ORIGINAL ARTICLE

In vitro effects of Thai medicinal plants on human lymphocyte activity

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Abstract

Sriwanthana, B., Treesangsri, W., Boriboontrakul, B., Niumsakul, S., and Chavalittumrong, P.
***In vitro* effects of Thai medicinal plants on human lymphocyte activity**
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We assessed the effects of *Cleistocalyx nervosum var paniala, Gynostemma pentaphyllum, Gynura procumbens, Houttuynia cordata, Hyptis suaveolens, Portulaca grandiflora, Phytolacca americana* and *Tradescantia spathacea* on lymphocyte proliferation and the effects of *C. nervosum, G. pentaphyllum, H. suaveolens* and *P. grandiflora* on natural killer (NK) cells activity. All of the extracts significantly stimulated human lymphocyte proliferative responses at various concentrations depending on each extract. The extracts of *C. nervosum* and *H. suaveolens* were significantly enhanced NK cells activity while those of *G. pentaphyllum* and *P. grandiflora* did not alter NK cells function. Our results suggested that the extracts of those plants have stimulating activity on human lymphocytes and could be clinically useful for modulating immune functions of the body.

Key words : *Cleistocalyx nervosum*, *Gynostemma pentaphyllum*, *Gynura procumbens*, *Houttuynia cordata*, *Hyptis suaveolens*, *Portulaca grandiflora*, *Phytolacca americana*, *Tradescantia spathacea*, lymphocyte proliferation, NK activity, immunostimulating agents

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ฤทธิ์ของสารสกัดสมุนไพรไทยต่อการทำงานของลิมโฟซัยท์ในหลอดทดลอง

ว. สงขลานครินทร์ วทท. มีนาคม 2550 29(ฉบับพิเศษ 1) : 17-28

ได้ทำการศึกษาเพื่อทดสอบฤทธิ์ของสารสกัดสมุนไพรไทย 8 ชนิด คือ มะเกี๋ยง (*Cleistocalyx nervosum var paniala*) ปัญจขันธ์ (*Gynostemma pentaphyllum*) แป๊ะตำปึง (*Gynura procumbens*) พลูคาว (*Houttuynia cordata*) แมงลักคา (*Hyptis suaveolens*) แพรเซี่ยงไฮ้ (*Portulaca grandiflora*) พิษลักษณ์ (*Phytolacca americana*) และว่านกาบหอย (*Tradescantia spathacea*) ต่อการกระตุ้นการแบ่งตัวของลิมโฟซัยท์ (lymphocyte proliferation) และฤทธิ์ของปัญจขันธ์ แพรเซี่ยงไฮ้ มะเกี๋ยง และแมงลักคาต่อการทำงานของเซลล์ที่ทำหน้าที่จับกินสิ่งแปลกปลอมแบบไม่จำเพาะ [natural killer (NK) cells] พบว่าการแบ่งตัวของลิมโฟซัยท์เพิ่มขึ้นอย่างมีนัยสำคัญทางสถิติที่ความเข้มข้นต่าง ๆ ขึ้นกับชนิดของสารสกัด นอกจากนี้พบว่าสารสกัดมะเกี๋ยงและแมงลักคาช่วยเพิ่มการทำงานของ NK cell อย่างมีนัยสำคัญ ในขณะที่สารสกัดของปัญจขันธ์ และแพรเซี่ยงไฮ้ ไม่ทำให้การทำงานของ NK cell เปลี่ยนแปลง จากการศึกษานี้แสดงว่าสารสกัดที่ทำการทดสอบนี้มีส่วนช่วยกระตุ้นการทำงานของลิมโฟซัยท์ในหลอดทดลองซึ่งอาจนำไปสู่การประยุกต์ใช้เพื่อปรับเปลี่ยนภูมิคุ้มกันของร่างกายได้

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Cleistocalyx nervosum var paniala is a plant in the Family Myrtaceae. High contents of polyphenols and flavonoids in *C. nervosum* were known to have antioxidant and anticarcinogenic properties (Leelapornpisit *et al*., 2004). It is now popular for functional health food, cosmetic ingredients and health drinks.

Gynostemma pentaphyllum Makino is a perennial climber in the Family Cucurbitaceae. It is used for treatment of inflammation, cough, hyperviscosity of sputum and chronic bronchitis (Jiang-Xu, 1979; Lin *et al*., 1993). Gypenosides, dammarane-type saponins (Piacente *et al*., 1995; Hu *et al*., 1996; Liu *et al*., 2005) isolated from *G. pentaphyllum*, are major bioactive principles which have been reported to have various *in vitro* activities such as reducing cholesterol (Kimura *et al*., 1983; Huang *et al*., 2005), anti-tumor (Chen *et al*., 1999; Zhou *et al*., 2000; Wang *et al*., 2002; Chiu *et al*., 2003; Chen *et al*., 2004) anti-mutagenicity (Kulwat *et al*., 2005), anti-gastric ulcer (Rujjanawate *et al*., 2004), anti-thrombotic (Li and Jin, 1989; Tan *et al*., 1993), immunopotentiating (Zhang *et al*., 1990; Li and Xing, 1992) and anti-inflammatory (Lin *et al*., 1993) activities. Several lines of evidence have indicated its efficacy in experimental animals and patients, such as reducing levels of serum triglyceride and cholesterol in rats and quail (la Cour *et al*., 1995), hepatoprotection in rats (Lin *et al*., 1993; Lin *et al*., 2000), cardiovascular protection in anesthesized guinea pigs (Circosta *et al*., 2005), recovering leukocyte counts and lymphocyte proliferation in cancer patients with radiotherapy or chemotherapy and in irradiated mice (Hou *et al*., 1991; Chen *et al*., 1996) and increasing immune responses to ovalbumin in mice (Sun and Zheng, 2005).

Gynura procumbens (Lour.) Merr. is an annual evergreen shrub in the Family Compositae. It has been traditionally used as a topical anti-inflammatory and anti-allergy agent in Thailand (Jiratchariyakul *et al*., 2000). The ethanol extract of *G. procumbens* was reported to have anti-hyperglycaemic and anti-hyperlipidaemic activities in streptozotocin-induced diabetic rats (Zhang *et al*., 2000), and to possess anti-herpes simplex viral activity *in vitro* (Jiratchariyakul *et al*., 2000). The

ethyl acetate fraction of the ethanolic extract demonstrated anti-inflammatory in mice (Iskander *et al*., 2002). In addition, its aqueous extract could lower blood pressure in spontaneously hypertensive rats (Kim *et al*., 2006).

Houttuynia cordata Thunb is a plant in the Family Saururaceae and has been tradtionally used for treatment of dermatitis, urinary tract infection, and malaria (Bunyapaphatsara *et al*., 1999). *H. cordata* was reported to have various *in vitro* activities such as anti-cancer, anti-bacterial, anti-fungal, antiviral and anti-inflammatory activities (Chavalittumrong *et al*.,2003). Houttuynin sodium bisulphate, a mixture of sodium bisulphate and houttuynin, was used for the treatment of bovine mastitis (Hu *et al*., 1997). Sodium houttuyfonate, a mixture of sodium bisulfite and houttuynin, was demonstrated to have effects on antibody production, macrophage activities, lymphocyte proliferation and IL-2 secretion in mice (Wang *et al*., 2002).

Hyptis suaveolens (L.) Poit is a strong aromatic and mosquito-repellent plant in the Family Lamiaceae (Smitinand, 2001; Seyoum *et al*., 2002). It is used as carminative, antiseptic, sudorific and galactogogue agents (Saluja and Santani, 1993). The essential oil of *H. suaveolens* inhibited the growth of Gram-positive and Gram-negative bacteria and had a mild inhibitory effect on *C. albicans* and *Aspergillus nigers* (Iwu *et al*., 1990). Methanol extract of *H. suaveolens* inhibited the growth of *Candida albicans*, selected Gram-positive and Gram-negative bacteria (Rojas *et al*., 1992). The ethanolic extract of its leaves showed wound healing activity (Shirwaikar *et al*., 2003). Leaf powder of *H. suaveolens* could inhibit aflatoxin B production (Krishnamurthy and Shashikala, 2006). Suaveolol and methyl suaveolate, isolated from leaves of *H. suaveolens*, possessed anti-inflammatory activities (Grassi *et al*., 2006).

Portulaca grandiflora Hook. is a succulent plant in the Family Portulacaceae (Backer & Bakhuizen Van Den Brink, 1963; Liu & Chen, 1976). In oriental traditional medicine, *P. grandiflora* is used for the relief of sore throat, skin rash and detoxification. *P. grandiflora* was reported as an effective anti-HBsAg herb by ELISA technique for the recognition of anti-HBsAg capability (Zheng & Zhang, 1990). In addition, antimutagenic effect of *P. grandiflora* on the mutation induced by aflatoxin B1 and cyclophosphamide in mice was demonstrated (Liu *et al*., 1990).

Phytolacca americana L. is a plant in the Family Phytolaccaceae. Ethanolic extraction of its fresh roots is used as an emetic (Smitinand, 2001). Phytolacca mitogens, derived from the ethanolic extract of *P. americana* roots, were found to have a stimulating effect on murine B and T lymphocytes (Yokoyama *et al*., 1976). Saponin extracts from *P. americana* demonstrated antifungal (Shao *et al*., 1999) and anti-viral activities (Uckan *et al*., 2005).

Tradescantia spathacea Kerr. is a succulent herb in the Family Commelinaceae. In Thai folk medicine, it is used to relieve fever, cough and bronchitis. It was reported to possess anti-microbial, insecticidal, anti-inflammatory, anti-cancer and anti-fertility activities (Bunyapaphatsara *et al*., 2000).

A large number of plants used in traditional medicines have been shown to possess non-specific stimulating activities on humoral and cell-mediated immune responses (Azuma, 1987). At present, crude or partial extracts of medicinal plants are widely consumed because they are believed to strengthen human health by boosting the immune system, but with limited scientific information. The present study was, therefore, aimed to screen and elucidate *in vitro* effects of the above Thai medicinal plants on human lymphocyte proliferation and functions of natural killer (NK) cells.

Materials and methods

Plant material

Plants investigated in this study are summarized in Table 1. These plants were identified and confirmed by comparing with voucher specimens of known identities in the Forest Herbarium (RFD and BKF), Royal Forest Department, and the Bangkok Herbarium (BK), Department of Agriculture, Bangkok, Thailand.

Table 1. List of plants investigated

Name	Origin of collection	Plant parts/solvent	Voucher specimen number
Cleistocalyx nervosum var paniala	Chiang Mai	fruit/water	BK 3013
Gynostemma pentaphyllum Makino.	Chiang Rai	aerial part/water	BKF 135338
Gynura procumbens (Lour.) Merr.	Bangkok	leaf/water	BKF 40176
Houttuynia cordata Thunb.	Pittsanulok	aerial part/water	BKF 37022
Hyptis suaveolens (L.) Poit.	Chantaburi	aerial part/water	BKF 082711
Portulaca grandiflora Hook.	Nonthaburi	aerial part/water	RFD 99057
Phytolacca americana L.	Pathumthani	root, leaf, fruit, stem/ 50% ethanol	BK 999
Tradescantia spathacea Kerr.	Nonthaburi	leaf/water	BK 15143

Extraction

Fresh roots (405 grams), leaves (396 grams), fruits (988 grams) and stems (145 grams) of *P. americana* were extracted with 50% ethanol using a reflux method for 2 hours. The supernatants were dried under vacuum in a rotary evaporator. The yields of dried extracts obtained from roots, leaves, fruits and stems were 7.21,8.60,10.84 and 7.96 % w/w, respectively.

Dried *C. nervosum* fruits (40 grams), *G. pentaphyllum* (15 grams), *H. suaveolens* (100 grams) and *P. grandiflora* (15 grams) aerial parts were ground and extracted with distilled water using a reflux method for 2 hours. Filtrates were collected and dried under vacuum in a rotary evaporator. The yields obtained were 8.5, 20, 13, and 20.93% w/w for *C. nervosum, G. pentaphyllum, H. suaveolens* and *P. grandiflora,* respectively.

Fresh leaves of *G. procumbens* (100 grams) and *T. spathacea* (200 grams) were blended with water and filtrates were dried under vacuum in a rotary evaporator. The yields of the dried extracts obtained from *G. procumbens* and *T. spathacea* were 4.19 and 3.42 % w/w, respectively.

The fresh aerial part of *H. cordata* (580 grams) was blended with water and its filtrate was lyophilized to give a residue (yield: 2.91% w/w).

All of the dried extracts were kept in a refrigerator until use. Each extract was dissolved in sterile distilled water for an hour prior to the experiments. It was noted that all extracts, except that of *C. nervosum*, contained very minute amount of non-dissolved residues after 1-hour dissolution.

Subjects

Fresh heparinized blood from healthy Thai donors of the National Blood Bank, the Thai Red Cross Society were collected with permission. Their ages ranged from 20-50 years. None had a history of hepatitis B infection or had a risk for HIV-1 exposure.

Preparation of mononuclear cells

Mononuclear cells were separated from heparinized blood using Ficoll-Hypaque density gradient (Boyum, 1966). The mononuclear cells were counted and adjusted to an appropriate concentration in complete RPMI 1640 (RPMI 1640 medium supplemented with 2 mM glutamine, 10 mM HEPES, 100 U/ml penicillin G, and 100 µg/ml streptomycin) containing 10% fetal bovine serum (FBS; Grand Island Biological Company, Grand Island, NY, USA) for further assays.

Lymphoproliferation assay

Lymphocyte proliferative response to the extract was performed as described (Sriwanthana and Chavalittumrong, 2001). Briefly, purified mononuclear cells ($2x10^6$ cells/ml) were cultured in triplicate in 96-well microtiter plates (Costar, Cambridge, MA, USA) with the extracts at final

concentrations of 1 ng/ml, 10 ng/ml, 100 ng/ml, 1 µg/ml, 5 µg/ml, 10 µg/ml and 100 µg/ml in complete RPMI 1640 containing 10% FBS. The cultures were incubated at 37°C with 5% CO_2 for 72 hours. Lymphocyte proliferation was determined by uptake of ^{3}H-thymidine at 18 hours before harvesting. The radioactivity was measured by a liquid scintillation counter (Topcount Microplate Scintillation & Luminescence Counter, Packard Instrumental Co., CT, USA). The degree of activation was expressed as a stimulation index [S.I., i.e., the ratio of the ^{3}H-thymidine uptake in count per minute (CPM) of samples with extract to those without extract]. Phytohemagglutinin (Sigma, St. Louis, MO, USA) at 10 µg/ml was also added to the culture system to check for cell survival and used as a positive control of each assay.

Natural Killer (NK) cell activity assay

Peripheral blood mononuclear cells (PBMC) were washed, resuspended and adjusted to $2x10^6$ cells/ml in complete RPMI 1640 containing 10% FBS. PBMC were incubated in the absence or presence of the extracts at the final concentrations of 10 ng/ml, 100 ng/ml, 1 µg/ml, 10 µg/ml and 100 µg/ml at 37°C for 18 hours. After incubation, the cultures were washed and then used as effector cells for the assay of NK cell activity.

K 562 cells were used as target cells and were grown in complete RPMI 1640 containing 10% FBS. The target cells ($2x10^6$ cells) were labeled with 100 µCi of $Na_2{}^{51}CrO_4$ (specific activity 37.0 MBq/µg: Amersham, Buckinghamshire, UK) at 37°C 5% CO_2 for 60 minutes, and washed 3 times with cold RPMI 1640 containing 10% FBS.

The cytotoxicity assay was performed as described (Sriwanthana and Chavalittumrong, 2001). In brief, $2x10^3$ target cells/well and a PBMC effector-to-target cell ratios (E:T) of 90:1, 30:1, 10:1, and 3:1 were set up in triplicate in 96-well round-bottom microtiter plates (Corning Incorporated, Corning, NY, USA). The plates were incubated for four hours at 37°C with 5% CO_2. After incubation, supernatants from each well (100 µl) were transferred into tubes and counted in a Gamma counter (Cobra Series Gamma Counter Systems, Packard Instrumental Co., CT, USA). The percentage of cytolysis was calculated according to the following formula: %cytolysis = (experimental release - spontaneous release) / (maximal release - spontaneous release). Spontaneous release was measured by incubation of target cells with medium alone, while maximal release was measured by lysis of target cells with 5% Triton X-100. NK cell activity was expressed as lytic units (LU)/10^7 PBMCs as determined by least squares analysis derived from the percentage of specific lysis of all E:T ratios. One LU was defined as the number of effector cells required for 20% specific lysis of $1x10^4$ target cells.

Statistical analysis

Data were expressed as mean±SD. The significance of the results was calculated using Student's paired t-test and statistical significance was defined as $P<0.05$.

Results

Lymphocyte proliferation

The extract-induced proliferative responses were performed by culturing human lymphocytes in the presence or absence of each extract. It was shown that the responses were significantly elevated with the *H. cordata* and *P. grandiflora* extracts ranging from concentrations of 1 ng/ml to 100 µg/ml (Tables 2, 3). Our study showed that the water extract of *G. pentaphyllum* and *G. procumbens* significantly increased lymphocyte proliferation at the concentrations of 1 µg/ml to 100 µg/ml (Table 2). Tables 2 and 3 also demonstrate various patterns in stimulating effects of *C. nervosum, H. suaveolens* and *T. spathacea* on the proliferative responses of human lymphocytes.

P. americana roots significantly enhanced lymphocyte proliferation at the concentrations of 1 µg/ml to 100 µg/ml, while other parts of *P. americana* significantly stimulated or decreased lymphoproliferative responses at different concentrations as shown in Table 4.

Table 2. Effect on lymphocyte proliferation of normal PBMC

Concentration	*C. nervosum* (n = 20)	*G. pentaphyllum* (n = 14)	*G. procumbens* (n = 10)	*H. cordata* (n = 20)
control	1.00±0.00	1.00±0.00	1.00±0.00	1.00±0.00
1 ng/ml	1.87±0.56*	1.15±0.32	1.55±0.78	1.70±0.44*
10 ng/ml	1.43±0.40*	1.19±0.37	1.18±0.36	1.64±0.55*
100 ng/ml	0.91±0.32	1.19±0.33	1.13±0.25	1.12±0.23*
1 µg/ml	1.12±0.33	1.36±0.44*	1.34±0.34*	1.13±0.22*
5 µg/ml	2.37±0.82*	1.69±0.55*	1.75±0.47*	2.07±0.64*
10 µg/ml	2.32±1.00*	1.56±0.46*	1.75±0.46*	2.27±0.76*
100 µg/ml	1.47±0.72*	1.80±0.66*	2.03±0.96*	1.58±0.55*
PHA	368.63±141.87*	116.82±69.01*	326.96±213.30*	365.90±154.96*

Each value represents Mean±SD
***p< 0.05**

Table 3. Effect on lymphocyte proliferation of normal PBMC

Concentration	*H. suaveolens* (n = 20)	*P. grandiflora* (n = 60)	*T. spathacea* (n = 24)
control	1.00±0.00	1.00±0.00	1.00±0.00
1 ng/ml	1.20±0.24*	1.41±0.11*	1.82±1.55*
10 ng/ml	1.04±0.21	1.41±0.10*	1.41±0.27*
100 ng/ml	0.98±0.21	1.88±0.11*	0.95±0.19
1 µg/ml	1.26±0.38*	2.26±0.12*	1.06±0.19
5 µg/ml	1.56±0.49*	2.59±0.17*	1.70±0.40*
10 µg/ml	1.42±0.43*	2.64±0.19*	1.53±0.29*
100 µg/ml	1.28±0.39*	2.86±0.23*	1.16±0.29*
PHA	106.61±50.48*	120.17±14.10*	335.80±92.60*

Each value represents Mean±SD
***p< 0.05**

Table 4. Effect on lymphocyte proliferation of normal PBMC

Concentration	*P. americana* (root) (n = 18)	*P. americana* (leaf) (n = 18)	*P. americana* (fruit) (n = 18)	*P. americana* (stem) (n = 18)
control	1.00±0.00	1.00±0.00	1.00±0.00	1.00±0.00
1 ng/ml	1.06±0.23	1.22±0.39*	1.05±0.22	1.17±0.27*
10 ng/ml	0.97±0.25	0.98±0.22	0.94±0.20	1.00±0.24
100 ng/ml	0.94±0.25	0.85±0.21*	0.79±0.22*	0.92±0.27
1 µg/ml	1.19±0.37*	1.08±0.29	1.06±0.37	1.05±0.22
5 µg/ml	1.68±0.67*	1.28±0.37*	1.25±0.5*	1.41±0.36*
10 µg/ml	1.74±0.67*	1.21±0.35*	1.27±0.65	1.15±0.39
100 µg/ml	2.64±1.12*	1.53±0.52*	1.86±0.95*	1.56±0.76*
PHA	155.73±108.07*	167.59±118.86*	183.52±134.50*	156.74±100.85*

Each value represents Mean±SD
***p< 0.05**

Table 5. NK cell activity by normal PBMC

Concentration	*C. nervosum* (n = 20)	*G. pentaphyllum* (n = 12)	*H. suaveolens* (n = 12)	*P. grandiflora* (n = 12)
control	70.65±39.21	46.12±36.46	46.58±26.09	53.64±34.07
10 ng/ml	72.00±35.94	42.35±29.81	49.11±32.37	55.28±45.07
100 ng/ml	70.53±34.12	43.66±29.41	50.55±26.77	57.54±44.31
1 µg/ml	72.56±36.20	44.51±34.12	50.01±30.14	57.25±42.03
10 µg/ml	85.52±37.55*	49.10±35.61	56.94±30.95*	52.76±31.82
100 µg/ml	115.97±51.86*	42.25±31.69	54.35±27.68*	53.04±40.07
PHA	140.19±36.01*	105.66±48.55*	92.66±22.22*	97.14±27.99*

Each value represents Mean±SD
***p< 0.05**

NK cell activity

Results from other pharmacological studies performed in our institutes suggested that *C. nervosum , G. pentaphyllum, H. suaveolens* and *P. grandiflora* had high potential for further development as medicines. We, then, examined the effect of these plants on NK cell activity. No significant changes in NK cell activity, as expressed in LU/10^7 PBMC, were observed in the presence of the water extracts of *G. pentaphyllum* or *P. grandiflora* (Table 5). Both *C. nervosum* and *H. suaveolens* significantly enhanced NK cell activity, as expressed in LU/10^7 PBMC, at the concentrations of 10 µg/ml to 100 µg/ml, respectively (Table 5).

The activities, expressed as % lysis, of each extract are illustrated in Figures 1A -1D. Each of the extract significantly altered the function of NK cells at different E:T ratios.

Discussion

A number of herbal medicines and products are claimed to modify or boost immunity without scientific support. In addition, plants cultivated in different parts of the world may not have pharmacological activities as previously reported (Bauer, 2000). Therefore, attempts were made to verify 8 kinds of Thai medicinal plants for *in vitro* stimulating human lymphocyte activity.

Several lines of evidence have indicated non-specific immunostimulating or immunomodulating activities of a large number of medicinal plants in experimental animals. Those *in vivo* studies are cumbersome for initial screening of such activities. Using human lymphocytes in our studies, it was shown that the extracts of *G. pentaphyllum, H. cordata* and *P. americana* possess immunostimulating activities. These findings parallel reports from studies in mice (Li and Xing, 1992; Wang *et al*., 2002; Yokoyama *et al*., 1976; Zhang *et al*., 1990), suggesting that human lymphocytes could be used to determine effects of medicinal plants on the immune system. In addition, it may possibly indicate that *G. pentaphyllum, H. cordata* and *P. americana* cultivated in Thailand also possess immunostimulating activity. Furthermore, enhanced lymphocyte proliferation was demonstrated with the extracts of *G. procumbens* and *P. grandiflora*, suggesting possible roles of *G. procumbens* and *P. grandiflora* as putative immunostimulants.

The increase in lymphocyte stimulation, as quantified by stimulation index (S.I.), was also found in cultures containing the extracts of *C. nervosum, H. suaveolens* and *T. spathacea*. Significant alteration induced by these extracts, was not dose-dependent. This may possibly be due to variation in chemical constituents responsible for plant activities.

We assessed the effects of *C. nervosum , G. pentaphyllum, H. suaveolens* and *P. grandiflora* on NK cell activity, one of natural defense

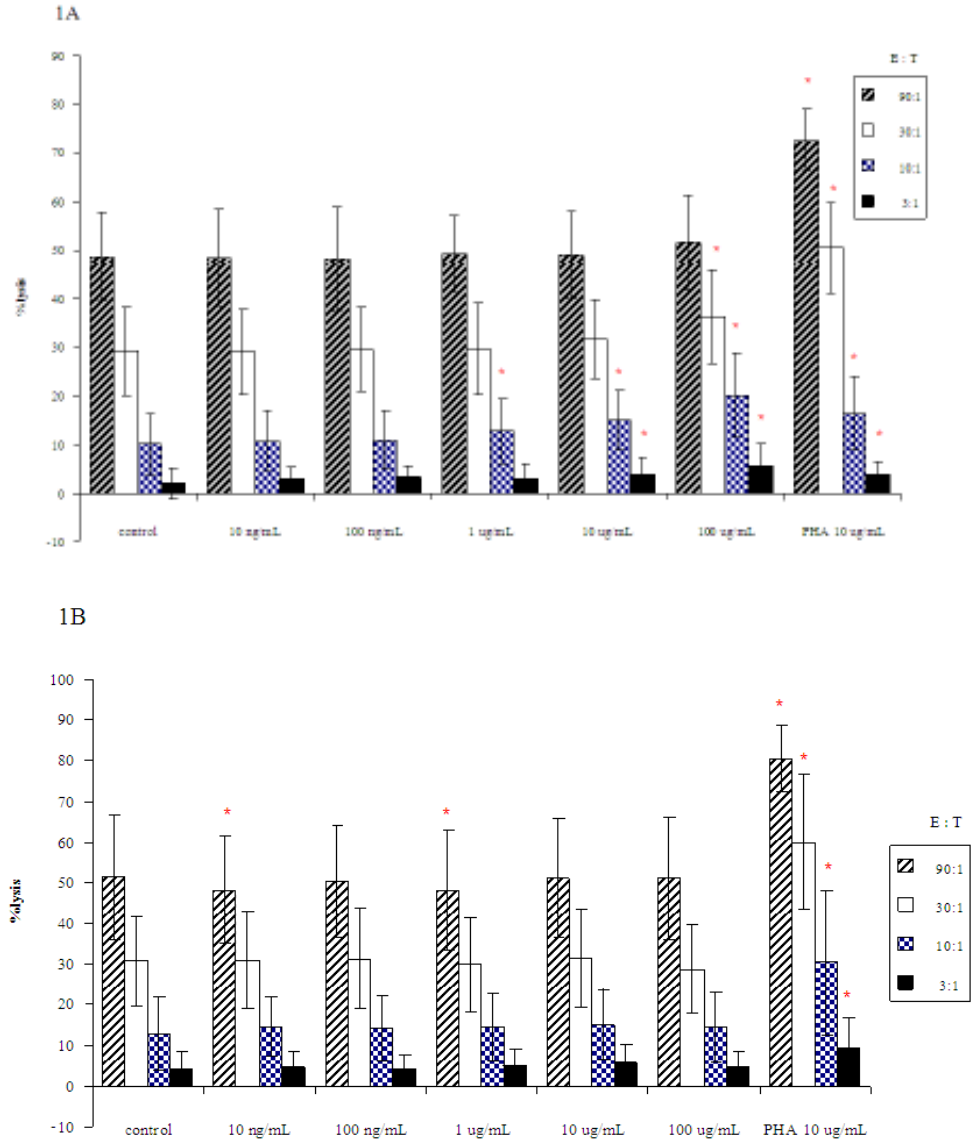

Figure 1. ***In vitro* effects of *C. nervosum* (1A), *G. pentaphyllum* (1B), *H. suaveolens* (1C) and *P. grandifora* (1D) on NK activity. PBMC were cultured in the presence and absence of different concentrations of each extract for 18 hrs. NK cell activity was determined by culturing stimulated PBMC with ^{51}Cr-K562 cells in triplicate at different E:T ratios for 4 hrs at 37^{o}C. Supernatants were counted for radioactivity and % lysis calculated. Each bar represents mean±SD. *p< 0.05**

mechanisms against a variety of infections and cancers (Kuby, 1997). There is no definite rule in reporting NK cell activities. The activities, reported as LU/10^{7}PBMC, apparently demonstrated the effects of each extract as compared with the ones reported in % lysis. Calculation as % lysis may not be comparable with other studies because of differences in E:T ratios used in each study. We, therefore, used activities in LU/10^{7} PBMC in order to assess the effects of each extract on NK cell

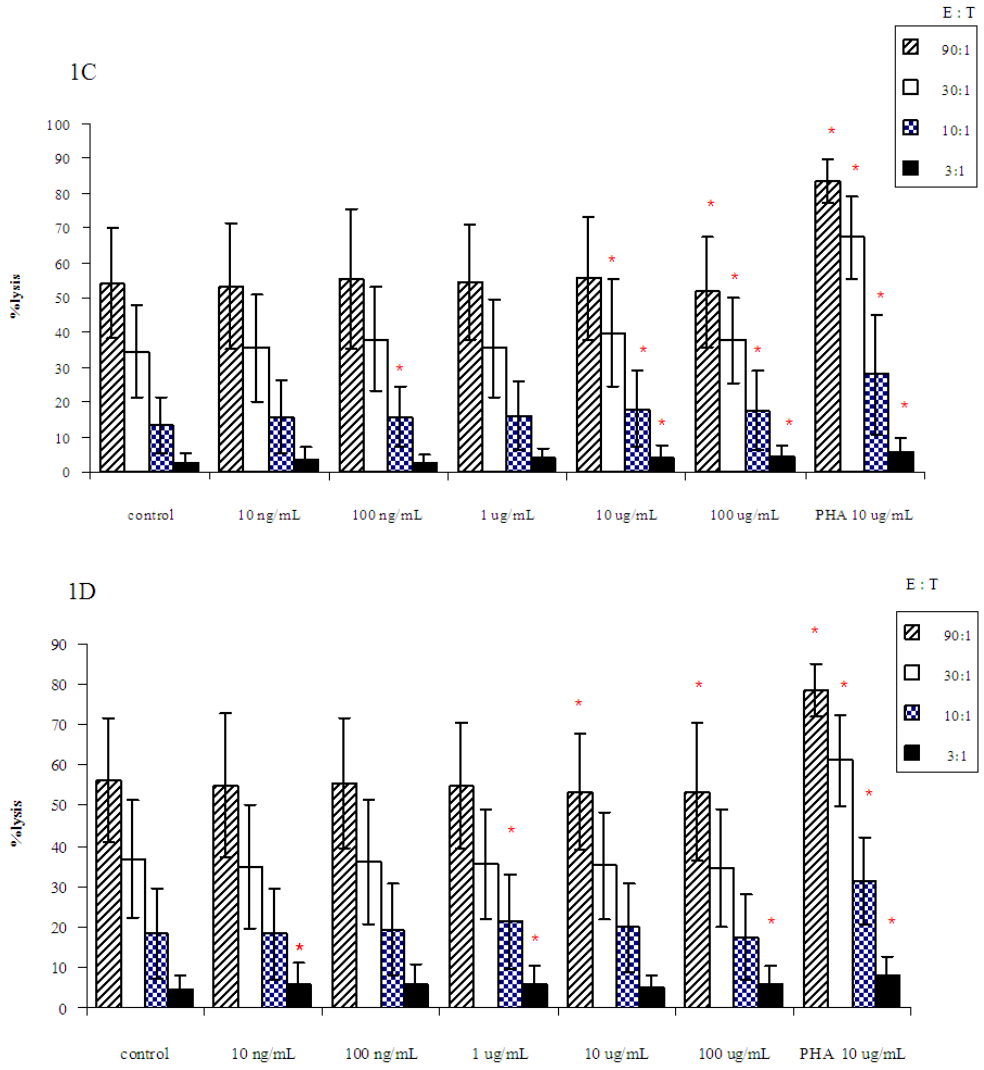

Figure 1. (continued)

function. Our studies demonstrated that the extracts of *C. nervosum* and *H. suaveolens* stimulated NK cell activity. Different immunomodulating profiles may provide a rational basis and support for plant selection aimed at drug discovery.

Our data revealed that those plant extracts show promising lymphocyte stimulating activities on non-specific cell-mediated immune responses. Purification of active components may be required for potential and clinical applications as immunomodulators.

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