

Acetylcholinesterase Inhibitory and Antioxidant Properties of Thai Vegetables

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Abstract—Objective: To screen *in vitro* AChE inhibition and DPPH radical scavenging activity of Thai vegetables.

Methods: Nineteen Thai vegetables were investigated for AChE inhibitory activity based on Ellman's colorimetric method and the antioxidant activity was also determined by 2,2-diphenyl-1-picrylhydrazyl (DPPH) assay. **Results:** The methanol extracted of pods of *Vigna unguiculata* (L.) Walp. exhibited the strongest activity against AChE with percent inhibition of 53.05 ± 1.88 . For antioxidant activity of Nineteen Thai vegetables, the leaves extract of *Spondias pinnata* (L.f.) Kurz. demonstrated the most potent antioxidant activity with an EC_{50} value of $1.89 \mu\text{g/mL}$ which was comparable to the positive control, L-ascorbic acid ($EC_{50} = 1.62 \mu\text{g/mL}$). four extracts, include leaves of *Ipomoea aquatica* Forsk., arial parts of *Mentha cordifolia* Opiz., leaves of *Ocimum basilicum* L. and whole plants of *Piper sarmentosum* Roxb. extracts showed good activity with EC_{50} as 4.49, 8.22, 9.19 and 10.18 $\mu\text{g/mL}$, respectively. **Conclusion:** These results will be useful for further research and development of dietary supplements from *Vigna unguiculata* (L.) Walp. and *Spondias pinnata* (L.f.) Kurz. with biological properties against Alzheimer's disease and provide a useful guidance for further studies to identify the active compounds responsible for these biological activities.

Index Terms—Alzheimer's disease, acetylcholinesterase, antioxidant activity, Thai vegetables

I. INTRODUCTION

Alzheimer's disease (AD) is the most common cause of dementia among the elderly people and accounts for more than 80% of dementia cases worldwide [1]. AD is an irreversible and chronic neurodegenerative disorder which is characterized by a cognitive dysfunction and memory loss [2]. The pathologies of AD are the progressive accumulation of the beta amyloid plaques

outside neurons in the brain and abnormal forms of the protein tau inside neurons. These changes are eventually accompanied by the damage and death of neurons [3]. Moreover, the pathophysiology of AD is related to oxidative stress and inflammation, failure of synaptic function and a depletion of neurotransmitter acetylcholine (ACh) by acetylcholinesterases (AChE) [4]. AChE is an enzyme participating in cholinergic neurotransmission. It breaks down ACh which terminates the neurotransmission process [5]. A common approach for treating AD is to enhance the ACh level in the brain using AChE inhibitors (AChEIs) [6]. There are only four AChEIs currently approved by the U.S. Food and Drug Administration (FDA). Three of these drugs are commonly prescribed, i.e., donepezil, rivastigmine and galantamine whereas tacrine, the first cholinesterase inhibitor, is rarely prescribed today because of associated side effects like hepatotoxicity [7]. However, these all drugs are known to have limitations for clinical use due to their unfavorable side-effects, short-half-lives and problems associated with bioavailability [1], [8]. Therefore, the search for finding potential new AChEIs from natural sources that have strong activity to inhibit AChE without side effects is still great interest. Numerous chemical constituents from medicinal plants have been reported for their effects on AChEIs. The active compounds are huperzine-A from *Huperzaserrata*, galantamine from *Galanthus nivalis* and *Narcissus* sp., α -viniferin from *Caragana chamlague*, ursolic acid from *Origanum majorana* including coronaridine, voacangine and rupicoline from *Tabernaemontana australis* [8], [9].

Since the development and progression of AD is known as a result from oxidative stress that damages the central nervous system [10]. It plays harmful physiological responses which may lead to develop cell damage and various diseases such as diabetes, atherosclerosis, inflammation and carcinogenesis [11].

The use of natural antioxidants play an essential role in the prevention of diseases caused by oxidative stress due to the antioxidants can scavenge free radicals and inhibit lipid peroxidation [12]. Therefore, one of the AD therapeutics is especially related to phytotherapy through the use of natural antioxidants to minimize oxidative imbalance [10]. It is accept that vegetables are important elements of a healthy diet due to they contain high protective substances such as fiber, minerals and antioxidant vitamins [13]. The consumption of vegetables could help prevent chronic diseases such as cardiovascular diseases, hypertension, type 2 diabetes mellitus, dementia and several types of cancers [14]. In Thailand, there are several vegetables that are important for human nutrition and health. Thai vegetables are great interest because they are rich sources of beneficial phytochemicals with pharmacological properties. Some native vegetables have been found to have bioactive compounds with antioxidant activity [15].

Since, the varying side effects of the synthetic AChEIs, the search for novel active compounds with safe therapeutic as alternative drugs in treating AD are very attractive. Therefore, this study aims to determine the AChE inhibitory and antioxidant properties of some vegetables in the Southern region of Thailand to provide a basic guideline for further develop better AChEIs as

well as provide a base for development of dietary supplement for patients suffering from AD.

II. MATERIALS AND METHODS

A. Chemicals

All the chemicals used in these experiments were of analytical grade. AChE enzyme, acetylthiocholine iodide (ATCI), 5,5-dithiobis-2-nitrobenzoic acid (DTNB), bovine serum albumin (BSA), galanthamine, 2,2-diphenyl-1-picrylhydrazyl (DPPH) and L-ascorbic acid were purchased from Sigma-Aldrich (Thailand). All other chemicals were purchased from Merck (Darmstadt, Germany).

B. Plant Materials

Nineteen vegetable materials were purchased from local markets in Songkhla province, the South of Thailand in April and May 2016. The voucher specimens are deposited at Faculty of Pharmaceutical Sciences, Prince of Songkla University. All materials were identified by a botanist of the Department of Pharmacognosy and Pharmaceutical Botany, Faculty of Pharmaceutical Sciences, Prince of Songkla University. Some vegetables are presented in Fig. 1



Some vegetables used for study the AChE inhibitory and antioxidant properties.

C. Extraction

The vegetable materials were cleaned, cut into small pieces and dried in a hot air oven at 55 °C. The dried materials were ground into fine powder. An amount of 100 g of each powdered samples was macerated with 600 mL of 95% methanol for 3 days at room temperature. The methanol extracts were filtered through Whatman No. 4 filter paper and concentrated under reduced pressure using a rotary vacuum evaporator until dryness. The

residue was macerated again in 95% methanol for 7 days and filtered. The filtrate was concentrated with the same process and combined with the crude extract from the first extraction and stored at 4°C for further analysis.

D. Enzyme and Samples Preparation

AChE used in the assay was isolated from electric eel (type VI-S lyophilized powder, 480 U/mg solid, 530 U/mg protein). The lyophilized enzyme was prepared in

the 50 mM Tris-HCl buffer, pH 8.0 to obtain 1130 U/mL stock solution. The enzyme stock solution was kept at 80 °C. The further enzyme dilution was dissolved in 0.1% BSA in buffer. The concentration of each extract was adjusted to 1 mg/mL stock solution.

E. Microplate Assay for AChE Inhibitory Activity

Inhibition of AChE activity was evaluated *in vitro* by the spectrophotometric Ellman's method [16] with minor modifications as detailed by Inganinant *et al.* [17] using acetyl-thiocholine as substrate. The assay was performed at a final concentration of 100 mg/mL. In brief, 125 µL of 3 mM DTNB, 25 µL of 15 mM ATCI, 50 µL of buffer and 25 µL of sample extract dissolved in buffer were added to the wells of 96-well microplate followed by 25 µL of 0.28 U/mL AChE. The hydrolysis of acetylthiocholine was determined by monitoring the formation of yellow 5-thio-2-nitrobenzoate anion produced from the reaction of DTNB with thiocholine released by the enzymatic hydrolysis of acetylthiocholine [1] at a wavelength of 405 nm on a microplate reader (Bio-Tek Instrument, USA) every 10 s for 2 min. In this method, galantamine was used as a positive control. The percentage of AChE inhibitory activity was calculated according to the formula (1).

$$\text{Inhibition (\%)} = [(V_{\text{control}} - V_{\text{sample}}) / V_{\text{control}}] \times 100 \quad (1)$$

where V_{control} is the mean velocity of control (containing all reactants, except the sample extracts) and V_{sample} is the mean velocity of the sample extracts. All treatments were performed in triplicate.

F. Antioxidant Activity

The antioxidant capacity of the extracts was measured on the basis of the scavenging activity of the stable DPPH free radical according to the method described by Yamasaki *et al.* [18] with some modifications. This method is an antioxidant assay based on electron-transfer that produces a violet solution in ethanol. DPPH free radical is stable at room temperature and reduced in the presence of an antioxidant molecule giving rise to colorless ethanol solution [19]. An amount of 100 µL of sample solution at various concentrations (1-200 µg/mL) was mixed with 100 µL of 0.1 mM DPPH solution in absolute ethanol, in a 96-well plate. The reaction mixture was vortexed and incubated at room temperature for 30 min. After that, absorbance of the mixture was detected using a microplate reader at 517 nm. The control solution was prepared by mixing absolute ethanol and DPPH solution. Antioxidant efficacy of the sample extracts was compared with those of L- ascorbic acid, a positive control. The experiment was carried out in triplicate for each sample. The percentage of antioxidant activity (%AA) was calculated according to the formula (2).

$$\%AA = [(A_{\text{control}} - A_{\text{sample}}) / A_{\text{control}}] \times 100 \quad (2)$$

where A_{control} and A_{sample} are the absorbance values of the control and the sample extracts, respectively. The results were expressed as EC_{50} value (concentration of the sample extract producing 50% scavenging of DPPH radicals) which was obtained by plotting the %AA versus

sample concentrations.

III. RESULTS AND DISCUSSION

A. AChE Inhibitory Activity

The nineteen vegetable extracts belonging to fourteen families were screened for their anti-AChE *in vitro* by the spectrophotometric method. AChE inhibitory activity was classified as potent (>50% inhibition), moderate (30-50% inhibition), inactive or low (<30% inhibition) suggested by Vinutha *et al.* [20]. The percent inhibition data of the extracts are presented in Table I. The result showed that at the concentration of 100 µg/mL, only the pods of *Vigna unguiculata* (L.) Walp., known as yard long bean in Leguminosae family exhibited potent effect against AChE with the percent inhibition of $53.05 \pm 1.88\%$. Moreover, five extracts of *Momordica charantia* L., *Ipomoea aquatica* Forsk., *Mentha cordifolia* Opiz., *Spondias pinnata* (L.f.) Kurz. and *Piper sarmentosum* Roxb. showed moderate activity of AChE inhibitor (%) at 45.63 ± 1.83 , 39.73 ± 2.73 , 37.00 ± 3.98 , 36.04 ± 1.88 and 31.42 ± 2.36 , respectively.

Previous studies reported, many species in Leguminosae family exhibited AChE inhibitory activity such as leaves ethanol extract of *Aeschynomene indica*, *Cassia occidentalis*, *Crotalaria juncea*, *Sesbania sesban* and *Caesalpinia pulcherrima* including stem barks ethanol extract of *Amburana cearensis*, roots methanol extract of *Caragana chamlagu* and seeds ethanol extract of *Glycine max* [21].

The aqueous and ethanol seeds extracts of *Vigna unguiculata* (L.) Walp. were reported as a good antibacterial activity [22] and its constituents composed of tannins and phenolic acids such as *p*-hydroxybenzoic acid, protocatechuic acid, 2,4-dimethoxybenzoic acid and cinnamic acid derivatives [23]. Therefore, the strong AChE inhibitory of our study may be due to the rich of phenolic acid contents. In addition, most of the AChE inhibitory compounds are known to contain heterocyclic nitrogen (e.g. alkaloidal) which play an important role, the high activity of *Vigna unguiculata* may be also due to the rich alkaloidal contents [24]. An interesting result in present study, the fruits extract of *Momordica charantia* L. or bitter melon in Cucurbitaceae family possessed moderate AChE inhibitory activity with percent inhibition of 45.63 ± 1.83 . This plant is both a nutritious and healthy food. These two vegetables (*Vigna unguiculata* (L.) Walp. and *Momordica charantia* L.) are interesting to further study of active compounds for AD.

B. Antioxidant Activity

The capacity of vegetable extracts on antioxidant property was evaluated through DPPH radical scavenging activity. The EC_{50} values of all extracts indicating antioxidant property are presented in Table I. The wild mango, *Spondias pinnata* (L.f.) Kurz., in Anacardiaceae family exhibited the most potent antioxidant capacity with an EC_{50} value of 1.89 µg/mL. Interestingly, the activity of this extract was comparable to that of the positive control, L-ascorbic acid ($EC_{50} = 1.62$ µg/mL).

Moreover, the morning glory, *Ipomoea aquatica* Forsk. also showed high activity with an EC₅₀ value of 4.49 µg/mL. Among four vegetables belonging to the Labiatae family, *Mentha cordifolia* Opiz. and *Ocimum basilicum* L. possessed good antioxidant activity with EC₅₀ values of 8.22 and 9.19 µg/mL, respectively. Furthermore, *Piper sarmentosum* Roxb. in Piperaceae family also showed good activity with EC₅₀ values 10.78 µg/mL. Our result for methanol extract of *Piper sarmentosum* Roxb. was in agreement with a previous study reported that this plant showed good activity with 51.47% DPPH scavenging [25]. A variety of antioxidant compounds, naturally occurring in a number of dietary sources have been identified as free radical or active oxygen scavengers.

Several Thai indigenous vegetables have previously been shown to exhibit excellent sources of antioxidant compounds. They contained high amounts of phenolic compounds, e.g., gallic acid, caffeic acid, catechins, anthocyanidins, quercetin and rutin [26]. These compounds found in dietary and medicinal plants that could inhibit oxidative stress by antioxidant mechanism which are known to be a powerful chain breaking antioxidant, radical scavengers, metal chelators, reducing agents, hydrogen donors, and singlet oxygen quenchers [4]. In the present study, the potent antioxidant activity of vegetable extracts was possibly due to the hydroxyl and superoxide radical scavenging of phenolic compounds.

TABLE I. IN VITRO AChE INHIBITION AND DPPH RADICAL SCAVENGING ACTIVITY OF THAI VEGETABLES

NO.	Scientific name	Family	Common name	Local name	Part used	AChE inhibition (%)	DPPH radical inhibition (EC ₅₀ , µg/mL)
1	<i>Centella asiatica</i> (L.) Urb.	Apiaceae	Gotu kola	Bua bok	Leaf	27.49 ± 4.48	39.77
2	<i>Citrus hystrix</i> DC.	Rutaceae	Leech lime	Makrut	Leaf	23.23 ± 0.14	23.46
3	<i>Coccinia grandis</i> (L.) Voigt.	Cucurbitaceae	Ivy gourd	Tamlueng	Leaf	29.47 ± 2.06	26.05
4	<i>Diplazium esculentum</i> (Retz.) Sw.	Athyriaceae	Vegetable fern	Phak kood	Leaf	25.38 ± 4.51	83.29
5	<i>Garcinia cowa</i> Roxb.	Guttiferae	Cowa	Chamuang	Leaf	11.45 ± 1.94	46.80
6	<i>Gnetum gnemon</i> L. var. <i>tenerum</i> Markgr.	Gentaceae	Karen vegetables	Phak miang	Leaf	22.29 ± 3.71	101.62
7	<i>Ipomoea aquatica</i> Forsk.	Convolvulaceae	Morning glori	Phak bung	Leaf	39.73 ± 2.73	4.49
8	<i>Lasia spinosa</i> (L.) Thwaites.	Araceae	-	Phak nam	Arial part	0.98 ± 4.20	76.21
9	<i>Limnocharis flava</i> Buch.	Limnocharitaceae	Yellow velvet leaf	Phak pai	Whole plant	16.60 ± 1.47	50.96
10	<i>Mentha cordifolia</i> Opiz.	Labiatae	Kitchen mint	Sa ra nae	Arial part	37.00 ± 3.98	8.22
11	<i>Momordica charantia</i> L.	Cucurbitaceae	Bitter gourd	Mara khinok	fruit	45.63 ± 1.83	17.67
12	<i>Ocimum africanum</i> Lour.	Labiatae	Hairy basil	Manglak	Arial part	23.02 ± 0.87	42.81
13	<i>Ocimum basilicum</i> L.	Labiatae	Sweet basil	Horapha	Leaf	9.67 ± 4.22	9.19
14	<i>Ocimum gratissimum</i> L.	Labiatae	Shrubby basil	Yee rah	Arial part	0.32 ± 5.13	44.21
15	<i>Oenonthe stolonifera</i> Wall.	Apiaceae	Water dropwort	Phak chee lom	Arial part	24.97 ± 2.47	40.27
16	<i>Piper sarmentosum</i> Roxb.	Piperaceae	Wildbetal leafbush	Cha phlu	Whole plant	31.42 ± 2.36	10.78
17	<i>Sauropus androgynus</i> (L.) Merr.	Euphorbiaceae	Star gooseberry	Phak wan ban	Leaf	10.15 ± 3.35	32.56
18	<i>Spondias pinnata</i> (L.f.) Kurz.	Anacardiaceae	Wild mango	Makok	Leaf	36.04 ± 1.88	1.89
19	<i>Vigna unguiculata</i> (L.) Walp.	Leguminosae	Yard long bean	Tua fuk yao	Pod	53.05 ± 1.88	73.20
	Galantamine			95.91 ± 2.46	-	95.91 ± 2.46	-
	Ascorbic acid			-	1.62	-	1.62

IV. CONCLUSION

Nineteen vegetable species belonging to fourteen plant species were investigated for AChE inhibitory and antioxidant properties. The most potent activity against AChE was the pods extract of *Vigna unguiculata* (L.) Walp. This plant should be considered for further studies to identify the active compounds responsible for the

AChE inhibitory activity. The assessment of antioxidant capacity, The leaves extract of *Spondias pinnata* (L.f.) Kurz. exhibited prominent activity. Several vegetables were the dietary sources of the potent antioxidant capability. The consumption of vegetables provides potential nutraceuticals for human health and may help in preventing or alleviating patients suffering from AD through AChE inhibition and scavenge free radicals.

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CONFLICTS OF INTERESTS

All authors have none to declare.

REFERENCES

[1] M. Jung and M. Park, "Acetylcholinesterase inhibition by flavonoids from *Agrimoniapilosa*," *Molecules*, vol. 12, pp. 2130-2139, 2007.

[2] M. H. Oh, P. J. Houghton, W. K. Whang, and J. H. Cho, "Screening of Korean herbal medicines used to improve cognitive function for anti-cholinesterase activity," *Phytomedicine*, vol. 11, pp. 544-548, 2004.

[3] A. Association, "Alzheimer's disease facts and figures," *Alzheimers & Dementia*, vol. 12, pp. 1-80, 2016.

[4] E. A. Adewusi and V. Steenkamp, "In vitro screening for acetylcholinesterase inhibition and antioxidant activity of medicinal plants from southern africa," *Asian Pac. J. Trop. Med.*, vol. 4, no. 10, pp. 829-835, 2011.

[5] M. Pohanka, M. Hrabnova, K. Kuca, and J. P. Simonato, "Assessment of acetylcholinesterase activity using indoxylacetate and comparison with the standard Ellman's method," *Int. J. Mol. Sci.*, vol. 12, no. 4, pp. 2631-2640, 2011.

[6] C. Sabphon, T. Sermboonpaisarn, and P. Sawasdee, "Cholinesterase inhibitory activities of xanthenes from *Anaxagorea luzonensis* A. Gray," *Journal of Medicinal Plant Research*, vol. 6, no. 4, pp. 3781-3785, 2012.

[7] J. C. Morris, "About FDA-approved cholinesterase inhibitors," *Cholinesterase Inhibitors Fact Sheet*, pp. 1-2, 2003.

[8] P. K. Mukherjee, V. Kumar, M. Mal, and P. J. Houghton, "Acetylcholinesterase inhibitors from plants," *Phytomedicine*, vol. 14, pp. 289-300, 2007.

[9] M. T. Andrade, *et al.*, "Indole alkaloids from *Tabernaemontana australis* (Muell. Arg) Miens that inhibit acetylcholinesterase enzyme," *Bioorg. Med. Chem.*, vol. 13, no. 12, pp. 4092-4095, 2005.

[10] B. C. M. Omena, *et al.*, "Antioxidant, anti-acetylcholinesterase and cytotoxic activities of ethanol extracts of peel, pulp and seeds of exotic Brazilian fruits: Antioxidant, anti-acetylcholinesterase and cytotoxic activities in fruits," *Food Res. Int.*, vol. 49, pp. 334-344, 2012.

[11] J. Worarathphoka, K. Intarapichet, and K. Indrapichate, "Antioxidant activity and cytotoxicity of six selected, regional, Thai vegetables," *AEJTS*, vol. 4, no. 2, pp. 108-117, 2012.

[12] S. Natarajan, K. P. Shanmugiahthevar, and P. D. Kasi, "Cholinesterase inhibitors from sargassum and gracilaria gracilis: Seaweeds inhabiting south Indian coastal areas," *Nat. Prod. Res.*, vol. 23, no. 4, pp.355-369, 2009.

[13] C. H. G. Van, *et al.*, "Consumption of vegetables and fruits and risk of breast cancer," *JAMA*, vol. 293, no. 2, pp. 183-193, 2005.

[14] A. Agudo, "Measuring intake of fruit and vegetables," *World Health Organization*, p. 40, 2005.

[15] N. Andarwulan, R. Batari, D. A. Sandrasari, B. Bolling, and H. Wijaya, "Flavonoid content and antioxidant activity of vegetables from Indonesia," *Food Chem.*, vol. 121, no. 6, pp. 1231-1235, 2010.

[16] G. L. Ellman, K. D. Courtney, V. Andres, and R. M. Featherstone, "A new and rapid colorimetric determination of acetylcholinesterase activity," *Biochem. Pharmacol.*, vol. 7, no. 2, pp. 88-95, 1961.

[17] K. Ingkaninan, P. Temkitthawon, K. Chuenchom, T. Yuyaem, and W. Thongnoi, "Screening for acetylcholinesterase inhibitory activity in plants used in Thai traditional rejuvenating and neurotonic remedies," *J. Ethnopharmacol.*, vol. 89, no. 2-3, pp. 261-264, 2003.

[18] K. Yamasaki, A. Hashimoto, Y. Kokusenya, T. Miyamoto, and T. Sato, "Electrochemical method for estimating the antioxidative

effects of methanol extracts of crude drugs," *Chem. Pharm. Bull.*, vol. 42, no. 8, pp. 1663-1665, 1994.

[19] D. J. Huang, B. X. Ou, and R. L. Prior, "The chemistry behind antioxidant capacity assays," *J. Agric. Food. Chem.*, vol. 53, no. 6, pp. 1841-1856, 2005.

[20] B. Vinutha, *et al.*, "Screening of selected Indian medicinal plants for acetylcholinesterase inhibitory activity," *J. Ethnopharmacol.*, vol. 109, no. 2, pp. 359-363, 2007.

[21] J. M. B. Filho, *et al.*, "Natural products inhibitors of the enzyme acetylcholinesterase," *Revista Brasileira De Farmacognosia*, vol. 16, no. 2, pp. 258-285, 2006.

[22] D. Sandeep, "Evaluation of antibacterial activity of seed extracts of *Vigna unguiculata*," *Int. J. Pharm. Pharm. Sci.*, vol. 6, no. 2, pp. 75-77, 2014.

[23] P. Siddhuraju and K. Becker, "The antioxidant and free radical scavenging activities of processed cowpea (*Vigna unguiculata* (L.) Walp.) seed extracts," *Food Chem.*, vol. 101, pp. 10-19, 2007.

[24] J. Nino, J. A. Hernandez, Y. M. Correa, and O. M. Mosquera, "In vitro inhibition of acetylcholinesterase by crude plant extracts from Colombian flora," *Mem. Inst. Oswaldo. Cruz.*, vol. 101, no. 7, pp. 783-785, 2006.

[25] P. Buachum, R. Wongsiri, and O. In-noi, "Study on antioxidant activity of crude extracts from some local and general vegetables," presented at the North Eastern Science and Technology Conference, Thailand, 2014.

[26] P. Maisuthisakul, "Phenolic constituents and antioxidant properties of some Thai plants," in *Phytochemicals-A Global Perspective of Their Role in Nutrition and Health*, V. Rao, Ed., Thailand, 2012, pp. 187-212.



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S. Langyanai and J. Puripattanavong, "Study on antioxidant activity of Thai medicinal plants in Songkhla Rajabhat University," in *Proc. 4th CDD Int Conf.*, Phuket, Thailand, 2016, pp. 151-152.

S. Langyanai and J. Puripattanavong, "Acetylcholinesterase inhibitory activity from *Bauhinia integrifolia* Roxb. extract," in *Proc. 3rd CDD Int Conf.*, Krabi, Thailand, 2014, pp. 192-193.



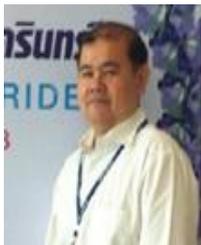
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P. Chaniad, C. Wattanapiromsakul, S. Pianwanit, and S. Tewtrakul, "Anti-HIV-1 integrase compounds from *Dioscorea bulbifera* L. and molecular docking study," *Pharm Biol.*, vol. 54, no. 6, pp. 1077-1085, Feb. 2016.

S. Tewtrakul, P. Chaniad, S. Pianwanit, C. Karalai, C. Ponglimanont, and O. Yodsauae, "Anti-HIV-1 integrase activity and molecular

docking study of compounds from *Caesalpiniasappan* L.,” *Phytother. Res.*, vol. 29, no. 5, pp. 724-729, May 2015.



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J. Puripattanavong and S. Tewtrakul, “Anti-allergic and anti-inflammatory compounds from *Aglaia andamanica* leaves,” *Songklanakarin J. Sci. Technol.*, vol. 37, no. 1, pp. 37-41, 2015.

J. Puripattanavong, T. Amnuaikit, N. Radenahmad, and P. Rattanaburee, “Phytochemical study and its quantity and quality of fresh and freeze-dried young coconut juice (*Cocos nucifera* L.),” *Adv. Mat. Res.*, vol. 884-885, pp. 490-493, Jan. 2014.