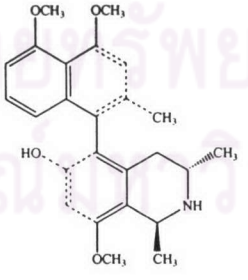
CHAPTER I

INTRODUCTION

Plants from the families Ancistrocladaceae and Dioncophyllaceae have yielded a number of napthylisoquinoline alkaloids that structurally differ in the position of biaryl linkage, the degree of oxidation in the isoquinoline ring and the degree of *O*-methylation. Despite the growing number of such alkaloids, simple isoquinoline compounds devoid of the naphthalene unit had not been reported from either family until recently.

Ancistrocladus, the only genus of the plant family in Ancistrocladaceae, is composed of nearly 20 species distributed in tropical Asia, Malasia, and West Africa (Gereau, 1997). The alkaloid found can be divided into two groups as 5-naphthyl-1,2,3,4-tetrahydroquinoline derivatives and 7-naphthyl-1,2,3,4-tetrahydroquinoline derivatives. The 5-naphthyl isoquinoline alkaloids, one of the very few novel structural types discovered in the past one or two decades, ancistrocladine (1) is the most predominant representative of this growing group which more than 20 related natural products have been reported (Bringmann, 1984). The structural features common to all of these fascinating alkaloids, especially the hindered naphthalene isoquinoline linkage gives rise to the highly stable atropisomers, constitute a synthetic challenge.



Alkaloids from Ancistrocladaceae (1)

Several 5-naphthyl-1,2,3,4-tetrahydroisoquinoline alkaloids isolated from Ancistrocladaceae such as ancistrocladine (1a), hamatine (1b), ancistrobrevine B (2) and ancistrobertsonine A-C (3-5), ancistrolikokines A-C (6-8) and ancistrocongolines A-C (9-11), etc are known as moderately to highly active antimalarial agents against

Plasmodium falciparum, the pathogen of malaria which cause more than one million deaths per year (Bringmann *et al*, 1999). The IC_{50} value of the 5-naphthyl-1,2,3,4-tetrahydroquinoline derivatives are summarized in table 1 (Bringmann *et al*, 1999-2000; Bringmann, Messer, Brun and Mudogo, 2002).

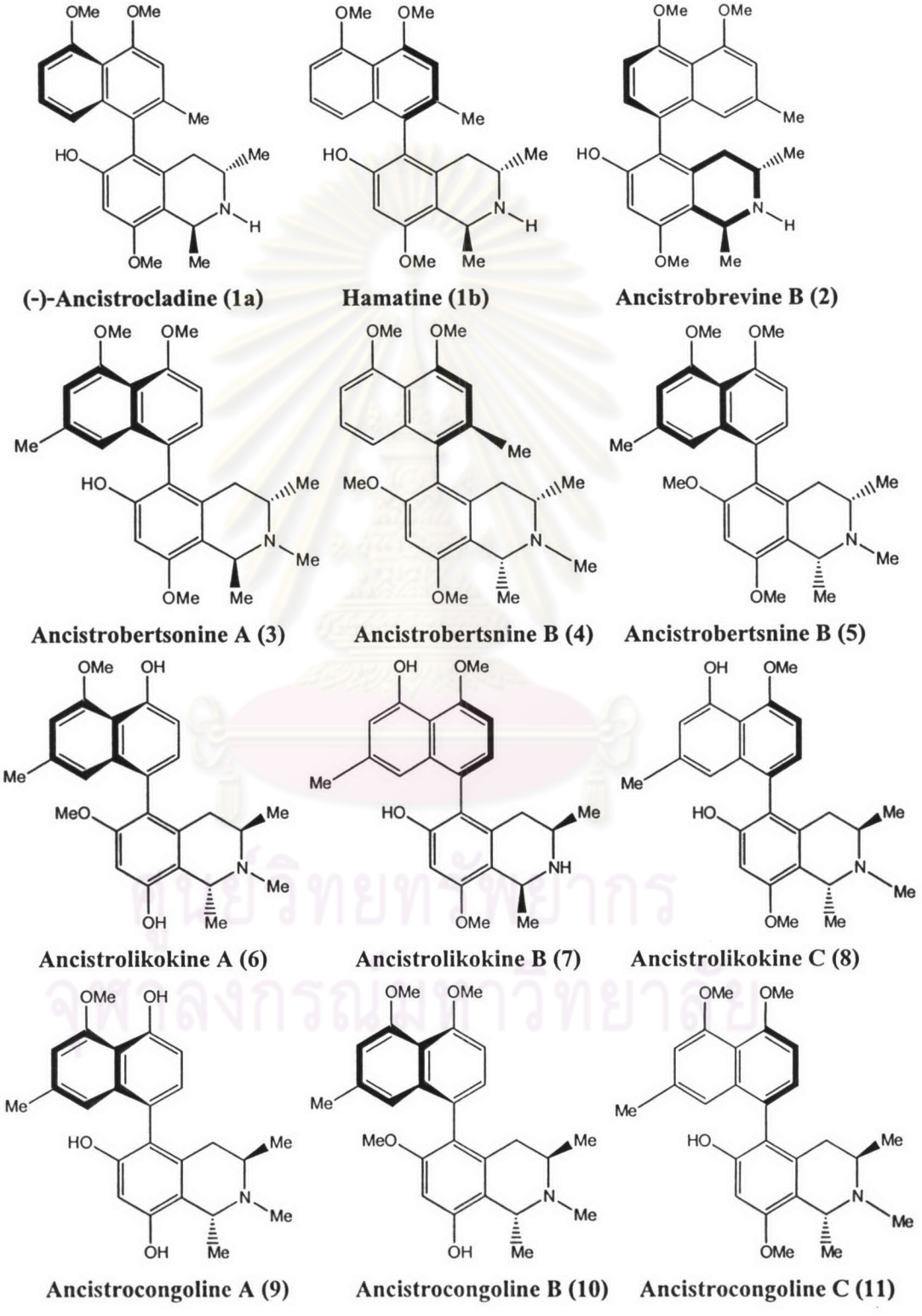


Figure 1 5-Naphthyl-1,2,3,4-tetrahydroisoquinoline alkaloids of *Ancistrocladaceae sp.*

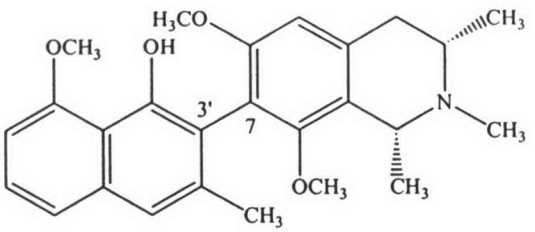
Table 1 IC_{50} values of the 5-Naphthyl-1,2,3,4-tetrahydroisoquinoline alkaloids (isolated from *Ancistrocladaceae sp.*) on chloroquine resistant (K1) and susceptible (NF5) strains of *Plasmodium falciparum.*

Compounds	*K1-strain IC_{50} [μM]	**NF54-strain IC_{50} [μM]
Ancistrobrevine B	**2.00**	**4.70**
Ancirtrobertsonine A	**15.90**	**> 23.70**
Ancirtrobertsonine B	**9.00**	**> 23.00**
Ancirtrobertsonine C	**4.50**	**10.10**
Ancistrolikokine A	**0.34**	**0.47**
Ancistrolikokine B	**0.53**	**1.37**
Ancistrolikokine C	**2.27**	**15.29**
Korupensamine	**0.18**	**0.063**

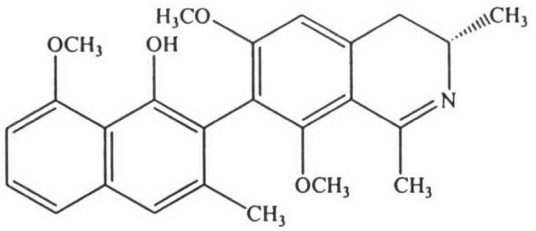
* *P. falciparum*, K1-strain (chloroquine resistant)

** *P. falciparum,*NF 54-strain (chloroquine susceptible)

A new member of the naphthalene-isoquinoline series of alkaloids, ancistrotectorine, was isolated from the leaves of the Thai medicinal plant *Ancistrocladus tectorius* (Ancistrocladaceae) (Ruangrungsi *et al.*, 1985). This plant is known in Thai as "Khon-ma-daeng". Its leaves have been used in a folk medicine in Thailand for the treatment of edema and dermatitis (Na Songkla, 1882). Its roots have been used for the treatment of malaria and dysentery in Burma and Malaysia (Burkill, 1935, vol.1). In 1985, Ruangrungsi and co-workers isolated two compounds of 7,3′-linkage type of naphthylisoquinoline alkaloids from the leaves known as ancistrotectorine (12) and ancistrocladidine (13) from *A. tectorius* and *A. beyneanus*, respectively.



Ancistrotectorine (12)



Ancistrocladidine (13)

The pharmacological activity of ancistrotectorine, which is antispasmodic action, has been studied. This activity was tested on the intestinal contraction induced by acetylcholine, histamine, serotonin, barium chloride, and potassium chloride. Moreover, it also shows the contraction inhibitory induced by oxytocin and serotonin in the rat uterus and guinea-pig ileum (Pasupat, 1985). This compound also exhibits this action on the smooth muscle of blood vessel and aorta induced by potassium chloride, calcium chloride, norepinephrine, serotonin and histamine (Phusiraphan, 1987). The above data showed that ancistrotectorine had a direct effect on smooth muscle and caused a non-specific relaxation (Boonprasphai *et al.*, 1990).

Several biological activities can be found in the 1,2,3,4-tetrahydro-isoquinoline (THIQ) derivatives such as smooth muscle relaxant, antimalarial, anticonvulsant, antiviral and cytotoxic activities (Hoye *et al.*, 1999; Rosaria *et al.*, 2000). Various biological activities of 1,2,3,4 THIQ derivatives were illustrated in Figure 2.

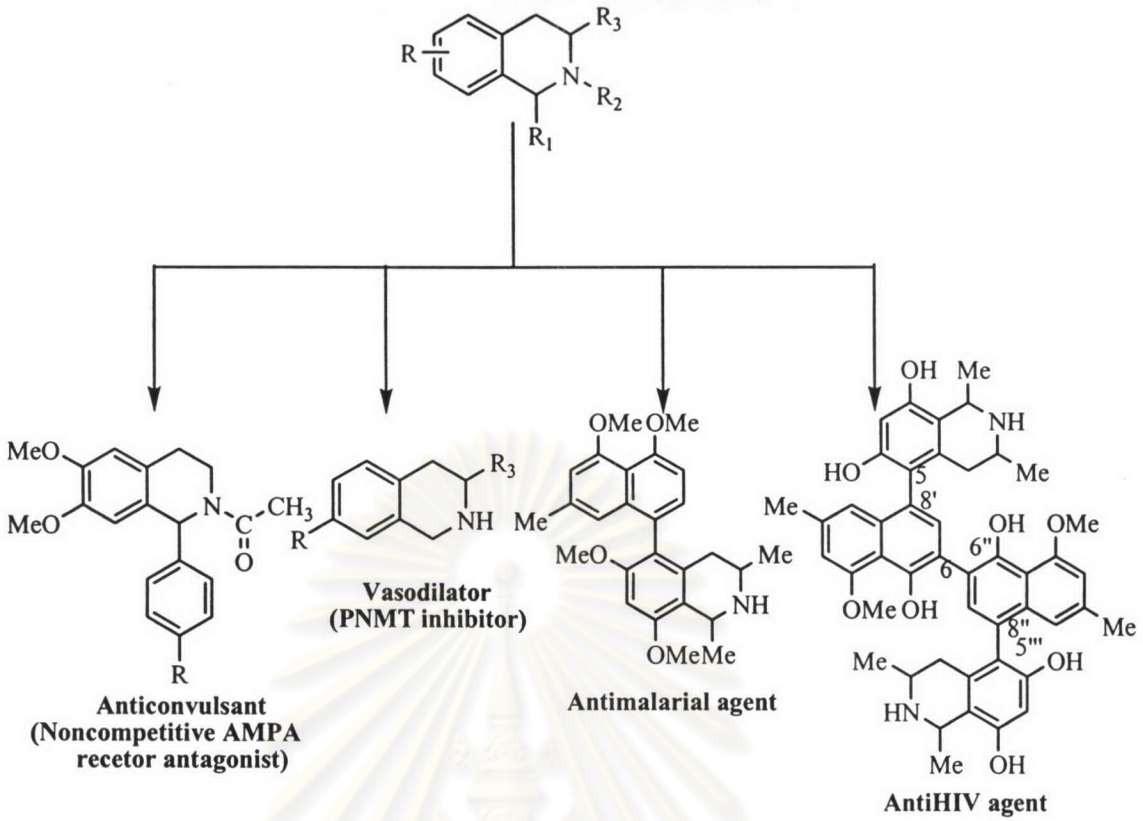
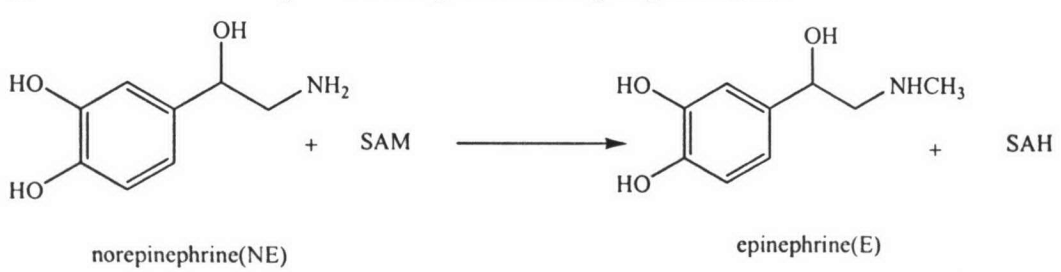


Figure 2 Diagram illustrated the versatile pharmacological activities of the 1,2,3,4 –THIQ derivatives

As of the previous reports, the 1,2,3,4–terahydroisoquinoline nucleus has been used for structural modification to improve the versatile biological activities as well as antimalarial and smooth muscle relaxation activity. Biomimeticly, the 1,2,3,4–tetrahydroisoquinoline derivative are considered as rigid analogues of β-phenyl-ethylamine structure which is an ideal pharmacophore of adrenergic drug (Delgado and Remeis, 1998) and also exhibits phenylethanolamine *N*-methyltransferase (PNMT) inhibition activity. PNMT was referred to norepinephrine *N*-methyltransferase (NMT) and catalyzed the terminal step in the biosynthesis of epinephrine (E).



PNMT is located mainly in the adrenal medulla, but small quantities is also found in the brain of mammals (Saavedra *et al.*, 1974), where it is localized in areas concerned with the control vital functions such as secretion of pituitary hormone (Crowley and Terry, 1981), heart rate and blood pressure. Reduction in the level of central epinephrine by PNMT inhibitors was initially believed to be the cause of its antihypertensive effect (Goldstein *et al.*, 1982; Terry *et al.*, 1981). Later, it was found that most of the widely-studied PNMT inhibitors were non-selective and had α_2-adrenoceptor binding affinity (Stolk *et al.*, 1984; Ruffolo *et al.*, 1984). Several means for inhibiting epinephrine biosynthesis seem possible and most obviously such an action might be produced by inhibitors of the enzyme that are chemically related to the natural substrate, e.g. S-adenosylmethionine (SAM), Norepinephine (NE), S-adenosylhomocysteine (SAH), and other compounds which related to SAM inhibition.

However, the majority PNMT inhibitors are structurally related to NE. Cyclization of phenylethylamine into a fused piperidine ring gives tetrahydroisoquinoline (THIQ) compound that greatly enhances PNMT-inhibitory potency and also exhibits strong affinity for the α_2-adrenoceptor (Grunewal, 1999). Thus, 1,2,3,4-tetrahydroisoquinoline compound has been modified to improve the PNMT-inhibitor activity based on the idea of β-phenylethylamine modification. The structure of β-phenylethylamine consists of a benzene ring and an ethylamine side chain. The substitution can be modified at: 1) the terminal amino group, 2) the alpha- or beta-positions of the ethylamine side chain and 3) the substitution on the aromatic ring (Delgado and Remeis, 1998), as illustrated in Figure 3.

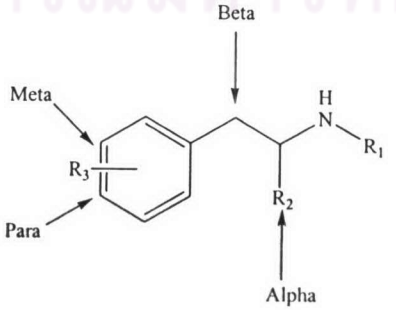


Figure 3 Site of structural modification of phenylethylamine.

R^1, Substitution on Amino Nitrogen

When R^1 is increased in size from hydrogen in norepinephrine to methyl in epinephrine to isopropyl in isoproterenol, the activity at α-receptors decreases, and activity at β-receptors increases. These compounds were used to define alpha and beta activity long before receptor proteins could be isolated and characterized. The activity at both α and β-receptors is maximal when R^1 is methyl as in epinephrine, but α-agonist activity is dramatically decreased when R^1 is larger than methyl and is negligible when R^1 is isopropyl as in isoproterenol, leaving only β activity. Presumably, the β-receptor has a large lipophilic binding pocket adjacent to the amine-binding aspartic acid residue, which is absent in the α-receptor. As R^1 becomes larger than butyl, affinity for α_1-receptors returns, but not intrinsic activity, which means large lipophilic can afford compounds with α_1-blocking activity. In addition, the *N*-substituent can also provide selectivity for different β-receptors, with a t-butyl group affording selective for β_2-receptors.

R^2, Substitution α to the Basic Nitrogen, Carbon-2

Small alkyl groups, methyl or ethyl, may be present on the carbon adjacent to the amino nitrogen. Such substitution slows metabolism by monoamine oxidase (MAO) but has little overall effect on duration of action in catecholamines because they remain substrates for catechol-*O*-methyltransferase (COMT). While at the *beta*-position, particularly the hydroxyl substitution may be important for storage of sympatomimetic amine in neural vesicle.

R^3, Substitution on the aromatic ring

The aromatic ring are substituted at *meta*- and *para*-position (catechol moiety) are maximal agonist activity at adrenergic receptors and the excellent substrate for catechol-*O*-methyltransferase (COMT). For example, the natural 3′,4′-dihydroxy substituted ring present in norepinephrine provides excellent receptor activity for both α and β-sites, but such catechol-containing compounds have poor oral activity because they are hydrophilic

and rapidly metabolized by COMT. But the absence of one or the other of these groups, particularly the hydroxyl at *meta*-position, without other substitutions on the ring may be dramatically reduced the potency of the compounds. Thus, it can be replaced catechol moiety with resorcinol structure gave the drug metaproterenol. Since the resorcinol ring is not the substrate for COMT enzyme.

The two principle enzymes involved in adrenergic drug metabolism are monoamine oxidase (MAO) and catechol-*O*-methyltransferase (COMT). Both of these enzymes are distributed throughout the body, with high concentration found in the liver and kidney. MAO is associated primarily with the outer membrane of the mitochondria, which COMT is found primarily in the cytoplasm. Neither MAO nor COMT exhibits a high degree of substrate specificity.

MAO oxidatively deaminate a variety of compound that contains an amino group attached to a terminal carbon. The catecholamine metabolism indicate that in the adrenergic neuron of human brain and peripheral tissue NE is oxidatively deaminated by MAO to give 3,4-dihydroxyphenyl glycolaldehyde (Figure 4).

Methylation by COMT occurs almost exclusively on the *meta*-hydroxyl group of catechol. For example, the action of COMT upon NE and E give normetanephrine and metanephrine (Figure 4). In fact, regardless of whether the first metabolism step is oxidation by MAO or methylation by COMT.

When the phenyl ring has no phenolic substituents, i.e., R^3 = H, these phenylethanolamines may have both direct and indirect activity. Direct activity (i.e. agonist) is the simulation of an adrenoceptor by the drug itself while indirect activity is the result of displacement of norepinephrine from its storage granules or reuptake inhibitor resulting in non-selective stimulation of the adrenoceptor by the displaced norepinephrine.

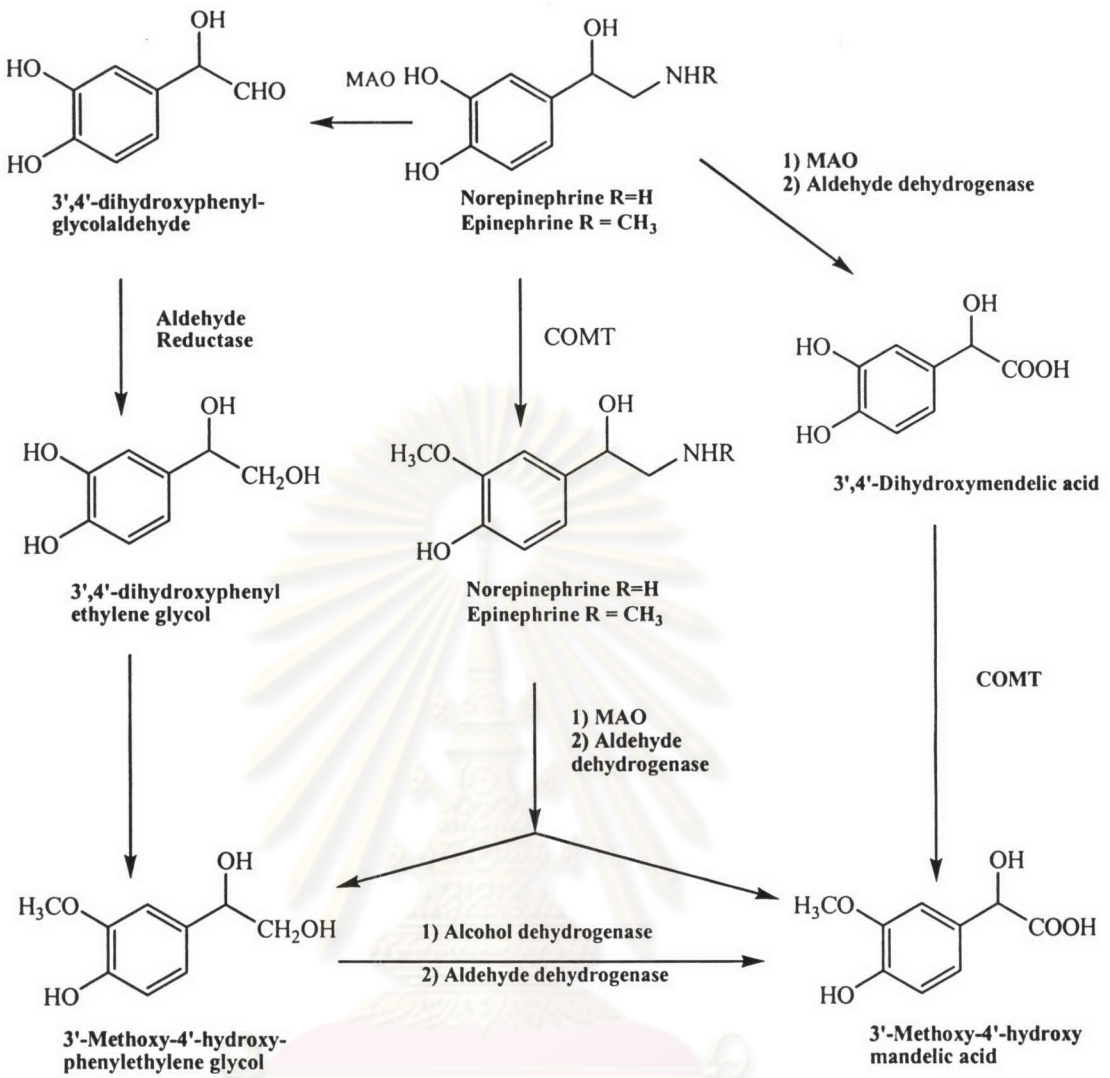


Figure 4 The metabolism of norepinephrine and epinephrine by MAO and COMT

Moreover, the 5-naphthyl-1,2,3,4-tetrahydroisoquinoline derivatives have reported as the potent antimalarial activity which were capable of curing most forms of malaria including chloroquine-resistant strains of *P. falciparum* and should be considered the traditional substances of choice for these resistant strains (Bringmann *et al.*, 1999)

Assumption to design the target compounds possessing an antimalarial activity

Malaria is an acute infectious disease caused by four species of the protozoal genus plasmodium. The parasite is transmitted to humankind through the bite of female

anopheles mosquito, which thrives in humid, swampy areas. *Plasmodium falciparum* is the most dangerous species, causing an acute, rapidly fulminating disease characterized by persistent high fever, orthostatic hypotension and massive erythrocytosis(swollen and reddish condition of the limbs). *Plasmodium falciparum* infection can lead to capillary obstruction and death if treatment is not instituted promptly. *Plasmodium vivax* causes a milder form of the disease. *P. malariae* is common to many tropical region but *Plasmodium ovale* is rarely encountered. The antimalarial drugs can be divided the various categories following the mode of action and the chemical structures but they have also some toxic effects such as hemolytic anemia, abdominal discomfort, visual disturbances, electrocardiographic abnormalities (prolonged QT interval, cardiac arrest), hallucination and depression, etc. The side-effects lead to the failure of treatment and the life-threatening in the therapeutic course. Resistance acquired by the mosquito to insecticides, and by the parasite to drugs is the important problem, has led to new therapeutic challenges, particularly in the treatment of *P. falciparum*.

To maintain the biological activity of antimalaria, the target structures should be the 5-aryl-1,2,3,4-tetrahydroisoquinoline derivatives to be analogue of the 5-naphthyl-1,2,3,4-tetrahydroisoquinoline derivatives which were separated from the plants in the family Ancistrocladaceae (Figure 5). The synthetic approach is simplification of these derivatives which can eradicate the atropisomer event in the synthetic process. Previous preliminary *in vitro* test, it showed the antispasmodic activity in the 1-substituted -1, 2, 3, 4-THIQ derivatives. In contrast, non-substituted derivatives have not been found in this activity. Thus, the substituent in the 1-position may be responsible the antispasmodic activity. The 1-substiuted-5-aryl-1,2,3,4-THIQ derivatives may be show the antimalarial activity but it also contain the constipation side-effect.

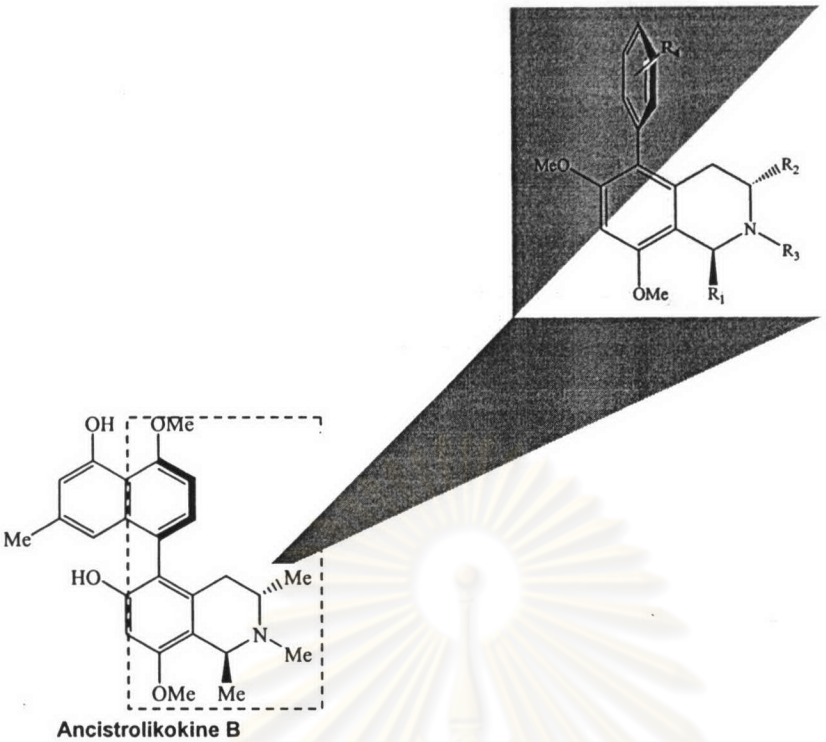
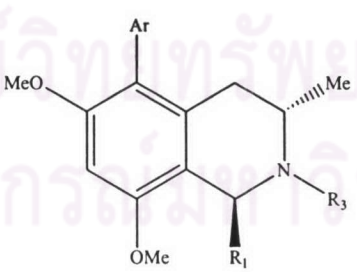


Figure 5 Simplification of the 5-naphthyl-1,2,3,4-tetrahydroisoquinoline structure to 5-aryl-3-methyl-1,2,3,4-tetrahydroisoquinoline derivatives to evaluate the antimalarial activity.

All above results support the appropriate parent structure in this study is 5-Aryl-3-methyl-1,2,3,4-tetrahydroisoquinoline derivatives. The modification in the parent structure shall be made in the C-1 position and C-5 position to maintain the potent antimalarial activity and to lessen smooth muscle relaxant toxicity. (Figure 6)



R_1 = H , $COOBu^n$

R_3 = Benzyl

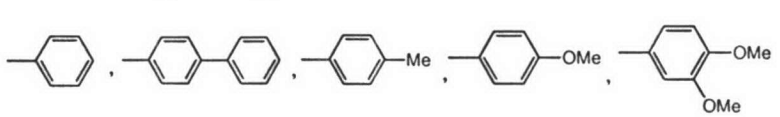
Ar = 

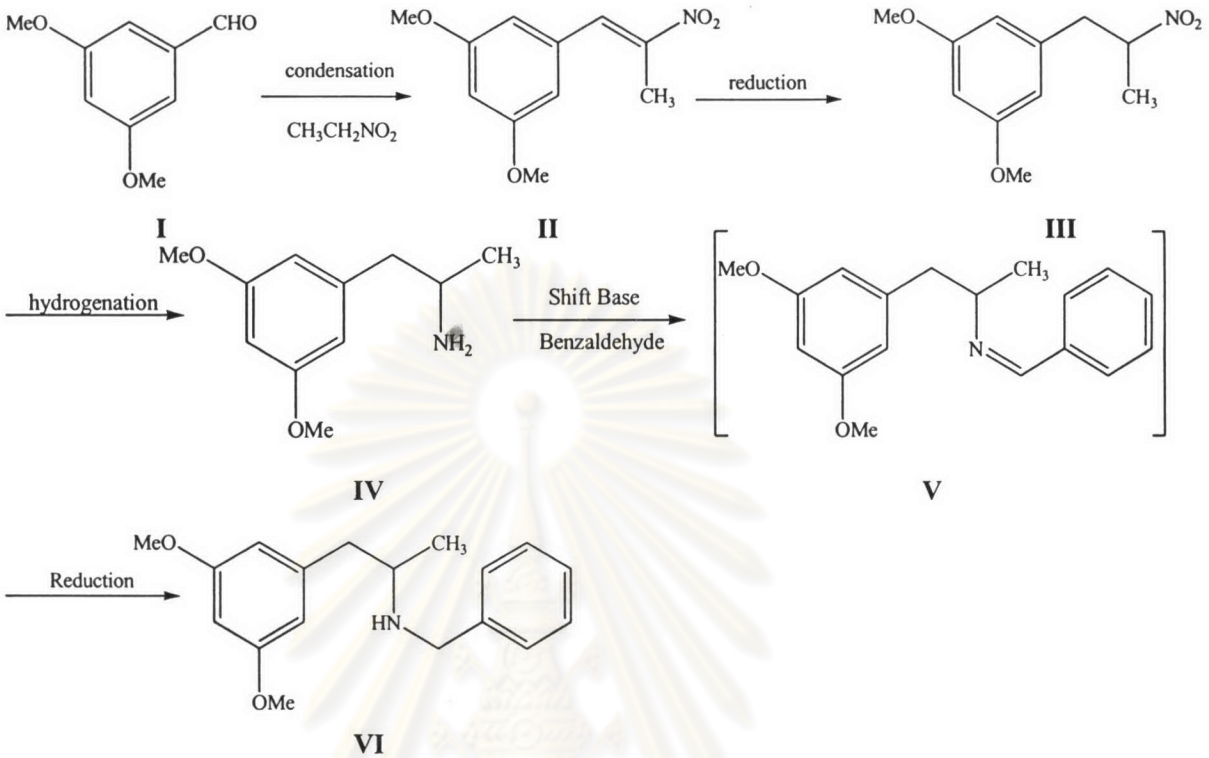
Figure 6 The chemical structures of target compounds

The synthetic approach for the research

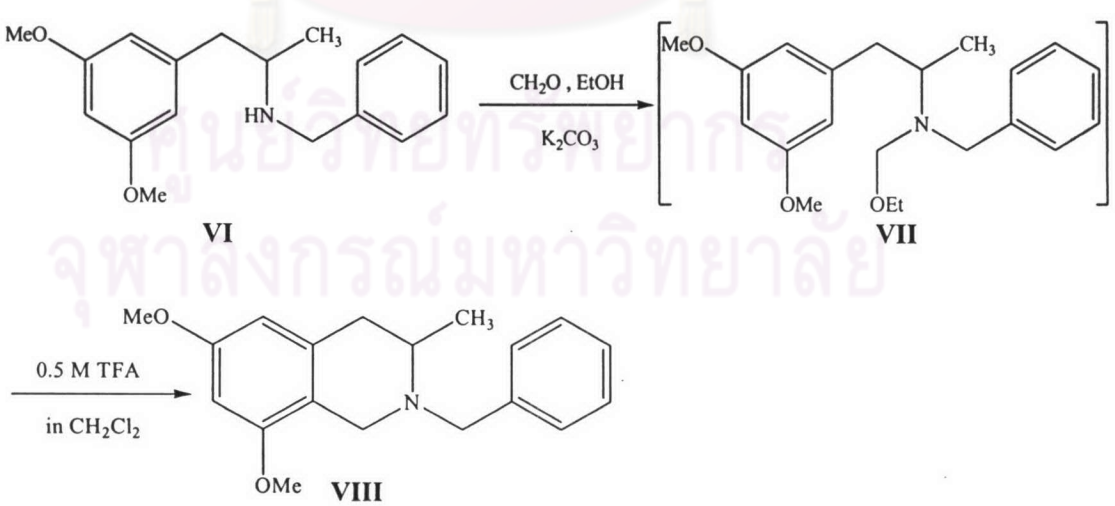
The 5-aryl-3-methyl-1,2,3,4-tetrahydroisoquinoline derivatives are synthesized by the several steps. In the study, the synthetic approach is divided into five major steps as follows.

1. Procedure of the *N*–benzylphenylethylamine synthesis (Scheme 1)
2. Procedure of the 1,2,3,4-tetrahydroisoquinoline cyclization through *N,O*-acetal intermediate using paraformaldehyde (Scheme 2)
3. Procedure of the 1,2,3,4-tetrahydroisoquinoline cyclization through *N,O*-acetal intermediate using butyl glyoxal (Scheme 3)
4. The synthetic procedures for the 5-iodo-6,8-dimethoxy-3-methyl-1,2,3,4-tetrahydroisoquinoline derivatives using the aromatic iodination (Scheme 4)
5. To synthesize the 5-aryl-3-methyl-1,2,3,4-tetrahydroisoquinoline derivatives via Suzuki Coupling reaction (Scheme 5)

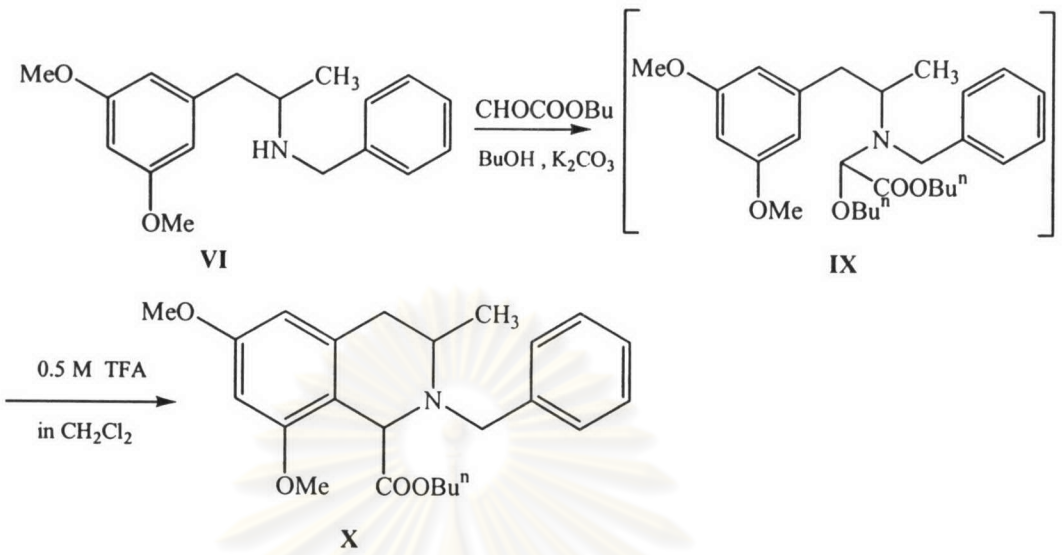
Scheme 1: The synthetic procedures of *N*-benzyl-2-(3′,5′-dimethoxyphenyl)-1-methylethylamine



Scheme 2: The synthetic procedures for 6,8-dimethoxy-3-methyl-1,2,3,4-tetrahydro-isoquinoline through *O,N*-acetal cyclization using paraformaldehyde



Scheme 3: The synthetic procedures for 6,8-dimethoxy-3-methyl-1,2,3,4-tetrahyroisoquinoline through *O,N*-acetal cyclization using butyl glyoxal



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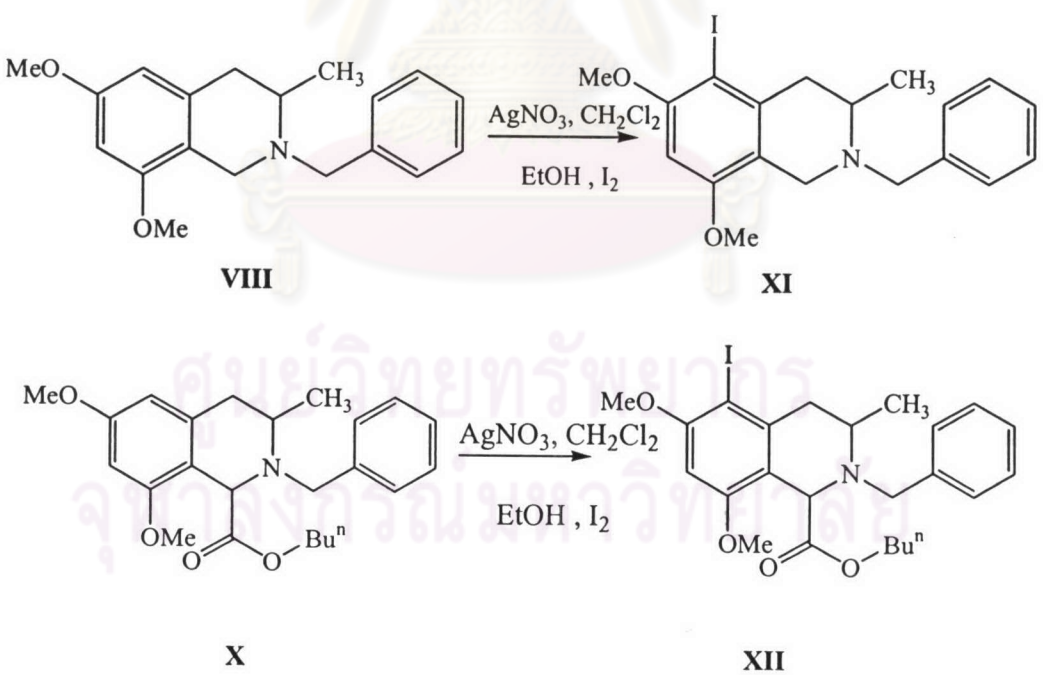
Scheme 4: The synthetic procedure for the 5-iodo-6,8-dimethoxy-3-methyl-1,2,3,4-tetrahydroisoquinoline derivatives using the aromatic iodination



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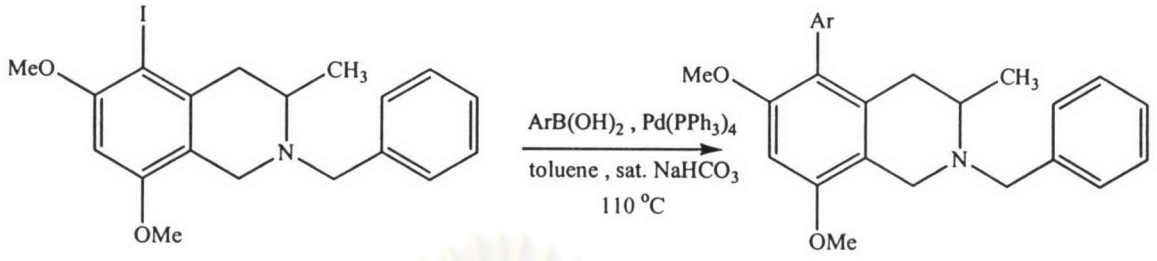
Scheme 5: The corresponding 5-aryl-3-methyl-1,2,3,4-tetrahydroisoquinoline derivatives synthesis via Suzuki Coupling reaction.



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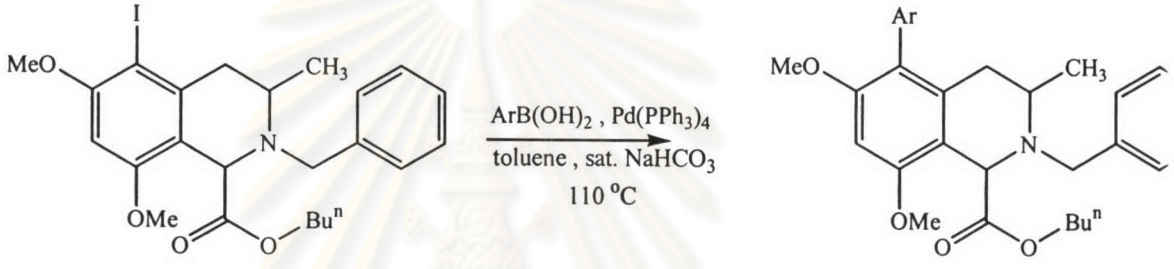

XII **XIV**