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Biological evaluation of plants of Laos used in the treatment of tuberculosis in Lao traditional medicine

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Abstract

Tuberculosis has existed in Southeast Asia for thousands of years. Many traditional treatments involve herbal remedies. Over time, these traditional treatments have had the chance to become refined based on efficacy and safety. It was therefore hypothesized that plants that were used in the past and are still used today to treat symptoms associated with tuberculosis are more likely to contain anti-tubercular compounds than plants that have not been used continuously. To try to deduce which plants were used in Laos in the past, a collection of palm leaf manuscripts was studied and a list of plants used to treat symptoms associated with tuberculosis was compiled. Interviews were then conducted with contemporary healers to see if the same plants are still being used today. Plants that were found in the manuscripts and/or are presently used by healers were collected, extracted and were evaluated in an anti-tubercular assay. This paper presents the methods used to identify and collect plants used to treat symptoms indicative of tuberculosis, and the results of anti-TB assays to test for activity.

Keywords

Ethnomedicine; Laos; mycobacteria; palm leaf manuscript; Tham

Introduction

Due to many circumstances, Laos has remained relatively secluded from the outside world. It is surrounded by China and Myanmar (formerly Burma) to the North, Vietnam to the East, Cambodia to the South, and Thailand to the West, and has been referred to as “the land in between” due to its position. The country covers 236,800 km² (CIA World Factbook, 2007), an area slightly larger than the size of Utah. Undeveloped forest covers more than 40% of the country, and 80% of the population lives in rural areas as subsistence farmers. This

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situation places most of the population into close contact with medicinal plants from forested areas. Therefore, in the event of health problems, it is less expensive and easier to turn to traditional herbal remedies than to visit a government clinic for medicine. The magnitude of traditional medicine use in Laos makes it an ideal place to study ethnomedicine.

The government recognizes the importance of traditional medicine (TM) in Laos, and has established the Traditional Medicine Research Center (TMRC) under the Ministry of Public Health. An important focus of their research is to gather and catalogue indigenous traditional medicine treatments, the greater part of which consists of herbal remedies, and to publish the results of their research to serve as a reference for use by contemporary healers and the wider public. As part of its function and mandate, the TMRC has established smaller traditional medicine units called provincial traditional medicine stations (TMSs) (Soejarto et al., 1999). Initially, a TMS was established in each of 10 provinces. Each TMS is also affiliated with a traditional medicine hospital. Today, three more TM stations have been added, for a total of 13 TM stations in operation. The staff of a TMS work in direct contact with rural traditional healers who both give and receive information on treatment methods currently being used around the country.

Lao Traditional Medicine or “Ya Phurn Meuang Lao” dates back 4500 years (to the first group of Lao nation in southern part of present China), while the well documented practices started as early as the 14th century, after the establishment of the First Kingdom (Lane Xang Kingdom) (Viravong, 1973). While the earliest practices of traditional medicine in Lao PDR are still unclear, it is evident that Buddhist and Sino-Indian influences helped shape the traditional pharmacopoeia and practices used in traditional medicine. In addition, in many of the Buddhist temple-compounds, or *wats* found in every village in Lao PDR, young monks are trained in traditional medicine by older monks. Information about the uses of medicinal plants also appears in palm-leaf manuscripts located in the libraries of the larger *wats*, written several generations ago in old Lao and Pali, the language of the sacred Buddhist texts (Abhakorn, 2000).

Lao palm leaf manuscripts date back to the 1400s CE. They are typically written on leaves of the palmyra palm (*Borassus flabellifer* L., Arecaceae), which are very thick and durable. The leaves are pressed, dried, and cut into uniform shapes, usually around 10 cm in width and range anywhere from 15 to 60 cm in length. The leaves are cleaned and sanded to get a uniform writing surface, then transcribed using a stylus of wood or metal. It has actually been hypothesized that the characters used in the manuscripts developed their rounded shapes because too many straight lines has a tendency to tear the leaves. As this scratching does not give any color, a mixture of soot and oil is then rubbed on to the leaves to make the characters stand out. The leaves are attached together with string, and are often bound between two flat pieces of wood. The length often indicates the type/subject of palm leaf manuscript, with medicinal manuscripts being around the length of a forearm, while religious manuscripts are typically the length of an entire arm.

Buddhist monks in Laos have been regarded as healers for many centuries, and have kept records of their treatments in palm leaf manuscripts. Many of these manuscripts are recorded in scripts that are known only by monks or lay-people who have studied them for many years. The TMRC and the National Library of Laos have hundreds of these medical manuscripts that have yet to be translated into a modern language. There is a vast amount of information waiting to be uncovered.

In the recent past, the use of palm leaf manuscripts has declined and many are becoming subject to mould and insects before being recopied. In recognition of the amount of

information at risk of being lost, Dara Kanlaya headed up the Palm Leaf Manuscript Preservation (PLMP) Project in 1989 (Abhakorn, 1996). This project allowed researchers to travel to rural areas of the country with microfilm equipment to capture images of important manuscripts without removing them from their respective community *wats* (temples). The microfilm is kept at the National Library of Laos, which currently holds around six thousand actual bundles of palm leaves dating back to 1465 along with more than 1030 rolls of microfilm containing more than 54,000 palm leaf manuscript fascicles from more than 830 *wats*.

The TMRC holds an ample library about traditional medicines, including a collection of medical palm leaf manuscripts. Many of these manuscripts have been translated from their original scripts into the modern Lao language, and some have been translated into English. It has been one of the goals of the International Cooperative Biodiversity Group (ICBG) project, a collaborative research between the University of Illinois at Chicago (UIC), government institutions in Vietnam, and the TMRC in Laos, to explore past and contemporary traditional medicinal plant treatments as a potential source of biologically active compounds for pharmaceutical development (Soejarto et al., 1999, 2006). In this search, the information contained in the palm leaf manuscripts represents therapeutic practices of the past.

One of the disease targets in the ICBG project is tuberculosis (TB). Although TB is conventionally treated using modern antibiotics such as isoniazid, rifampin, pyrazinamide and ethambutol, the causative agent, *Mycobacterium tuberculosis*, has developed strains resistant to conventional antibiotics. Additionally, in countries such as Vietnam and Laos, modern drugs are often expensive and difficult to obtain for much of the population. There is an urgent need for alternative treatments. The aim of this research is to identify some of the herbal remedies used in contemporary and past traditional medical practices for the treatment of tuberculosis in Laos, and to investigate their effectiveness.

Materials and methods

Ethnographic field work

Two months of ethnographic fieldwork were conducted. During this time, palm leaf manuscripts, the majority of which were written in the Tham script to transcribe the Lao language, were perused for indications of tuberculosis. Because entries in these manuscripts were categorized according to symptoms, the manuscripts were investigated for symptoms of tuberculosis (chronic cough, cough with weight loss, cough with blood, cough with fever). The treatments for these entries were recorded, and a list of plants used in the treatments was compiled.

Field interviews were conducted in different regions of the country, where traditional healers were asked a series of questions about their familiarity with tuberculosis and the plants they use for treatment. Before beginning the interview, the healer was told about the goal of the project and was given the option to decline the interview or to withhold any information they did not feel comfortable giving. All of the interviews were conducted in the Lao language and digitally recorded, under a protocol approved by the UIC Institutional Review Board (IRB Protocol number 2007-0369). These field interviews yielded a second list of plants. The two lists were then compared, and the plants cited most frequently in the manuscripts and by healers were prioritized for collection. However, while in the field, if a plant was recognized that had been cited in the manuscripts, or that is presently being used by a healer for TB treatment, it was also collected.

Plant material collected and taxonomic identification

A team of healers and trained investigators from the TMRC and the ICBG project (UIC) carried out fieldwork to locate and collect the plants in both lists mentioned above. Samples of 500-1000 g (dry weight), namely the plant part(s) as indicated in the treatment, were collected. Each sample was supported by a set of fully annotated voucher herbarium specimens. Samples were dried in the sun using nylon mesh bags either in the field or at the TMRC's facilities. Herbarium specimens were identified by comparison with identified specimens in the collection holdings of the TMRC Herbarium and the John G. Searle Herbarium of the Field Museum of Natural History in Chicago, as well as by using standard floristic treatises, including Pham Hoang Ho's *Cayco Vietnam* (1999-2003). A set of voucher herbarium specimens has been deposited at each of the two institutions.

Extraction

Extraction of samples was performed at the pharmacognosy laboratories of the TMRC in Vientiane. A 25 g sample of dried plant material was soaked in 250 mL of 90% ethanol and repeated twice, using 200 mL of ethanol of the same concentration. The samples were condensed using a Heidolph Laborota 4000 rotavapor. From each plant, 2.5 g of extract was transferred into plastic bottles and transported to the University of Illinois at Chicago (UIC).

Test microorganisms

Plant extracts were tested against the following microorganisms: *Mycobacterium tuberculosis* H37Rv (ATCC 27294), *Mycobacterium smegmatis* (ATCC 607), *Escherichia coli* (ATCC 25922), *Staphylococcus aureus* (ATCC 29213), and *Candida albicans* (ATCC 10231). All test organisms were cultured at 37°C until log phase. Cultures were then harvested; aliquots were made into 1.5 mL screw-cap tubes, and stored at -80°C until use.

Antimicrobial assays

Crude plant extracts were weighed and dissolved in dimethyl sulfoxide (DMSO) to make a stock concentration of 10 mg/mL. Primary bioassays were tested at 100 µg/mL at the laboratories of the Institute for Tuberculosis Research (ITR) at UIC, using the Microplate Alamar Blue Assay (MABA) and *M. tuberculosis* (H37Rv) (Collins & Franzblau, 1997; Franzblau et al., 1998). Extracts that exhibited 85% inhibition or higher were submitted for minimum inhibitory concentration (MIC) testing.

A primary bioassay was also conducted against *S. aureus*, *E. coli*, *C. albicans*, and *M. smegmatis* to test the extracts for tuberculosis specificity. *S. aureus* and *E. coli* were tested according to a modified protocol described by NCCLS documents M7-A2 and M100-S3 (NCCLS, 1990; NCCLS, 1991). *C. albicans* was tested using a modified protocol described by NCCLS document M27-A2 (NCCLS, 2002). *M. smegmatis* was tested using MABA (Collins & Franzblau, 1997; Franzblau, et al., 1998).

Cytotoxicity assay

Cytotoxic effects of plant extracts were determined by using VERO cells (ATCC CCL-81) and rifampin as a positive control. The testing protocol followed the methods used by Falzari et al. (2005). The IC₅₀ values were calculated for crude extracts that exhibited at least 85% inhibition. The selectivity index was calculated by taking the IC₅₀ value and dividing it by the MIC value: $SI = IC_{50}/MIC$.

Results

Sixty-nine plants associated with treatments of respiratory illness were found from 29 entries in the manuscripts. Of these, four plants were cited twice and three plants were cited three times. Twenty-three interviews were conducted in three different regions of the country, leading to a list of 185 plants used by contemporary healers. All healers interviewed were male with an average age of 61.43 years, ranging from 36 to 85. The average number of years of formal schooling (in the French education system) reported was 3.64 years, ranging from no conventional formal schooling at all through completion of a university degree. Four healers asserted that they were educated at local Buddhist temples, 12 were living in Bolikhamxai province, one in Vientiane province, and 10 in Champasak province.

Of a total list of 206 plants, 11 plants were both named by healers and cited in the manuscripts. Sixty-six total plants were collected, 15 of these were cited in the manuscripts and 54 were named by traditional healers; 7 of these were both found in the palm leaf manuscripts and named by a healer. Fifty-one plants were taxonomically identified to the species level.

Upon submission to the TB bioassay, 4 plants showed inhibition above 90% and 3 showed inhibition above 85% (Table 1). Of the six plants that had inhibition above 85%, only one reported a selectivity index of greater than ten (Table 2). This extract also showed significant activity against *S. aureus*, *E. coli*, *C. albicans*, and *M. smegmatis* (Table 1).

Discussion and conclusions

Other plants in the Southeast Asian region have also been investigated and shown to possess anti-TB properties. One example is found in a study conducted by Soe et al. (2006) who tested five unnamed anti-TB ethnobotanicals from Myanmar, a country bordering on Laos. Five of these plants showed *in vitro* activity against *M. tuberculosis* H37Rv, and were given to patients with multi-drug resistant (MDR) TB who had not shown satisfactory response to the conventional first- or second-line anti-TB drugs for at least two years. After three months of administration of the extracts, in addition to the conventional second-line drugs, 13 of 15 of these patients had sputum conversion to negative.

Other ethnobotanicals containing anti-TB properties in Asia include *Justicia adhatoda* Nees (Acanthaceae), used in Nepal and India. This plant, also known as *Adhatoda vasica* contains the anti-TB compound vasicine in the leaves and led to the development of bromhexine and ambroxol, semi-synthetic derivatives added to cough syrups (Grange, 1996). The rhizome of *Alpinia galanga* (L.) Willd. (Zingiberaceae), containing a high concentration of the active compound 1'-acetoxy chavicol acetate (ACA) (Gautam, 2007), is used commonly as a spice in India and Thailand, and has shown activity against streptomycin-resistant mycobacteria (Chopra, 1957).

For plant species listed in our present study, previous findings include anti-TB testing on other *Polyalthia* species. The plant showing the highest percentage inhibition and selectivity index, *P. longifolia* Thw., has been cited as active against *M. fortuitum* and *M. smegmatis* using the disc diffusion method (Faizi, 2003). *P. cerasoides* (Roxb.) Benth. & Hook. f. ex Bedd., has produced three anti-TB compounds (Kanokmedhakul, 2007), and *P. evecta* Finet & Gagnep. has also shown activity against *M. tuberculosis* (Kanokmedhakul, 2006).

While some interesting plants were cited in the manuscripts or by contemporary healers, there were no plants that were both mentioned in the manuscripts and by healers that showed greater than 80% inhibition of *M. tuberculosis*. This does not support the original

hypothesis, but the small sample size could be misleading. The translation of more medical manuscripts and input from more contemporary healers should give a clearer indication of whether there are clear links between past and present healing systems.

All healers interviewed said that if they suspected that a member of their family had TB, they would first treat it with traditional herbal remedies. If that didn't work, 10 out of 23 healers said they would turn to a biomedical clinic. Eight healers estimated that they had treated an average of 212 patients for cough, with one well-known 85-year-old healer claiming to have treated around 1,000 people for cough, indicating the numbers of people who turn to traditional healers before government-sponsored clinics. This means that if a herbal treatment is working, tuberculosis will most likely not be diagnosed or reported at a clinic.

Difficulties encountered in our study included different plant species grouped under the same local name, as well as the same plant species having different names in different regions of the country. This could obscure the correlation of plant names from healers to those in the manuscripts if the trend was the same at the time when the manuscripts were written. For future research, the healer will lead the researchers to the plants so that there will be less confusion about the taxonomic identification of plants presently used.

Because the specific symptoms of tuberculosis vary from patient to patient, and to concretely diagnose tuberculosis requires equipment not likely to be found in rural settings where the majority of the population reside and seek primary health care, the term for cough was used to try to include treatments for anything that might be pulmonary tuberculosis. This allowed for many treatments to be included that may have nothing to do with tuberculosis.

Extracts (infusion or decoction) used by healers were derived from single plants, but there were no treatments designed by healers that contained only one plant. This research, therefore, did not look at the effectiveness of the whole treatments, but of smaller parts of the treatments. In the future, it could be beneficial to observe and obtain the actual herbal preparation from the healer to be tested in the bioassays, which could allow active synergistic effects to be detected.

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Percentage inhibition of crude EtOH extracts from plants used to treat different types of cough in Laos.

Table 1

Collection number	Scientific name	Plant data	Common name	Percentage inhibition				
				<i>S. aureus</i>	<i>E. coli</i>	<i>C. albicans</i>	<i>M. smegmatis</i>	<i>M. tuberculosis H37Rv</i>
BGE_080	<i>Polyalthia</i> sp. (Annonaceae)		tin tang teeyah	86.28%	94.22%	96.05%	83.65%	99.37%
BGE_051	<i>Haplophragma adenophyllum</i> (Wall. ex G. Don) Dop (Bignoniaceae)		kay pa	66.12%	27.91%	105.36%	-84.63%	97.74%
BGE_104	<i>Annona reticulata</i> L. (Annonaceae)		Kantalohit (peurak)	-24.11%	31.14%	65.93%	13.01%	97.36%
BGE_093S	<i>Aegle marmelos</i> (L.) Correa (Rutaceae)		mak toum	18.28%	27.05%	41.74%	-76.65%	96.55%
BGE_087	<i>Vitex trifolia</i> L. (Verbenaceae)		phi sua	-4.62%	23.80%	-12.68%	-63.69%	88.63%
BGE_066	<i>Feroniella lucida</i> Swingle (Rutaceae)		sung (mak)/kohk sung	4.15%	26.54%	22.68%	-11.46%	86.35%
BGE_068	<i>Justicia cf. adhatoda</i> L. (Acanthaceae)		hou ha (kohk hou ha)	6.38%	28.55%	46.01%	-127.39%	85.83%
BGE_072	<i>Litsea cubeba</i> Pers. (Lauraceae)		see khai tone	35.17%	26.11%	-14.97%	-125.71%	79.56%
BGE_084	<i>Tabernaemontana pallida</i> Hu (Apocynaceae)		phet pa (mak)	-15.91%	25.03%	-22.57%	-9.81%	76.84%
BGE_050	<i>Oroxylum indicum</i> (L.) Kurz (Bignoniaceae)		lin mai (mak)	57.67%	26.53%	93.91%	1.53%	74.50%
BGE_100	<i>Solanum lasiocarpum</i> Dunal (Solanaceae)		mak euk = mak keuah euk	18.11%	30.82%	-13.99%	-69.95%	71.26%
BGE_103	<i>Azadirachta indica</i> Juss. (Meliaceae)		kohm khat dao (kohm kadao)	23.43%	30.35%	1.55%	-115.13%	71.09%
BGE_046	<i>Millingtonia hortensis</i> L. f. (Bignoniaceae)		khang khong/tah salong	20.21%	31.34%	-18.12%	-103.18%	68.53%
BGE_081	<i>Myriopterion extensum</i> (Wight & Arn.) K. Schum. (Asclepiadaceae)		oy sam souan	20.39%	20.36%	43.20%	-61.20%	66.84%
BGE_058	<i>Mitragyna rotundifolia</i> (Roxb.) Kuntze (Rubiaceae)		tohm pai	19.39%	22.01%	-37.05%	-4.39%	62.86%
BGE_078	<i>Micromelum falcatum</i> (Lour.) Tanaka (Rutaceae)		summat nyai	22.69%	25.38%	79.57%	-43.48%	60.47%
BGE_089	<i>Cassia tora</i> L. (Fabaceae)		nya lap meun	20.50%	24.49%	38.76%	-127.09%	55.49%
BGE_053	<i>Stemona cochinchinensis</i> Gagnep. (Stemonaceae)		sam sip (hua)	14.73%	30.75%	-4.93%	-236.89%	52.98%
BGE_043	<i>Elephantopus</i> sp. (Asteraceae)		khi fai noh koum	-8.25%	29.98%	-23.93%	-180.72%	46.36%
BGE_083	<i>Micromelum minutum</i> Wight & Arn. (Rutaceae)		summat khao	-6.37%	24.37%	-23.00%	-8.33%	45.80%
BGE_045	<i>Colubrina pubescens</i> Kurz (Rhamnaceae)		khan toum	1.13%	30.83%	-21.08%	-158.80%	38.83%
BGE_108	<i>Saccharum officinarum</i> L. (Poaceae)		oy dam	-4.08%	29.11%	42.05%	-63.15%	33.93%
BGE_105	<i>Spondias</i> sp. (Anacardiaceae)		koh (mak koh/peurak)	-12.85%	28.40%	95.61%	0.19%	33.71%
BGE_077	<i>Clerodendrum paniculatum</i> L. (Verbenaceae)		pouang phi deng	13.14%	28.07%	31.61%	-89.38%	32.07%
BGE_055	Fam. indet.		hang yen	13.48%	30.22%	-8.07%	-50.35%	30.32%
BGE_099	<i>Sauropus androgynus</i> (L.) Merr. (Euphorbiaceae)		wan ban (hak)	-0.01%	27.54%	-34.59%	-40.93%	27.85%

Collection number	Scientific name	Plant data	Percentage inhibition					
			Common name	<i>S. aureus</i>	<i>E. coli</i>	<i>C. albicans</i>	<i>M. smegmatis</i>	<i>M. tuberculosis H37Rv</i>
BGE_079	<i>Glycosmis citrifolia</i> (Willd.) Lindl. (Rutaceae)		sohm seun	6.97%	22.37%	7.24%	-59.04%	26.19%
BGE070	Fam. indet.		kohm soum	39.58%	30.66%	75.96%	-28.97%	26.06%
BGE_062	<i>Nephrolepis exaltata</i> (L.) Schott (Davalliaceae)		good ngo = good kee pa	-5.76%	25.24%	-25.55%	-12.00%	25.56%
BGE_106	<i>Ficus glomerata</i> Roxb. (Moraceae)		deuah kieng	21.24%	30.08%	57.58%	-9.51%	20.31%
BGE_064	<i>Irvingia malayana</i> Oliver ex Bennett (Irvingiaceae)		bohk	31.32%	37.52%	97.62%	-99.58%	18.67%
BGE_074	<i>Jatropha gossypifolia</i> L. (Euphorbiaceae)		niao deng (mak)	12.88%	25.68%	-9.88%	-25.67%	18.41%
BGE_091	<i>Capparis micrantha</i> DC. (Capparaceae)		sai sou tone (hak)	-0.03%	19.32%	14.44%	-87.86%	18.29%
BGE_060	<i>Gardenia philastreii</i> Pierre ex Pit. (Rubiaceae)		kee kung; khai nao; kuk mohk	6.33%	22.65%	-1.00%	-44.53%	17.57%
BGE_075	<i>Ziziphus oenopia</i> (L.) Mill. (Rhamnaceae)		nam lep mayoh	16.92%	25.18%	-18.29%	-33.45%	16.63%
BGE_094	<i>Melothria heterophylla</i> Cogn. (Fabaceae)		tam yay	4.20%	27.63%	-21.91%	-10.62%	14.55%
BGE_101	<i>Moringa oleifera</i> Lamk. (Moringaceae)		ee houm (hak)	16.00%	26.51%	-15.15%	-39.44%	14.25%
BGE_085	<i>Strychnos nux-blanda</i> A.W. Hill (Loganiaceae)		toum kah khao	-10.98%	25.67%	39.87%	-11.63%	14.18%
BGE_063	<i>Gardenia obtusifolia</i> Roxb. (Rubiaceae)		khai nao (noy)	10.74%	29.52%	2.51%	-52.44%	13.26%
BGE_086	<i>Dimocarpus longan</i> Lour. (Sapindaceae)		kha leen	29.70%	24.54%	24.95%	-23.18%	11.75%
BGE_073	<i>Jatropha curcas</i> L. (Euphorbiaceae)		niao khao (mak)	7.23%	27.73%	-6.72%	-63.86%	11.75%
BGE_092	<i>Chaetocarpus castanocarpus</i> Thw. (Euphorbiaceae)		bohk khai	3.64%	27.48%	93.91%	24.02%	10.81%
BGE_069	Fam. indet.		kho phou	48.77%	22.64%	67.21%	-29.24%	10.77%
BGE_047F	<i>Spondias dulcis</i> Parkins. (Anacardiaceae)		kohk keuum	22.05%	41.29%	93.08%	-23.28%	10.42%
BGE_065	<i>?Solanum</i> sp. (Solanaceae)		mak keuah keun (hak)	-12.99%	27.16%	-15.41%	-18.91%	8.59%
BGE_067	<i>?Partnari</i> sp. (Chrysobalanaceae)		pohk	40.14%	24.50%	72.83%	-107.22%	5.91%
BGE_044	<i>Randia longiflora</i> Lamk. (Rubiaceae)		khat khao	13.55%	32.00%	-8.87%	-150.62%	4.60%
BGE_093F	<i>Aegle marmelos</i> (L.) Correa (Rutaceae)		mak toum	25.03%	28.24%	8.98%	-101.30%	2.37%
BGE_107	<i>Drynaria bonii</i> H. Christ. (Polypodiaceae)		good hohk	18.68%	32.76%	-20.31%	32.71%	1.80%
BGE_090	Fam. indet.		phi mob	25.40%	23.27%	-17.16%	-65.51%	1.57%
BGE_047S	<i>Spondias dulcis</i> Parkins. (Anacardiaceae)		kohk keuum	8.92%	38.08%	62.54%	-229.81%	-0.56%
BGE_061	<i>Acronychia pedunculata</i> (L.) Miq. (Rutaceae)		kohm loh wan joh	77.53%	26.94%	30.24%	-42.07%	-0.78%
BGE_071	<i>Melastoma decemfidum</i> Roxb. (Melastomataceae)		ben ah/en ah	16.58%	34.52%	99.03%	-19.21%	-3.86%
BGE_059	<i>Lasia spinosa</i> (L.) Thw. (Araceae)		bo nam; pak nam	49.12%	29.91%	66.45%	-230.66%	-5.00%
BGE_082	<i>?Ardisia</i> sp. (Myrsinaceae)		tin cham kohn	-5.86%	26.68%	97.30%	56.78%	-5.84%
BGE_048	<i>Heteropanax fragrans</i> (Roxb.) Seem. (Araliaceae)		oy sang	-14.32%	26.45%	38.09%	-18.97%	-7.21%

Plant data			Percentage inhibition				
Collection number	Scientific name	Common name	<i>S. aureus</i>	<i>E. coli</i>	<i>C. albicans</i>	<i>M. smegmatis</i>	<i>M. tuberculosis H37Rv</i>
BGE_102	<i>Amaranthus spinosus</i> L. (Amaranthaceae)	pak hohm (hak)	3.48%	25.92%	-25.64%	-77.59%	-7.37%
BGE_057	<i>Lycopodium cernuum</i> L. (Lycopodiaceae)	good ngo	5.93%	30.94%	-20.60%	-11.63%	-8.58%
BGE_088	<i>Smilax</i> sp. (Smilacaceae)	keuah khao mohk	0.22%	35.72%	-19.05%	-68.67%	-9.35%
BGE_049	<i>Vitex peduncularis</i> Wall. ex Schauer (Verbenaceae)	tin nohk	-10.10%	25.62%	53.16%	-34.39%	-9.69%
BGE_056	<i>Ficus hispida</i> L.f. (Moraceae)	deua pong	7.02%	31.08%	38.11%	-85.61%	-11.49%
BGE_098	<i>Dalbergia</i> sp. (Fabaceae)	padoung deua khai = padoung poh = padoung koh	-1.96%	26.57%	-18.37%	-49.24%	-12.04%
BGE_054	<i>Caryota mitis</i> Lour. (Arecaceae)	tao hang	17.20%	36.61%	-3.52%	-63.11%	-17.72%
BGE_052	<i>Salacta chinensis</i> L. (Celastraceae)	tah kai	30.60%	23.29%	78.71%	-153.42%	-18.14%
BGE_076	<i>Antidesma diandrum</i> (Roxb.) Roth (Euphorbiaceae)	mao (mak)	23.41%	27.40%	97.31%	-56.28%	-22.86%

Table 2*In vitro* selectivity for active ethanol. extracts from plants used to treat cough.

Extract	Percentage Inhibition	MIC (µg/mL)*	IC ₅₀ (µg/mL)*	Selectivity Index
BGE_051	97.74	83.25	83.98	1.0088
BGE_066	86.35	91.54	51.31	0.56052
BGE_068	85.83	NA	>100	NA
BGE_080	99.37	0.72	51.49	71.514
BGE_087	88.63	81.02	54.76	0.67588
BGE_093	96.55	54.88	47.35	0.86279
BGE_104	97.36	49.17	0.41	0.00833
Rifampin	100	0.06	97.79	1629.8

*
Test Concentration at 100 µg/mL.