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Full Length Research Paper

Antimicrobial, antioxidant and anticancer activities of Thai local vegetables

S. Nanasombat[1]* and N. Teckchuen [2]

[1]Department of Applied Biology, Faculty of Science, King Mongkut's Institute of Technology Ladkrabang, Bangkok, Thailand.
[2]Department of Chemistry, Faculty of Science, King Mongkut's University of Technology Thonburi, Bangkok, Thailand.

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Thai local vegetables (20 species) were screened for their antimicrobial and antioxidant activities. Methanolic extracts of *Cassia siamea*, *Garcinia cowa*, *Limnophila aromatica* and *Polygonum odoratum* exhibited strong antioxidant and antibacterial activities. The sensitive bacteria to these extracts were *Bacillus cereus*, *Listeria monocytogenes* and *Staphylococcus aureus.* Among all plant extracts, the extract of *P. odoratum* had the highest phenolic content and antioxidant activity with EC_{50} value of 315.35 µg extract/mg DPPH (2,2-diphenyl-1-picrylhydrazyl). These plant extracts were also tested for cytotoxicity against human oral epidermal carcinoma (KB), breast adenocarcinoma (MCF-7) and small cell lung carcinoma (NCI-H187). *P. odoratum* extract was moderately active against MCF-7 (IC_{50} value of 6.01 µg/ml). Major types of active compounds in *P. odoratum* extract were identified by HPLC method. Flavonoids found in this plant were rutin, catechin, quercetin, kaempferol and isorhamnetin. Among these compounds, rutin was found in the highest amount (3.77% w/w dry extract).

Key words: *Polygonum odoratum*, *Cassia siamea*, *Garcinia cowa*, *Limnophila aromatica*, phenolic compounds, flavonoids.

INTRODUCTION

In recent years, phytochemicals in vegetables have received a great deal of attention mainly on their role in preventing diseases caused as a result of oxidative stress which releases reactive oxygen species such as singlet oxygen and various radicals as a damaging side-effect of aerobic metabolism. These radicals are possibly involved in a number of disorders including cardiovascular malfunctions, tissue injury, DNA damage and tumour promotion (Ames, 1983; Horton, 2003). Several studies suggest that antioxidants could prevent accumulation of these reactive oxygen species and be beneficial for treatment of these pathologies (Horton, 2003). Diets rich in fruits and vegetables have been reported to associate consistently with reduced risk of a variety of tumors, especially epithelial cancers of the respiratory and gastrointestinal tract (Swanson, 1998). Therefore, there has been interest in in finding natural antioxidants from plant materials.

Thai local vegetables comprised more than 150 species of plants, consuming either fresh or cooked vegetables. Some are used as ingredients in Thai cuisine. Apart from being a good source of nutrients such as carbohydrate, protein, dietary fiber, vitamins and minerals, they are known for their therapeutic value. Some plants were reported to possess antimicrobial activity (Alzoreky and Nakahara, 2002). Recent research has done on investigation of antioxidant, antimicrobial, anti-inflammatory and anticancer activities of Thai medicinal plants (Mahidol et al., 2002). Antioxidant activity of *Acacia pennata*, *Cassia siamea*, *Centella asiatica*, *Coccinia grandis*, *Ocimum americanum* and *Limnophila aromatica* leaves was reported (Chanwitheesuk et al., 2005). However, there are many species of Thai local vegetables that have not been investigated on these properties. Thus, the aims of this work were to determine antimicrobial, antioxidant and anticancer activities of some Thai local vegetables and

*Corresponding author. E-mail: knsuree@kmitl.ac.th. Tel.: 66 2737 3000 ext. 6264. Fax: 66 2326 4354.

Table 1. Thai local vegetables used in this study.

Botanical names	Common names/ Thai names	Plant part
Acacia pennata subsp. *insuavia*	Thorny tree/ *Cha-Om*	Leaves
Anethum graveolens Linn.	Dill/ *Phak-Chee-Laao*	Leaves
Cassia siamea Britt.	Cassod Tree/ *Khi-Leck*	Leaves
Centella asiatica (Linn.) Urban	Asiatic pennywort/ *Boa-Bok*	Leaves
Coccinia grandis (l.) Voigt.	Ivy gourd/ *Tum-Lueng*	Leaves
Coriandrum spp.	*Hom-Yae*	Leaves
Diplazium esculentum (Retz.) Sw.	Paco/ *Phak-Good*	Leaves
Eryngium foetidum Linn.	Garden parsley/ *Phak-Chee-Farang*	Leaves
Garcinia cowa Roxb.	*Cha-Muang*	Leaves
Lasia spinosa Thw.	*Phak-Nam*	Leaves
Limnophila aromatica Merr.	Rice paddy herb/ *Phak- Ka-Yeang*	Leaves
Morinda citrifolia Linn.	Noni, Indian Mulberry/ *Yau*	Leaves
Momordica charantia Linn.	Bitter cucumber/ *Ma-Ra-Khi-Nok*	Fruits
Ocinum americanum Linn.	Lemon basil/ *Meang-Luck*	Leaves/ flowers
Ocimum gratissimum Linn.	Tree basil, Shrubby basil / *Yee-Ra*	Leaves
Polygonum odoratum Lour.	Vietnamese coriander/ *Phak-Paew*	Leaves
Sesbania grandiflora Desv.	Vegetable Humming Bird/ *Kae*	Flowers
Sesbania javanica Miq.	Sesbania flower/ *Sa-No*	Flowers
Spilanthes acmella Murr.	Para cress/ *Phak-Krad*	Leaves/ flowers
Tiliacora triandra Diels.	Bamboo grass/ *Ya-Nang*	Leaves

identify bioactive compounds of the selected vegetable.

MATERIALS AND METHODS

Microorganisms

19 microbial strains were used in this study. *Bacillus cereus* DMST 5040, *Enterobacter aerogenes* DMST 8841, *Escherichia coli* DMST 4212, *Klebsiella pneumoniae* DMST 8216, *Listeria monocytogenes* DMST 11256, *Pseudomonas fluorescens* DMST 20076, *Salmonella* Rissen DMST 7097, *Salmonella Typhimurium* DMST 0562 and *Yersinia enterocolitica* DMST 9380 were obtained from the culture collection of the department of medical sciences, ministry of public health, Thailand. *Staphylococcus aureus* TISIR 118, *Lactobacillus plantarum* TISTR 050, *Leuconostoc mesenteroides* TISTR 053, *Pediococcus pentosaceus* TISTR 414, *Candida lipolytica* TISTR 5655, *Hanseniaspora uvarum* TISTR 5153, *Pichia membranaefa-ciens* TISTR 5093, *Rhodotorula glutinis* TISTR 5159, *Schizosaccharomyces pombe* TISTR 5205, and *Zygosaccharomyces rouxii* TISTR 5044 were obtained from the TISTR culture collection at the Thailand institute of scientific and technological research.

Culture preparation

Each microbial culture was transferred individually to 5 ml of deMan Rogosa Sharpe medium (MRS broth, Difco) for lactic acid bacteria, Tryptic soy broth (TSB, Difco) for other bacteria and Saboraud dextrose broth (SDB, Difco) for yeasts. Cultures of bacteria and yeasts were incubated at 37 and 30°C for 24 and 72 h, respectively. The microbial cells were harvested by centrifugation at 3000 rpm for 15 min, washed twice and resuspended in 0.1% peptone water. Turbidity was adjusted to match that of 5 McFarland standard (0.5 for yeasts) to obtain a cell concentration of 10^7 CFU/ml.

Extraction of plant materials

20 species of Thai local vegetables (Table 1) were purchased at retail in Bangkok and Chonburi, Thailand during October-November, 2005. The plant materials were cut into small pieces, freeze dried and powdered. Then, 10 g of each were soaked in 150 ml methanol and shaked at 200 rpm for 24 h at ambient temperature. The mixtures were filtered. The filtrates were evaporated using vacuum rotary evaporator and air dried at 40°C. Stock solutions of crude methanolic extracts were prepared by diluting the dried extracts with 10% dimethyl sulphoxide (DMSO) solution to obtain a final concentration of 400 mg/ml.

Screening of Thai local vegetable extracts using disk diffusion technique

The disk diffusion test was performed using the procedure as described by Jorgensen et al., 1999. The inoculum suspension of each microbial strain was swabbed on the entire surface of Mueller-Hinton agar (MHA, Difco) for bacteria and SDA for yeasts. Sterile 6 mm filter paper discs were aseptically placed on MHA and SDA surfaces. Crude methanolic extracts (15 µl) were immediately added to discs. A 15 µl aliquot of 10% DMSO was also added to a sterile paper disc as a negative control. Penicillin G (10 unit/ml) and amphotericin B (10 mg/ml) were used as positive controls. The plates were left at ambient temperature for 15 min to allow excess prediffusion of extracts prior to incubation at 37°C for 24 h for bacteria and at 30°C for 72 h for yeasts. Diameters of inhibition zones were measured. The experiment was done in triplicate.

Determination of the minimum inhibitory concentration

The microbroth dilution test was performed to determine minimum

inhibitory concentrations (MICs) using the procedure as described by Jorgensen et al., 1999. 100 µl of Mueller-Hinton broth (MHB, for bacteria) or SDB (for yeasts) were added into each well of a microtiter plate. The 100 µl aliquot of stock solution of crude methanolic extract (400 mg/ml) was added and subsequently 2-fold serially diluted with broth. The cell suspension (20 µl) at concentration of 10^7 cells/ml was added into each well. The final concentrations of the extracts ranged from 166.7 to 2.6 mg/ml. The negative control were also performed using 10% DMSO. Penicillin G (50,000 - 0.005 unit/ml) was used as a positive control drug. Duplicate wells were run for each concentration of the extracts. After incubation, the turbidity was measured at 620 nm using the microplate reader (iEMS Reader MF, Labsystems). The lowest concentration that inhibited visible growth was recorded as the MIC.

Determination of free radical scavenging activity using DPPH method

Free radical scavenging activity of the extracts was measured according to Brand-Williams et al. (1995). Stock solution of extracts (10,000 µg/ml) was prepared and diluted to obtain final concentrations of 1,000, 500, 100, 50, 10 and 1 µg/ml in methanol. The α-tocopherol was used as a positive control. Each diluted extract (75 µl) was added to 2.925 ml of a 0.025 g/l DPPH (2,2-diphenyl-1-picrylhydrazyl) solution in methanol. The reaction mixtures were then incubated in the dark for 30 min and the absorbance at 515 nm was measured using a UV-visible spectrophotometer (Shimadzu, UV 1601). The remaining DPPH concentration in the reaction mixture was calculated from the DPPH standard curve and the remaining DPPH (%) was calculated using the following equation.

$$\%DPPH^{\cdot}_{REM} = [DPPH^{\cdot}]_T / [DPPH^{\cdot}]_{T=0} \times 100$$

Where $[DPPH]_T$ and $[DPPH]_{T=0}$ were the concentration of DPPH at steady state and zero time, respectively. The remaining DPPH concentration (%) in each reaction mixture was then plotted against µg of antioxidant (extract) /mg of DPPH to obtain EC_{50} value (concentration of antioxidant needed to decrease the initial DPPH by 50%). The lower the EC_{50}, the higher the antioxidant power. The EC_{50} values were calculated using the following linear regression of plots. Antiradical efficiency (AE = 1/ EC_{50}) was calculated.

$$[\%DPPH_{REM}] = b\,[\mu g \text{ antioxidant/ mg DPPH}] + a$$

Total phenolic assay

Total phenolic contents of all plant extracts were determined by the procedure as described by Tepe et al. (2005). Each stock solution of the extracts was prepared to obtain the concentration of 1,000 µg/ml and 100 µl of each were transferred to a flask containing 46 ml distilled water. Folin-Ciocalteu reagent (1 ml) was then added. The mixture was shaken thoroughly and allowed to stand for 3 min. 3 ml of 2% Na_2CO_3 were added to the mixture and allowed to stand for 2 h with intermittent shaking. The absorbance was measured at 760 nm using UV-Visible spectrophotometer (Shimadzu, UV 1601). Standard curve of gallic acid was prepared using the similar procedure. The results were expressed as µg GAE (gallic equivalents)/mg dry extract.

Cytotoxicity assay

Determination of anticancer activity

The selected plant extracts were tested for their cytotoxicity against KB (human epidermal carcinoma of cavity, ATCC #CCL-17) and MCF-7 (human breast adenocarcinoma, ATCC #HTB-22) by colorimetric cytotoxicity assay that measured cell growth from cellular protein content (Skehan et al., 1990) and NCI-H187 (human small cell lung carcinoma, ATCC #CRL-5804) by the procedure as described by Plumb et al. (1989). Ellipticine and doxorubicin were used as positive controls and DMSO was used as a negative control. Cytotoxicity was expressed as IC_{50} value which is the concentration of extract needed to inhibit cell growth by 50%.

Determination of cytotoxicity against vero cells

The selected plant extracts were tested for their cytotoxicity against normal vero cells (African green monkey kidney fibroblast) in 96 well microplates. Vero cell suspension (190 µl) containing 1.0×10^5 cells/ml and 10 µl of tested sample were added into each well. After incubation in 5% CO_2 at 37°C for 72 h, the cytotoxicity was determined by colorimetric method as described by Skehan et al. (1990).

HPLC analysis of the selected plant extract

The dried extract was dissolved in methanol and subsequently 10-fold diluted in the mixture of methanol-acetonitrile-water (40: 15: 45, v/v/v) containing 1.0% acetic acid (mobile phase). This mixture was filtered through a 0.45 µm nylon syringe filter before injection (20 µl) into a reverse phase ZORBAX SB-C18 analytical HPLC column (4.6 mm × 250 mm i. d.) at ambient temperature. The elution system was isocratic and the flow rate was 0.8 ml/min. Wavelengths of detection were set at 297 nm for catechin and 368 nm for rutin, quercetin, kaempferol and isorhamnetin. The chromatographic peaks were confirmed by addition of standard compounds and comparing their retention times with those of the reference compounds. The amount of each compound in the extract was calculated from their standard curves. The HPLC analysis was performed in triplicate.

RESULTS AND DISCUSSION

Preliminary screening of Thai local vegetable extracts

5 out of 20 plant species had antimicrobial activity against some microbial strains (Table 2). *G. cowa* extract had the broadest antimicrobial activity. It inhibited growth of 8 bacterial species. *L. aromatica* and *P. odoratum* extracts were active against 6 species, while the extracts of *A. pennata* and *C. siamea* inhibited growth of only 2 species. All plant extracts were inactive against all yeast strains tested.

Minimum inhibitory concentrations

The extracts of *G. cowa*, *L. aromatica* and *P. odoratum* had higher antibacterial activity than those of *A. pennata* and *C. siamea* (Table 3). *B. cereus* and *S. aureus* were the most susceptible strain to *L. aromatica* extract (2.6 mg/ml MIC), while the most vulnerable species to *G. cowa* extract were *L. monocytogenes* and *Y. enterocolitica* (5.2 mg/ml MIC). *B. cereus* was also susceptible to ex-

Table 2. Antimicrobial activity of crude methanolic extracts of Thai local vegetables using disk diffusion test

Microorganisms	Diameter of Inhibition Zone (mm)[a]						
	Acacia pennata	***Cassia siamia***	***Garcinia cowa***	***Limnophila aromatica***	***Polygonum odoratum***	**Penicillin G**	**Amphotericin B**
Bacillus cereus	8.5 ± 1.0	9.3 ± 1.6	20.2 ± 3.6	21.0 ± 5.2	15.3 ± 5.5	17.7 ± 0.6	-[b]
Enterobacter aerogenes	-	-	-	-	-	-	-
Escherichia coli	-	-	8.2±1.3	-	8.2 ± 0.3	-	-
Klebsiella pneumoniae	-	-	9.7 ± 1.5	-	-	-	-
Listeria monocytogenes	-	7.5 ± 0.5	11.0 ± 1.0	12.2 ± 3.4	11.5 ± 0.9	38.7 ± 1.1	-
Pseudomonas fluorescens	-	-	13.3 ± 0.8	9.7 ± 4.2	10.2 ± 2.8	-	-
Salmonella Rissen	-	-	8.5 ± 1.3	-	-	9.7 ± 0.6	-
Salmonella Typhimurium	-	-	-	8.7 ± 0.6	-	22.7 ± 0.6	-
Staphylococcus aureus	-	-	10.2 ± 0.3	12.5 ± 2.5	11.2 ± 2.9	41.7 ± 0.6	-
Yersinia enterocolitica	-	-	12.2 ± 4.3	11.2 ± 3.9	11.0 ± 4.1	34.0 ± 0.0	-
Lactobacillus plantarum	7.7 ± 0.8	-	-	-	-	23.8 ± 2.3	-
Leuconostoc mesenteroides	-	-	-	-	-	24.3 ± 3.1	-
Pediococus pentosaceus	-	-	-	-	-	22.3 ± 2.3	-
Candida lipolytica	-	-	-	-	-	15.0 ± 1.0	-
Hanseniaspora uvarum	-	-	-	-	-	-	15.3 ± 0.6
Pichia membranaefaciens	-	-	-	-	-	-	10.2 ± 0.3
Rhodotorula glutinis	-	-	-	-	-	-	22.0 ± 0.0
Schizosaccharomyces pombe	-	-	-	-	-	-	21.7 ± 1.2
Zygosaccharomyces rouxii	-	-	-	-	-	-	22.2 ± 0.3

[a] Data are mean of three replications.
[b] No inhibition was observed.

Table 3. Minimum inhibitory concentrations of crude methanolic extracts of Thai local vegetables.

Microorganisms	Minimum inhibitory concentrations (mg/ml)					
	Acacia pennata	***Cassia siamea***	***Garcinia cowa***	***Limnophila aromatica***	***Polygonum odoratum***	**Penicillin G (unit/ml[a])**
Bacillus cereus	41.7	5.2	20.8	2.6	41.7	5.0×10^3
Escherichia coli	>166.7	>166.7	20.8	>166.7	166.7	5.0×10^3
Klebsiella pneumoniae	>166.7	>166.7	10.4	>166.7	83.3	5.0×10^3
Listeria monocytogenes	>166.7	20.8	5.2	20.8	83.3	5.0×10^2
Pseudomonas fluorescens	>166.7	>166.7	10.4	20.8	83.3	$>5.0 \times 10^4$
Salmonella Rissen	>166.7	>166.7	10.4	>166.7	>166.7	5.0×10^4
Salmonella Typhimurium	>166.7	>166.7	>166.7	10.4	>166.7	5.0
Staphylococcus aureus	>166.7	>166.7	41.7	2.6	83.3	$>5.0 \times 10^4$
Yersinia enterocolitica	>166.7	>166.7	5.2	41.7	166.7	$>5.0 \times 10^4$
Lactobacillus plantarum	20.8	>166.7	166.7	83.3	20.8	5.0×10^4

[a] Unit/ml for Penicillin G only.

extract of *C. siamea* with 5.2 mg/ml MIC.

In this study, *G. cowa* extract had strong antibacterial activity. Fruits of *G. cowa* have been reported to contain 15 tetraoxygenated xanthones and these compounds had antibacterial activity (Panthong et al., 2006). In addition, bark of *G. cowa* has been reported to inhibit growth of *Aspergillus flavus* (Joseph et al., 2005). Recently, Negi et al. (2008) reported that crude hexane and chloroform extracts from fruit rinds of *G. cowa* inhibited growth of *Bacillus coagulans*, *Bacillus subtilis*, *B. cereus* and *S. aureus*. Antimicrobial activity of *L. aromatica* extract may be due its essential oil which consists of limonene and peril-aldehyde as major compounds, flavone and flavonoids (Bui et al., 2004).

Table 4. DPPH radical-scavenging activity and total phenolic contents of plant extracts.

Plant species	EC_{50} (µg extract / mg DPPH)[a] ± SD	Antiradical efficiency ($\times 10^{-5}$)[a] ± SD	Total phenolic content (µg Gallic Acid Equivalents (GAE)/mg dry extract)[a] ± SD
Acacia pennata	3,565.7 ± 41.5	28.1 ± 0.3	45.3 ± 5.8
Anethum graveolens	2,297.8 ± 80.4	43.6 ± 1.5	12.0 ± 0.0
Cassia siamea	349.9 ± 5.7	285.8 ± 4.7	35.3 ± 5.8
Centella asiatica	6080.0 ± 80.5	16.4 ± 0.2	7.8 ± 1.9
Coccinia grandis	6,640.2 ± 52.4	15.1 ± 0.1	14.4 ± 1.9
Coriandrum spp.	1,868.8 ± 62.3	53.6 ± 1.8	13.3 ± 0.0
Diplazium esculentum	3,353.2 ± 50.6	29.8 ± 0.04	3.7 ± 2.9
Eryngium foetidum	4,739.7 ± 61.3	21.1 ± 0.3	3.7± 2.9
Garcinia cowa	1597.5 ± 99.3	62.7 ± 4.0	2.0 ± 0.0
Lasia spinosa	3,105.6 ± 45.9	32.2 ± 0.5	2.0 ± 0.0
Limnophila aromatica	550.5 ± 12.2	181.7 ± 4.0	42.0 ± 0.0
Momordica charantia	4,230.8 ± 33.8	23.6 ± 0.2	3.7 ± 2.9
Morinda citrifolia	990.5 ± 26.1	101.0 ± 2.8	0.3 ± 2.9
Ocimum americanum	1,817.3 ± 61.3	55.1 ± 1.8	25.3 ± 5.8
Ocimum gratissimum	3,321.9 ± 54.9	30.1± 0.5	14.4 ± 1.9
Polygonum odoratum	315.4 ± 18.8	317.9 ± 19.5	52.0 ± 0.0
Sesbania grandiflora	13,425.9 ± 97.0	7.4 ± 0.1	12.0 ± 0.0
Sesbania javanica	2,265.6 ± 83.0	49.0 ± 1.9	2.0 ± 0.0
Spilanthes acmella	4,963.7 ± 79.9	20.2 ± 0.3	18.7 ± 2.9
Tiliacora triandra	3,903.9 ± 32.9	25.6 ± 0.2	13.3 ± 0.0
∝-tocopherol	322.4 ± 0.6	310.9 ± 0.4	-[b]

[a] Data are mean of three replications. [b] not determined

Antioxidant activity and total phenolic content

Among all plant extracts, extract of *P. odoratum* had the highest antioxidant activity with an EC_{50} of 315.4 µg extract/mg DPPH (AE = 317.9×10^{-5}) followed by α-tocopherol and the extracts of *C. siamea*, *L. aromatica*, *M. citrifolia*, *G. cowa* and etc. (Table 4). Total phenolic contents of all extracts were in the range of 0.3 - 52.0 µg GAE/mg dry extract. The extract of *P. odoratum* contained the highest content of phenolic compounds (52.0 µg GAE/mg dry extract). Antioxidant activity of these plant extracts may be due to their active constituents. Starkenmann et al. (2006) found many organic compounds ((*Z*)-3-hexenal, (*Z*)-3-hexenol, decanal, undecanal and dodecanal) in *P. odoratum*. Thongsaard et al. (2001) reported that leaves and flowers of C. siameas had an actice compound, barakol. Flavonoids, nevadensin 7-*O*-glycoside and 8-hydroxylated flavones were detected in *L. aromatica* (Bui et al., 2004), while 5 xanthones, *cowa* garcinone A - E were isolated from latex of *G. cowa* (Mahabusarakam et al., 2005).

Cytotoxicity against cancer cell lines and normal vero cells

As a result of strong antimicrobial and antioxidant activities, *C. siamea*, *G. cowa*, *L. aromatica* and *P. odoratum* were selected for further investigation on anticancer activities. The only plant extract showing inhibitory effect on breast cancer cell (MCF-7) proliferation was *P. odoratum* with the IC_{50} of 6.01 µg/ml (Table 5). This was probably related to its strongest antioxidant activity. This correlation has previously been found in other plant extracts (Mackeen et al., 2000; Li et al., 2008). In addition, vero cells (normal cells) showed a particularly low sensitivity to these extracts with the IC_{50} of >50.0 µg/ml. Therefore, they were not toxic to these cells. Boivin et al. (2009) reported that cruciferous vegetable extracts inhibited all tested cancer cell line proliferation. The antiproliferative effect of vegetables was found to be specific with the cells of cancerous origin and largely independent of their antioxidant properties.

Major active compounds in the selected plant extract

The HPLC analysis indicated that *P. odoratum* contained some flavonoids such as quercetin, rutin, catechin, isorhamnetin and kaempferol (Figure 1). Among all compounds, rutin was the most abundant constituent (3.77% w/w dry extract), followed by catechin (0.34%), quercetin (0.079%), kaempferol (0.009%) and isorhamnetin (0.007%). Rutin in combination with other flavonoids may cause strong antioxidant activity and antiproliferative ef-

Table 5. IC_{50} value of the selected plant extracts against three cancer cell lines.

Plant species	Cell lines (IC_{50}, µg/ml[a])[b]		
	KB	MCF-7	NCI-H187
Cassia siamea	>20.0	>20.0	>20.0
Garcinia cowa	>20.0	>20.0	>20.0
Limnophila aromatica	>20.0	>20.0	>20.0
Polygonum odoratum	>20.0	6.01 ± 0.08	>20.0
Ellipticine	0.61 ± 0.10	0.64 ± 0.16	0.60 ± 0.00
Doxorubicine	0.18 ± 0.02	0.23 ± 0.10	0.03 ± 0.00

[a] Data are mean of two replications.
[b] The extracts with an IC_{50} value of >20 were considered inactive, but >10.0 - 20.0 was weakly active, 5.0-10.0 was moderately active and < 5.0 was strongly active.

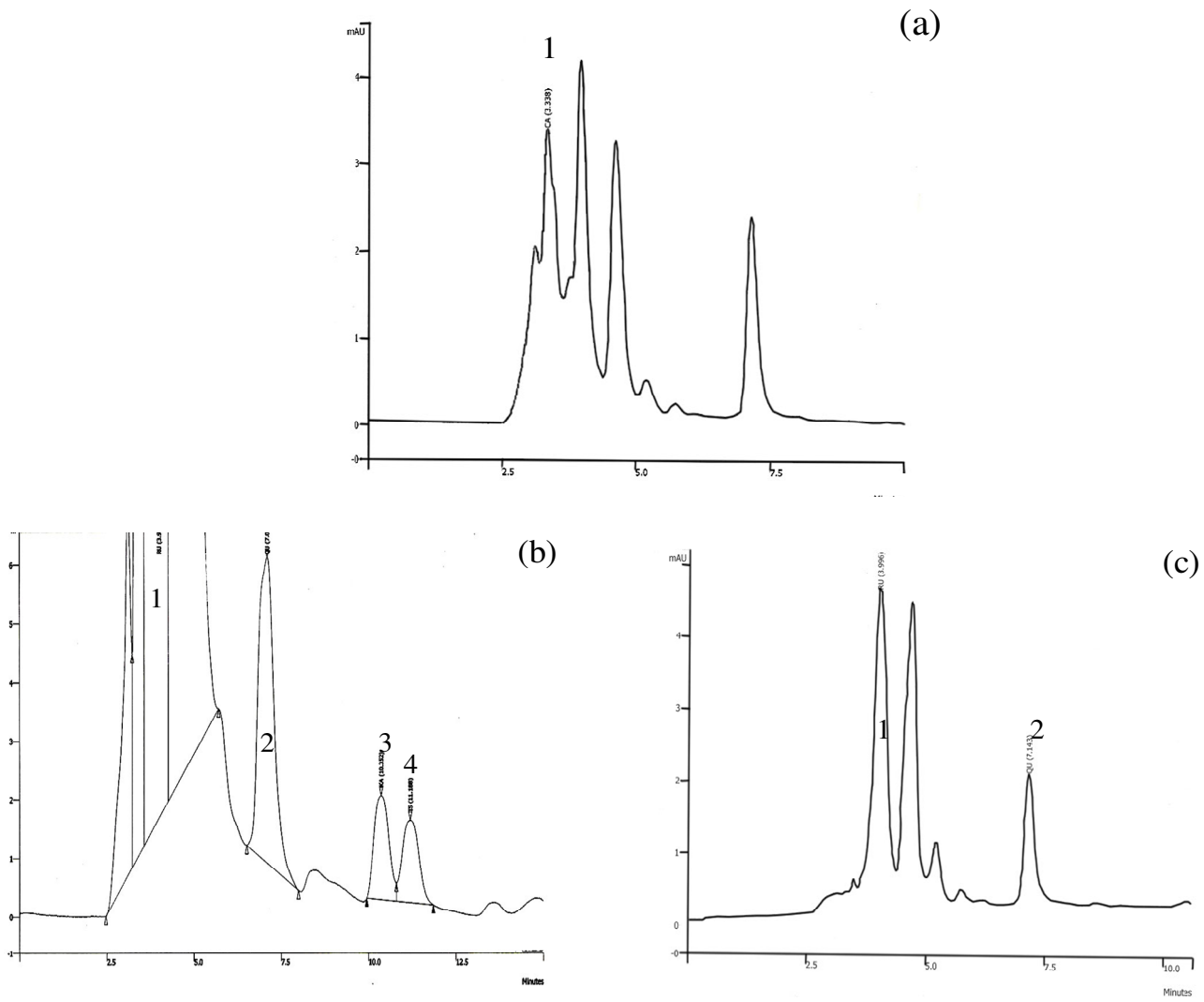

Figure 1. Chromatogram of active compounds in *Polygonum odoratum* extract:
(a) 100 ppb extract injected (measured at 297 nm), peak 1: catechin
(b) 500 ppb extract injected (measured at 368 nm), peak 1: rutin; peak 2: quercetin; peak 3: kaempferol; peak 4: isorhamnetin
(c) 100 ppb extract injected (measured at 368 nm), peak 1: rutin; peak 2: quercetin

fect on breast cancer cells. Plant extracts containing catechin, epicatechin, quercetin, kaempferol, rutin, and etc. have shown to decrease proliferation of breast, pancreatic, prostate and other cancer cell lines (Kampa et al., 2007).

In conclusion, *P. odoratum* possessed strong antioxidant activity and anti-breast cancer activity. Significant amounts of flavonoids existed in its leaves, especially rutin which found in higher amount compared to other compounds. Therefore, supplementing a balanced diet with *P. odoratum* leaves may have beneficial health effects. In addition, this plant could be used as a source of rutin for production of anti-breast cancer drug, or could be beneficial in food preservation by applying it as a natural antioxidant in high-fat food products to delay lipid oxidation. Its ability to inhibit lipid oxidation in food products requires further studies.

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