into many types of indole alkaloids. Secologanin, from the monoterpenoid biosynthesis pathways, was derived from isopentenyl diphosphate (IPP) which is the C_5 precursor of all isoprenoids. IPP was synthesized either via the cytosolic mevalonate pathway or via the plastidic 1-deoxy-D-xylulose-5-phosphate (DXP) pathway (alternatively named the non-mevalonate pathway). Each pathway appears to produce unique isoprenoids and operates either independently or simultaneously depending on the tissue type. On the other hand, tryptamine, the indole nucleus precursor of all types of indole alkaloids, was synthesized via the shikimate pathway. Tryptamine was derived from chorismic acid which is the precursor of all aromatic amino acids (Hong *et al*., 2003; Bruneton, 1999; Dewick, 1998; van Beek *et al*., 1984;).

Figure 1.4 Biosynthesis of strictosidine aglycosine



The tryptamine portion could be recognized virtually in all indole alkaloid structures. The remainng fragment was usually a C_9 or C_{10} which came from terpenoid origin, secologanin (**Figure 1.4**). Therefore, the classification of the terpenoid indole alkaloids were established from the secologanin fragment. The relation of the three main groups of terpenoid indole alkaloids, the corynanthean type, the aspidosperma type and the ibogan type could be rationalized in term of rearrangements occurring in the secologanin part of the structure (**Figure 1.5**). Secologanin contained the ten carbon framework which showed in the corynanthean type. Then, the aspidosperma type and the ibogan type arose from the rearrangement of the corynanthean type. Results from the detachment of a three-carbon units rejoined to the remaining C_7 fragment in one of two different ways as shown (**Figure 1.5**). The alkaloids normally appeared to have lost the carbon atom which corresponded to the carboxylate function by hydrolysis/decarboxylation. Thus, the nine-carbon unit framework of the secologanin part was observed (Dewick, 1998; Bruneton, 1999).

Figure 1.5 Biosynthesis homogeneity of selected types of indole alkaloids

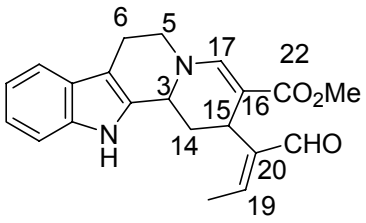


The alkaloids were arranged in 11 main groups according to the structural characteristics of their skeleton as showed in **Table 1.1** The class of miscellaneous alkaloids accommodated those obscured biogenesis (van Beek *et al.,* 1984).

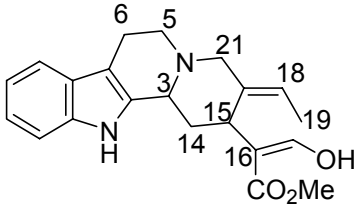
Table 1.1 Classification of the indole alkaloids occurring in *Tabernaemontana* species

Class (abbreviation)	Structure characteristics
Aspidospermatan (A)	C(2)-C(16)-C(15) unit, no C(3)-C(7) bond
Corynanthean (C)	C(2)-C(3)-C(14)-unit, N(4)-C(21) bond
Euburnan (E)	N(1)-C(16)-C(17)-C(20) unit
Ibogan (I)	C(2)-C(16)-C(17)-C(14) unit
Plumeran (P)	C(2)-C(16)-C(17)-C(20) unit
Strychnan (S)	C(2)-C(16)-C(15)-unit, C(3)-C(7) bond
Tacaman (T)	N(1)-C(16)-C(17)-C(14) unit
Vallesiachotaman (V)	C(2)-C(3)-C(14)-unit, N(4)-C(17) bond
Vincosan (D)	C(2)-C(3)-C(14)-unit, no N(4)-C(17) or N(4)-C(21) bond
Bis-indole (B)	Two indole alkaloids attached to each other
Miscellaneous (M)	-

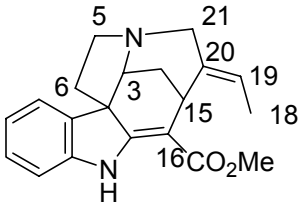
Figure 1.6 Types of the indole alkaloids occuring in *Tabernaemontana* species (van Beek *et al.,* 1984; Dewick, 1998; Bruneton, 1999)



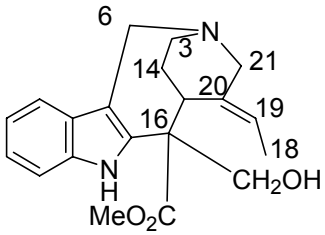
vallesiachotoman type



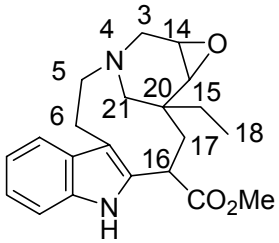
corynanthean type



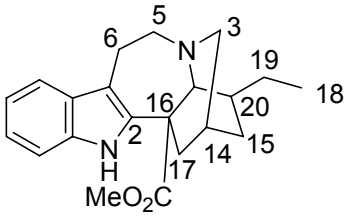
strychnan type



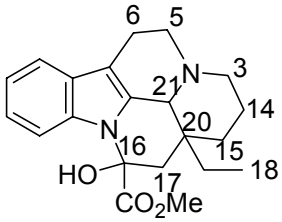
aspidospermatan type



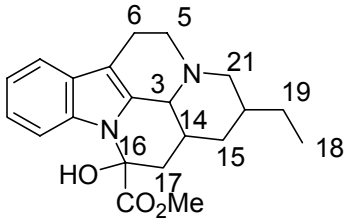
plumeran type



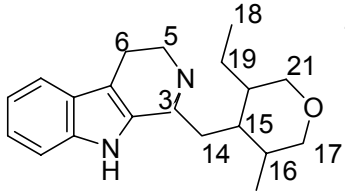
ibogan type



eburnan type



tacaman type

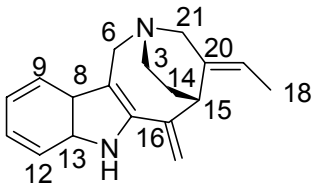


vincosan type

Structures of indole alkaloids from the *Tabernaemontana* species in Thailand

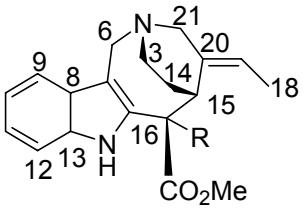
Aspidospermatan Type

A-1



apparicine

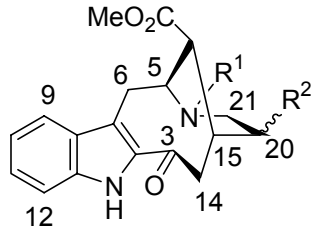
A-2



a R=CH_2OH; vallesamine
b R=CH_2OAc; *O*-acetylvallesamine

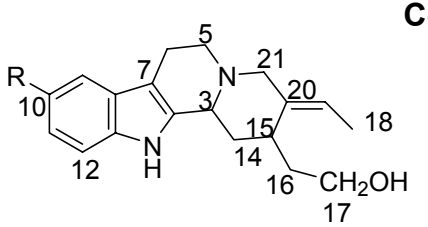
Corynanthean Type

C-1



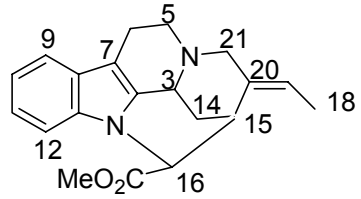
a R^1=Me, R^2=α-C_2H_5; dregamine
b R^1=Me, R^2=β-C_2H_5; tabernaemontanine
c R^1=Me, R^2=(=CH-Me); vobasine
d R_1=H, R^2=(=CH-Me); perivine

C-2



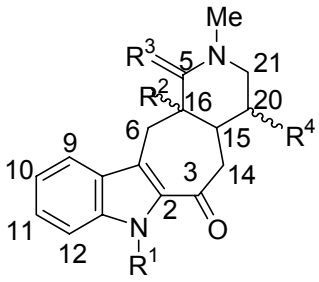
a R= H; geissoschizol
b R=OH; 10-hydroxygeissoschizol

C-3



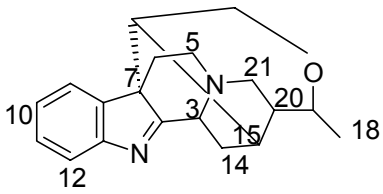
pleiocarpamine

C-4



a R^1=H, R^2=β-CO_2Me, R^3=H_2,
R^4=β-C_2H_5; ervatamine
b R^1=H, R^2=β-CO_2Me, R^3=H_2,
R^4=α-C_2H_5; 20-*epi*-ervatamine
c R^1=H, R^2=β-CO_2Me, R^3=H_2,
R^4=(=CH-Me);19-20-dehydroervatamine
d R^1=H, R^2=β-H, R^3=H_2,
R^4=(=CH-Me); Methuenine
e R^1=OMe, R^2=β-CO_2Me, R^3=H_2,
R^4=(=CH-Me);
N_1-methoxy-19-20-dehydroervatamine
f R^1=H, R^2=β-CO_2Me, R^3=O,
R^4=(=CH-Me);
5-*oxo*-19-20-dehydroervatamine

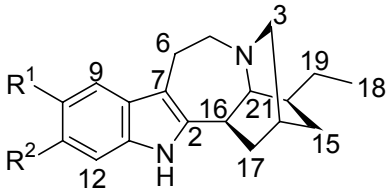
C-5



lahoricine

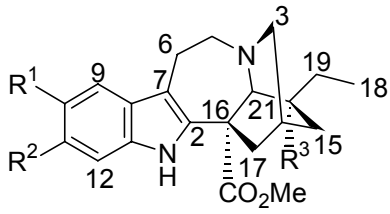
Ibogan type

I-1



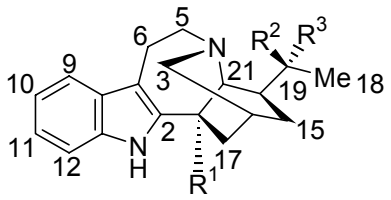
a R^1,R^2=H; ibogamine
b R^1=H,R^2=OCH_3; tabernanthine
c R^1=OCH_3,R^2=H; ibogaine
d R^1=OCH_3,R^2=OCH_3; ibogaline

I-2



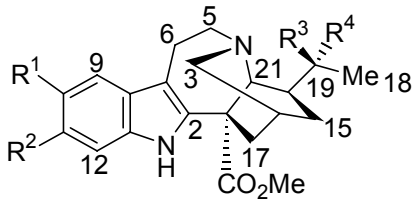
a R^1, R^2,R^3 =H; coronaridine
b R^1=OCH_3, R^2, R^3=H; voacangine
c R^1=H, R^2=OCH_3; isovoacangine
d R^1=OCH_3, R^2=OCH_3, R^3=OH;
20-hydroxy-conopharyngine

I-3



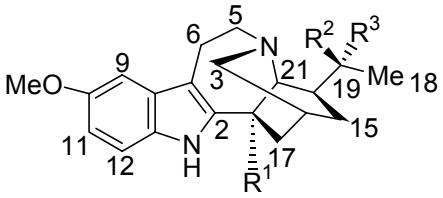
a R^1=H, R^2=OH, R^3=H; 19-(*S*)-hydroxyibogamine
b R^1=CO_2Me, R^2=OH, R^3=H; heyneanine
c R^1=CO_2Me,R^2=H, R^3=OH; 19-*epi*-heyneanine

I-4



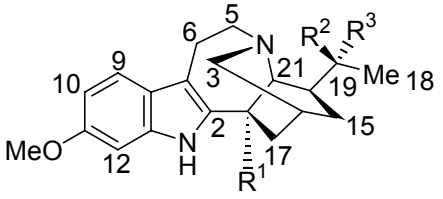
a R^1=OCH_3, R^2=H, R^3, R^4=O; voacryptine
b R^1=H, R^2=OCH_3, R^3, R^4=O; isovoacryptine
c R^1=OH, R^2=H, R^3=OH, R^4=H; 10-hydroxy-heyneanine

I-5



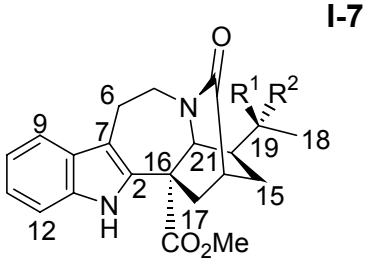
a R^1=CO_2Me, R^2=H, R^3=H; voacangine
b R^1=H, R^2=OH, R^3=H; iboxygaine
c R^1=CO_2Me, R^2=OH, R^3=H; voacristine
d R^1=CO_2Me, R^2=H, R^3=OH; 19-*epi*-voacristine

I-6



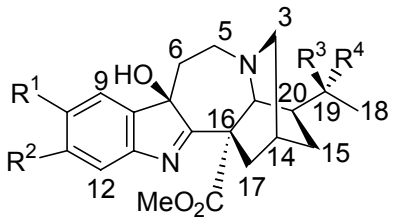
a R^1=CO_2Me, R^2=H, R^3=H; isovoacangine
b R^1=H, R^2=OH, R^3=H; tabernanthine
c R^1=CO_2Me, R^2=OH, R^3=H; isovoacristine

I-7



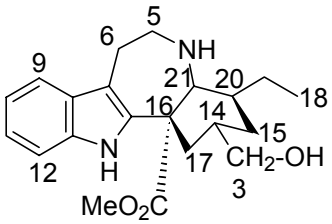
a R^1=H, R^2=H; 3-*oxo*-coronaridine
b R^1=OH, R^2=H; 3-*oxo*-heyneanine
c R^1=H, R^2=OH; 3-*oxo*-19-*epi*-heyneanine

I-8



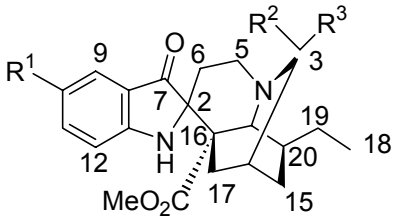
a $R^1=R^2=R^3=R^4=H$;
coronaridine hydroxyindolenine
b $R^1=OMe$, $R^2=R^3=R^4=H$;
voacangine hydroxyindolenine
c $R^1=R^2=H$, $R^3=OH$, $R^4=H$;
heyneanine hydroxyindolenine
d $R^1=OMe$, $R^2=H$, $R^3=OH$, $R^4=H$;
voacristine hydroxyindolenine

I-9



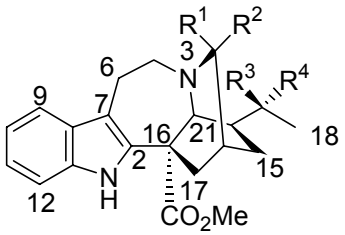
3-hydroxy-3,4-seco-coronaridine

I-10



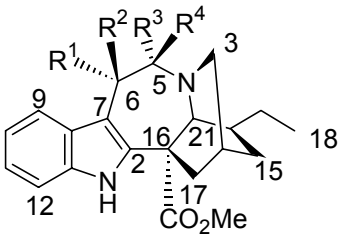
a $R^1= R^2= R^3=H$; coronaridine pseudoindoxyl
b $R^1=OMe$, $R^2=R^3=H$; voacangine pseudoindoxyl
c $R^1=H$, $R^2=CH_2COMe$, $R^3=H$; 3*R*-(2'-oxopropyl)-coronaridine pseudoindoxyl
d $R^1=H$, $R^2=H$, $R^3=CH_2COMe$; 3*S*-(2'-oxopropyl)-coronaridine pseudoindoxyl
e $R^1=OMe$, $R^2=CH_2COMe$, $R^3=H$; 3*R*-(2'-oxopropyl)-voacangine pseudoindoxyl
f $R^1=OMe$, $R^2=H$, $R^3=CH_2COMe$; 3*S*-(2'-oxopropyl)-voacangine pseudoindoxyl

I-11



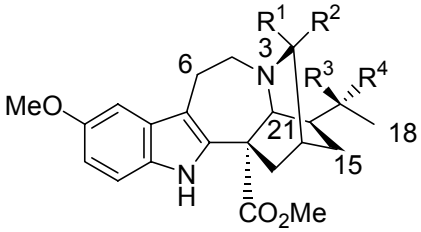
a R^1=H, R^2=OC_2H_5, R^3=H, R^4=H; 3*S*-ethoxy-coronaridine
b R^1=OC_2H_5, R^2=H, R^3=H, R^4=H; 3*R*-ethoxy-coronaridine
c R^1=H, R^2=OC_2H_5, R^3=OH, R^4=H; 3*S*-ethoxy-heyneanine
d R^1=OC_2H_5, R^2=H, R^3=OH, R^4=H; 3*R*-ethoxy-heyneanine
e R^1=H, R^2=OC_2H_5, R^3=H, R^4=OH; 3*S*-ethoxy-19-*epi*-heyneanine
f R^1=OC_2H_5, R^2=H, R^3=H, R^4=OH; 3*R*-ethoxy-19-*epi*-heyneanine
g R^1=H, R^2=CH_2COMe, R^3=H,R^4=H; 3*S*-(2'-oxopropyl)-coronaridine
h R^1=CH_2OMe, R^2=H, R^3=H, R^4=H; 3*R*-(2'-oxopropyl)-coronaridine
i R^1=H, R^2=$CHOHCH_3$, R^3=H, R^4=H; 3*S*-(β-hydroxyethyl)-coronaridine
j R^1=H, R^2=OH, R^3=H, R^4=H; 3*S*-hydroxy-coronaridine
k R^1=OH, R^2=H, R^3=H, R^4=H; 3*R*-hydroxy-coronaridine

I-12

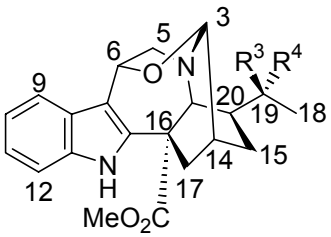


a R^1, R^2=H, R^3,R^4=O;
5-*oxo*-coronaridine
b R^1,R^2=O,R^3=OH, R^4=H;
5-hydroxy-6-*oxo*-coronaridine
c R^1,R^2=O, R^3,R^4=H
6-*oxo*-coronaridine
d R^1, R^2=H, R^3=CH_2COMe,R^4=H;
5*S*-oxopropyl-coronaridine
e R^1, R^2=H, R^3=H, R^4=CH_2COMe
5*R*-oxopropyl-coronaridine

I-13



a R^1=H, R^2=CH_2COMe, R^3=OH,
R^4=H; 3*S*-(2'-oxopropyl)-voacangine
b R^1=CH_2OMe, R^2=H, R^3=OH,
R^4=H; 3*R*-(2'-oxopropyl)-voacangine
c R^1, R^2=O, R^3=OH,
R^4=H; 3-*oxo*-voacangine

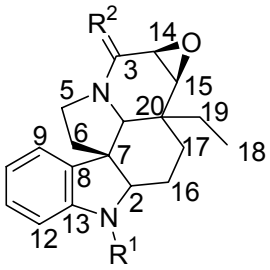


I-14

eglandine

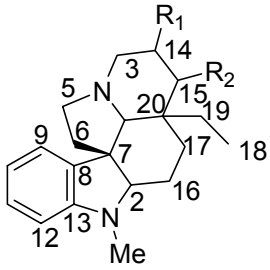
Plumeran type

P-1



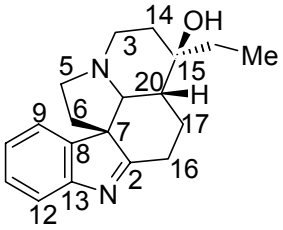
a R^1=Me, R^2=O; 3-*oxo*-mehranine
b R^1=Me, R^2=H_2; mehranine
c R^1=H, R^2=H_2; lochnericine

P-2



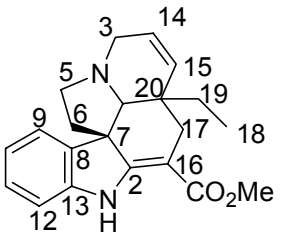
a R^1=αOH,H, R^2=βOH, H;
14α-15β-dihydroxy-*N*-methyl-aspidosperimidine
b R^1=H_2, R^2=H_2;
N_1-methyl-aspidosperimidine

P-3



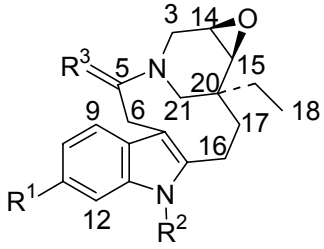
20*S*-hydroxy-1,2-dehydro-pseudoaspidospermidine

P-4



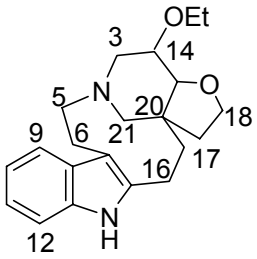
tabersonine

P-5



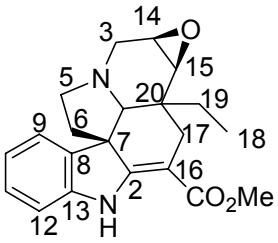
a R^1=H, R^2=H, R^3=H_2; voaphylline
b R^1=H, R^2=Me, R^3=H_2;
N(1)-methylvoaphylline
c R^1=OH, R^2=H, R^3=O; ervatinine
d R^1=H, R^2=H, R^3=OH,H; stapfinine

P-6



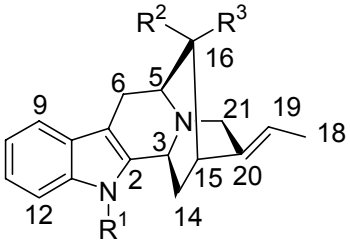
hyderabadine

P-7



pachysiphine

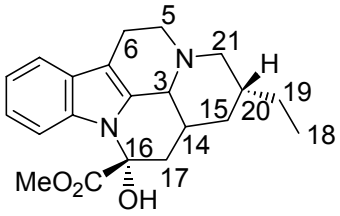
P-8



a R^1=H, R^2=CO_2Me, R^3=H;
pericyclivine
b R^1=Me, R^2=CH_2OH, R^3=CO_2Me;
voachalotine

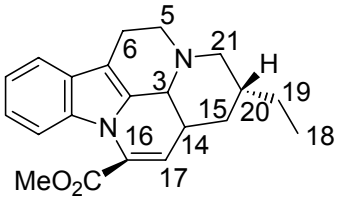
Tacaman Type

T-1



tacamine

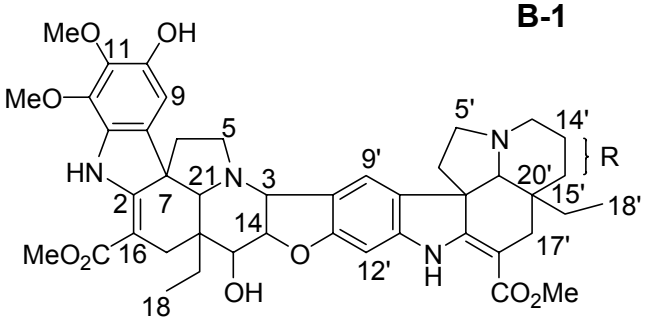
T-2



16,17-dehydro-tacamine

Bis-indole Type

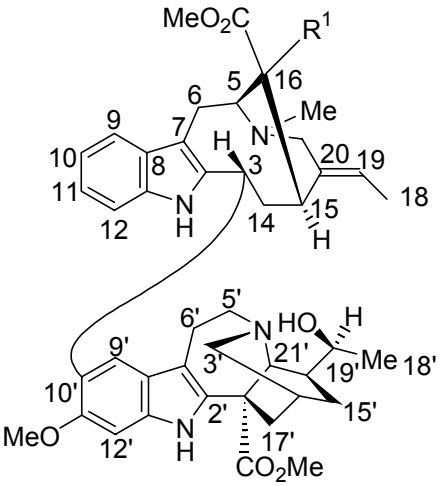
B-1



a R=  ;conophylline

b R=14'α-15'β,hydroxy; conophyllidine

B-2



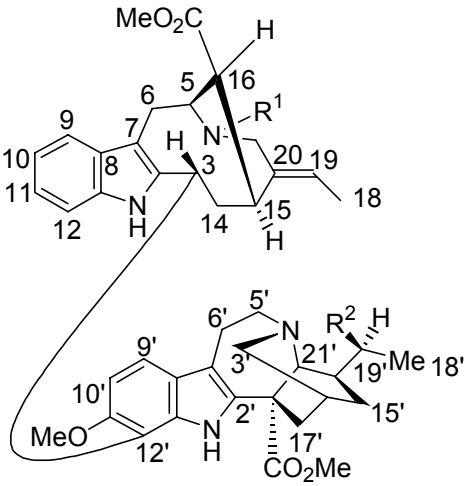
a R^1=H, R^2=Me, R^3=OH, R^4=H; 19'(*S*)-hydroxy-conoduramine

b R^1=CH_2OH, R^2=Me, R^3=OH, R^4=H; conodiparine A

c R^1=CH_2OH, R^2=Me, R^3, R^4=O; conodiparine C

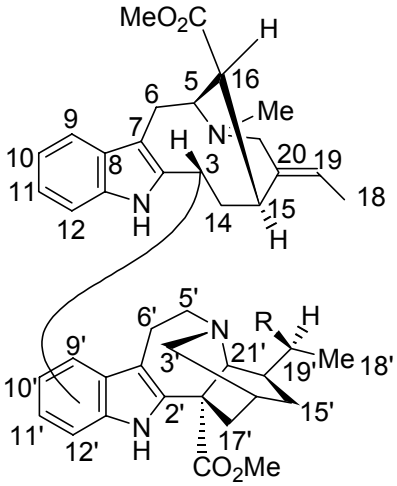
d R^1=CH_2OH, R^2=H, R^3=OH, R^4=H; ; conodiparine E

B-3



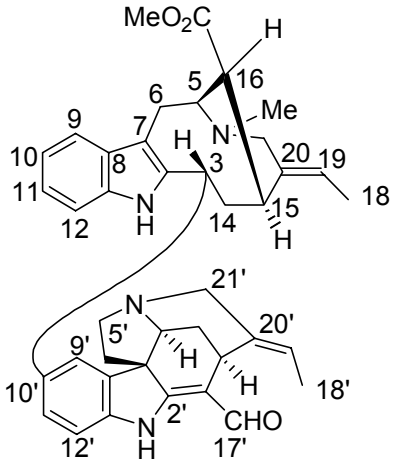
a R^1=Me, R^2=H;
conodurine
b R^1=Me, R^2=OH;
19'(*S*)-hydroxy-conodurine
c R^1=H, R^2=OH;
conodurinine

B-4



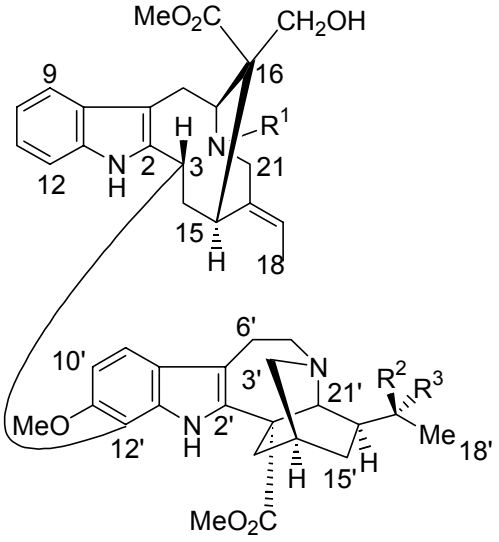
a C(3)-C(11'), R =H;
ervahanine A
b C(3)-C(11'), R =OH;
19'(*S*)-hydroxy-ervahanine A
c C(3)-C(10'), R =H;
ervahanine B
d C(3)-C(12'), R =H;
ervahanine C

B-5



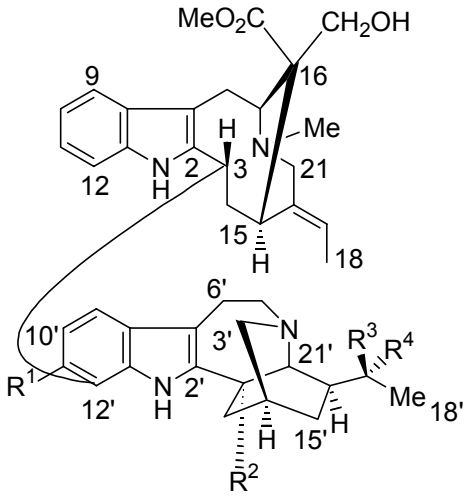
vobatricine

B-6



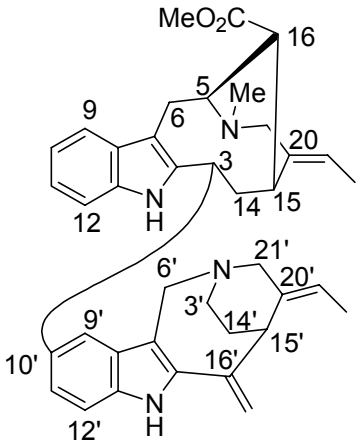
a R^1=Me, R^2 =OH, R^3=H; conodiparine B
b R^1=Me, R^2, R^3 =H; conodiparine D
c R^1=H, R^2 =OH, R^3=H; conodiparine F

B-7



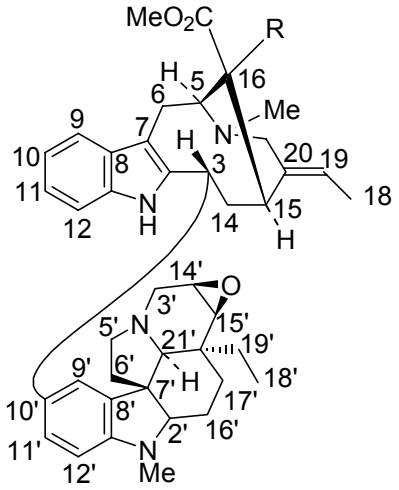
a R^1=OMe, R^2 =H, R^3 =OH, R^4 =H; conodutarine A
b R^1=OMe, R^2 =H, R^3, R^4 =O; conodutarine B
c R^1=OH, R^2 =CO_2Me, R^3 =OH, R^4 =H; cononitarine A
d R^1=OH, R^2 =CO_2Me, R^3 =H, R^4 =H; cononitarine B

B-8



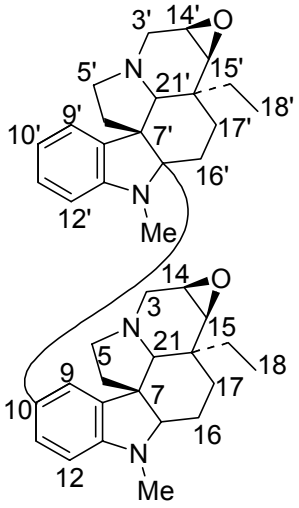
pseudovobparisine

B-9



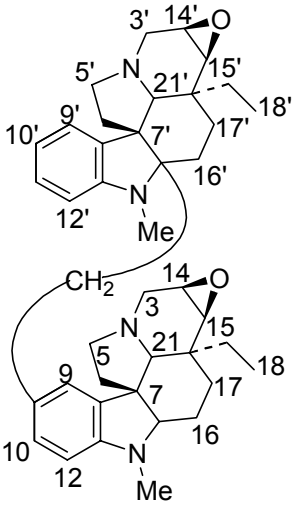
a R= H; tabernaemontabovine
b R=CH_2OH; tabernaemontavine

B-10



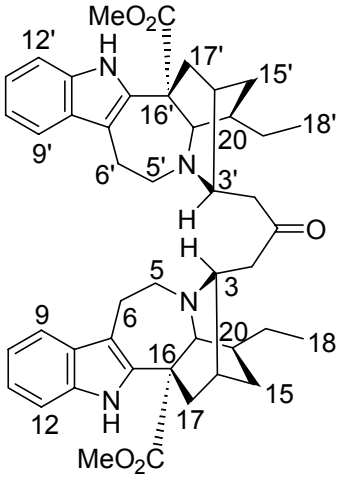
tabernaebovine

B-11



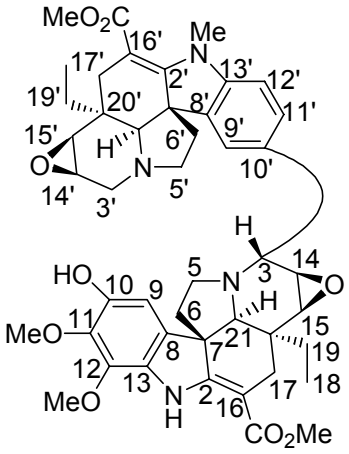
methylenebis-mehranine

B-12



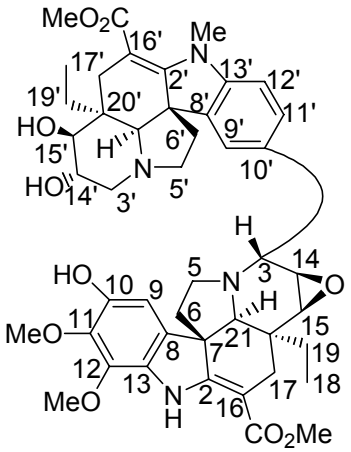
3,3'-oxopropyl-dicoronaridine

B-13



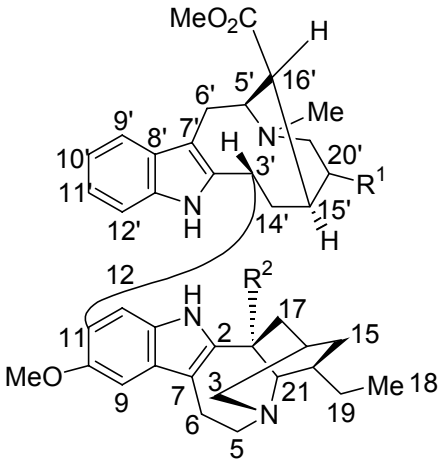
pedunculine

B-14



peduncularidine

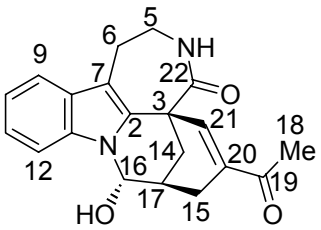
B-15



a R^1=(=CH-Me), R^2=CO_2Me; voacamine
b R^1=(=CH-Me), R^2=H; 16-decarmethoxy-voacamine
c R^1=α-C_2H_5, R^2=H; ervadivaricatine A
d R^1=β-C_2H_5, R^2=H; ervadivaricatine B

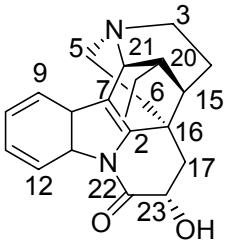
Miscellaneous type

M-1



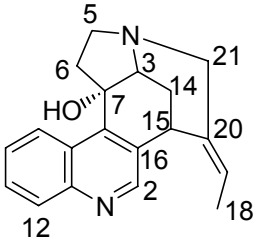
tronocarpine

M-2



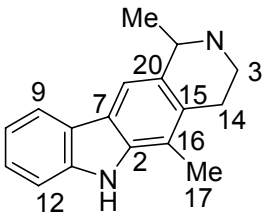
tronoharine

M-3



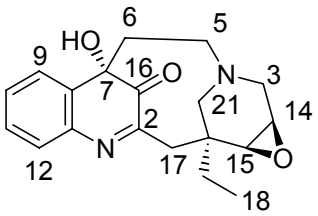
voastrictine

M-4



(3,14), (4,19)-tetrahydro
-olivacine

M-5



voaharine

1.3 The phytochemical study of *Tabernaemontana peduncularis* Wall.

Chemical study of the leaves and stem bark was performed by Zeches-Hanrot, *et al.* in 1995 and afforded two novel bis-indole alkaloids, pedunculine and peduncularidine, together with seven known alkaloids, coronaridine, coronaridine hydroxyindolenine, eglandine, heyneanine, eglandulosine, heyneanine hydroxyindolenine and N_1-methyl-aspidospermidine (Zeches-Hanrot *et al*., 1995).

1.4 The ethnopharmacology information of *Tabernaemontana peduncularis* Wall.

Tabernaemontana peduncularis Wall. roots are used against abscesses in the nose in Malaysia by decocting. A decoction is drunk in treating ulceration of the nose in tertiary syphilis. Moreover, other methods of administration are probably used. The plant is poisonous (van Beek *et al*., 1984; Perry, 1980). In Thai folklore medicine, the roots are used for the treatment of inflammation and fever (Taesotikul, 1997).

1.5 The biological activities of *Tabernaemontana* plants and their indole alkaloids

The studies in the pharmacological properties of *Tabernaemontana* plants were divided into two groups. The first group concerns tests for their chemotherapeutic activities e.g. antimicrobial, antitumor, antiprotozoal and antiviral. The second group includes general experiments on intact or isolated

organ systems e.g. central or peripheral nervous systems, cardiovascular system, smooth muscles and endocrines. These came from the most common use in folklore medicine concerned infectious diseases which chemotherapeutic investigations were dominating (Taesotikul, 1997; van Beek *et al.,* 1984). Many alkaloids from *Tabernaemontana* plants have been screened for antitumor activites under the auspices of the National Cancer Institute of the U.S. National Institute of Health. Additionally, in a screening of ethanolic extracts from 19 *Tabernaemontana* species, most of them showed a broad spectrum of antibacterial activities and some species also showed antiviral and antiamoebic activities (van Beek *et al.,* 1984).

The roots and stem barks of *Tabernaemontana* species accumulate monoterpenoid indole alkaloids, which display experimentally cytotoxic, anti-inflammatory, analgesic, hypotensive, anti-infectious and neurotropic properties (van Beek *et al.,* 1984). Conoduramine, conodurine, coronaridine, gabunine and vobasine were reported in a cytotoxic fraction of *T. holstii* (Kingston *et al*., 1977). Voacangine and voacamine isolated from the latex of *T. arborea* were cytotoxic against P-388 lymphotye leukemia cells (Kingston, 1978). Conodiparines A-D obtained from *T. corymbosa* showed appreciable activity in reversing resistance in vincristine-resistant KB cells (Kam *et al*., 1998). Other anti-inflammatory and analgesic effects were obtained with the administrations orally or intraperitonealy of an alcoholic and an aqueous extract of *T. divaricata* stem barks which contain coronaridine, heyneanine, voacamine, voacristine, voaphylline, vobasine,

tabersonine as well as the anti-inflammatory and analgesic salicylic acid. In addition an alcoholic extract of *T. divaricata* increased the pentobarbital induced sleeping time (Henriques *et al*., 1996) and intravenous administration of *T. divaricata* roots, stems, leaves and flower extracts caused sedation, lowered respiration and skeletal muscle tone in rodents. Most of the extracts showed analgesic properties with greater pharmacological activities from the roots and stems bark (Taesotikul *et al*., 1989a).

From *T. dichotoma,* stemmadenine, perivine, vobasine, coronaridine and dichomine exhibited hypotensive and muscle relaxant activities (Perera *et al*., 1985). Tubotaiwine and appricine from *T. pachysiphon* leaves exhibited analgesic activity via the opiate receptor (Ingkaninan *et al*., 1999).

Intravenous administration of an ethanol extract of *T. pandacaqui* stems, leaves and flowers induced hypotension in pentobarbital anesthetized rats. At high dose (100-300 mg/Kg), flower extract induced a transient hypertensive effect preceding hypotensive activity. On heart, chronotropic negative and inotropic negative responses were observed (Taesotikul *et al*., 1989b). Intravenous injection to rats of a crude alkaloidal fraction of *T. pandacaqui* induced 2 consecutive hypotensive and bradychardiac responses (Taesotikul *et al*., 1998a). Studies on carrageenin-induced rat paw edema, yeast-induced hyperthermia in rat and writhing response induced by acetic acid in mice showed that the alcoholic extract of stems of *T. pandacaqui* has significant anti-inflammatory, antipyretic and antinociceptive activities. These activities are due to alkaloidal components since

they were also observed when the crude alkaloidal fraction separated from alcoholic extract was tested in the same models (Taesotikul *et al*., 2003b)

Various extracts of *Tabernaemontana* species exhibited antibacterial, parasiticide and antiviral properties (van Beek *et al.,* 1984). *N*-demethylconodurine isolated from *T. van heurkii,* inhibited *Leishmania braziliensis* (10 mg/ml) whereas conodurine and conoduramine showed highest activities toward *Bacillus subtilis, Escherichia coli, Mycobacterium smegmatis, Pseudomonas aeruginosa* and *Staphylococcus aureus.* Using infected BALB/c mice, *N*-demethylconodurine was inactive whereas conoduridine was less active than *N*-methy1glucarnine antimonate, a drug of choice in the treatment of leishmaniasis (Munoz *et al*., 1994). Coronaridine showed potent antileishmanial activity, inhibiting promastigote and amastigote growth by pronounced alterations in their mitochondria (Delorenzi *et al*., 2001). Tabersonine and ibogamine inhibited the growth of *Bacillus subtilis* (Achenbach *et al*., 1997; van Beek *et al*., 1985)Whereas, voacangine inhibited the growth of *Mycobacterium tuberculosis* (Rastogi *et al*., 1998).

18-Methoxycoronaridine (18-MC), a novel iboga alkaloid congener that decreases drug self-administration in several animal models, may be a potential treatment for multiple forms of drug abuse. In animal models, 18-MC reduced intravenous morphine, cocaine, methamphetamine and nicotine self-administration, oral alcohol and nicotine intake, and attenuated signs of opioid withdrawal, but had no effect on responding for a nondrug reinforcer (water) and produced no apparent toxicity. Antagonists of $\alpha 3\beta 4$ nicotinic receptors may represent a totally novel

approach for treating multiple addictive disorders, and 18-MC might be the first of a new class of synthetic agents acting via this novel mechanism and having a broad spectrum of activity (Maisonneuve and Glick, 2003).

1.6 The objectives in this investigation

The previous phytochemical study of *Tabernaemontana peduncularis* was performed on the leaves and stem bark (Zeches-Hanrot *et al.,* 1995). While, the phytochemical study on the roots that have been used in folklore medicine has not yet been reported. These prompted the author to investigate the indole alkaloids from the roots of this plant in the hope of obtaining additional information which may lead to the better understanding of the occurrence and distribution of the alkaloids in this plant and this genus. Consequently, the main objectives in this investigation are as follows:

1. To isolate and purifiy indole alkaloidal compounds from the roots of *Tabernaemontana peduncularis*.
2. To determine the chemical structure and physical properties of each isolated alkaloid.