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Anticancer Effects of Some Medicinal Thai Plants

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Abstract

Ethanollic extracts from thirty Thai edible plants collected from Sa Keao province, Thailand, were screened for *in vitro* antiproliferative effect on HCT-116 human colon cancer cell line using cell titer 96 aqueous one solution cell proliferation assay. It was found that leaf extract of *Crateva adansnii*, fruit and leaf extracts of *Ardisia elliptica*, shoot extract of *Colocasia esculenta*, leaf extract of *Cratoxylum fomosum*, and leaf extract of *Millettia leucantha* exhibited antiproliferative activities. The fruit extract of *Ardisia elliptica* showed the highest antiproliferative activity. Ethanollic extract of the stems from *C. fenestratum* and its dichloromethane and aqueous fractions showed antiproliferative activity to human colorectal cancer cells (HCT-116) determined by cell growth assay. Berberine, one of the major alkaloid in the stems of *C. fenestratum*, also promoted antiproliferative effect. Extracts from the leaves of three *Azadirachta* species in Thailand, *A. indica*, *A. indica* var. *siamensis*, and *A. excelsa*, were reported to promote *in vitro* antioxidant effects determined by various methods. Ten *Russula* mushroom collected from northeastern part of Thailand were tested for *in vitro* antioxidant activities using photochemiluminescence assay for both lipid-soluble and water-soluble antioxidant capacities. *R. medullata* extract exhibited the highest antioxidant effects in both lipid-soluble and water-soluble models.

Keywords: anticancer, *Coscinium fenestratum*, berberine, *Azadirachta*, *Russula*

1. Introduction

Cancer cells uncontrollably divide to form masses of tissue, which are called tumors. Tumors can grow and interfere with the functions of many bodily systems including the digestive, nervous, and cardiovascular systems. Cancer has been reported to be the first in the rank of causes of the death in the Thai population. Liver, colon, and lung cancers are the most prevalent cancers in Thai males, while breast, cervical, and colon cancers are the most prevalent cancers in Thai females [1].

The development of cancer or carcinogenesis occurs through a multistep process involving the mutation, selection of cells with a progressive increasing capacity for proliferation, survival, invasion, and metastasis [2]. The first step in the process, tumor initiation, relates to the genetic alteration leading to the changes in normal cells. Then, in the promotion or development stage, the cells abnormally proliferate leading to the outgrowth of a population of clonally derived tumor cells [2]. This stage can be stimulated by carcinogens, which are a group of substances such as tobacco, asbestos, arsenic, radiation such as gamma and X-rays, sun light, polycyclic hydrocarbons, nitrosamines, and aflatoxins: these substances do not directly cause cancers but promote or aid the development of cancers [2, 3]. After that, tumor progression continues as additional mutations occur within the cells of the tumor population to further advantage the cancer cells, such as more rapid growth, which will allow them to become dominant within the late tumor population. The process is called clonal selection, since a new clone of tumor cells evolves on the basis of its increased growth rate or other properties such as survival, invasion, or metastasis. Clonal selection continues throughout tumor development, so tumors continuously become more rapid-growing and increasingly malignant [2].

2. Cancer therapy

The modern treatments for cancers mainly are surgery, radiation, and chemotherapy. However, most of chemotherapeutic drugs are not specific to only cancer cells, but also cause damage to normal cells, especially bone marrow, mucous glands, mucous membranes, hair, and nails and can lead to the suppression of the immune system [3]. The success of chemotherapy depends on the number of cancer cells, the proliferation rate, the duration of the drug administration, and the therapeutic interval. To avoid drug resistance, polychemotherapy is always used instead of monochemotherapy [3]. The anticancer drugs can also cause some other side effects including nausea, vomiting, agranulocytosis, inhibition of spermatogenesis and ovulation, alopecia, inflammation of mucous membranes, and teratogenesis [3].

Some compounds separated from natural products are now being developed as modern medicines for the treatments of cancers including paclitaxel, catharanthus alkaloids, and derivatives of podophyllotoxin.

Paclitaxel was separated from the bark of *Taxus brevifolia* Nutt. (Pacific Yew), which is a tree in Taxaceae. Paclitaxel will bind with β -tubulin and stimulate the aggregation of a tubulin subunit to become a nonphysiological microtubule composed of 12 proto-filaments, which cause the inhibition of cell cycles in mitosis and interphase (G_2 -phase) and lead to cell apoptosis. This compound is normally used in an injection formulation as the adjuvant chemotherapy for the treatments of ovarian, breast, and bronchial cancers [3].

Some alkaloids are separated from the leaves of *Catharanthus roseus* (L.) G. Don., such as vincristine and vinblastine. Vincristine is used for the treatment of lymphatic leukemia, neuroblastoma, and Wilms tumor, while vinblastine is used to treat lymphogranuloma (Morbus Hodgkin), lymphosarcoma, testicular carcinoma, and chorionic carcinoma [3].

Podophyllotoxin was separated from the rhizome of *Podophyllum peltatum* L. or American mandrake. Two derivatives of podophyllotoxin, etoposide and teniposide, are now being developed and used as anticancer drugs. Etoposide is used for the treatment of bronchial cancer, testicular carcinoma, and chorionic carcinoma, while teniposide is used to treat brain or bladder cancers [3]. The chemical structures of some anticancer compounds from natural products are shown in **Figure 1**.

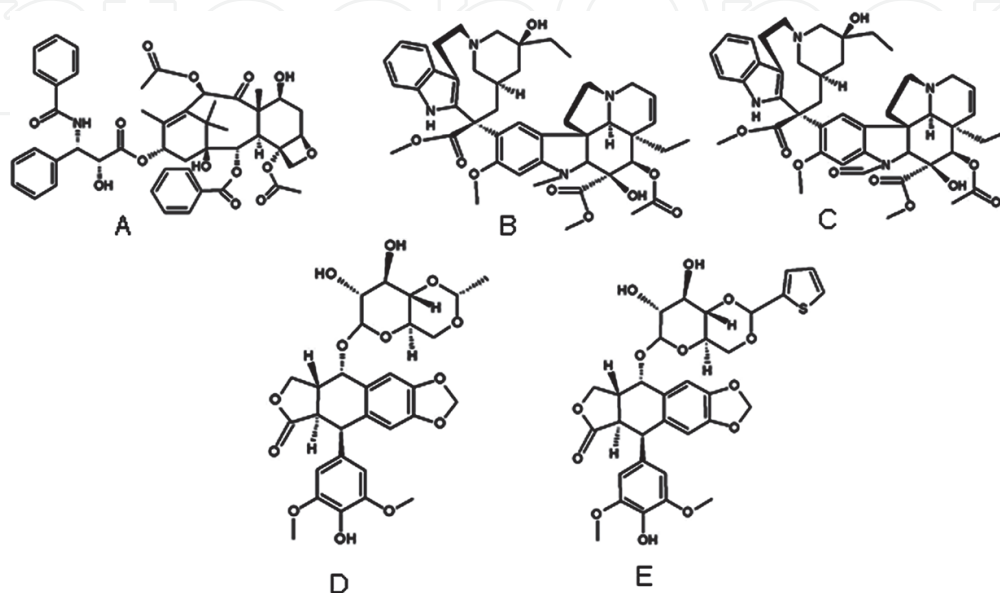


Figure 1. Chemical structures of some anticancer compounds from natural products. A = paclitaxel, B = vinblastine, C = vincristine, D = etoposide, E = teniposide.

3. Anticancer effects of medicinal plants and natural products

Natural products from plants, animals, marine sources, and minerals have been used for the treatments of ailments and diseases for a long time. In Thai traditional medicine, the word “cancer” could refer to the symptom of chronic wound, abscess, emaciation, and weak [4]. Active phytochemicals in plants can be classified into two main groups of primary metabolites, which are the compounds necessary for plant growth and development such as carbohydrates, proteins, and fats. Another group is secondary metabolites, which promote the defense mechanisms or support the lives of the plants; they include polyphenolic compounds, flavonoids, terpenoids, and alkaloids [5]. Ethanolic extracts from thirty Thai local edible plants collected from Wang Nam Yen district, Sa Keao province, Thailand were screened for the *in vitro* anti-proliferative effect on HCT-116 human colon cancer cell lines using a cell titer 96 aqueous one solution cell proliferation assay. It was found that six ethanolic plant extracts, including a leaf extract of *Crateva adansnii*, fruit and leaf extracts of *Ardisia elliptica*, a shoot extract of *Colocasia esculenta*, a leaf extract of *Cratoxylum fomosum*, and a leaf extract of *Millettia leucantha* exhibited antiproliferative activities on the HCT-116 cell line. The fruit extract of *Ardisia elliptica* showed the highest antiproliferative activities with an IC_{50} value of 5.12 ± 0.54 $\mu\text{g/ml}$ [6]. The mechanisms of the action of medicinal plants for anticancer effects have been reported as following [4]:

3.1. Inhibition of cell division in the cancer cell cycle

Alpha-mangostin from mangosteen (*Garcinia mangostana*) fruit rind promoted inhibitory effects to breast cancer cell line (MDA-MB-231) by inhibition of cell division in G1 and S phases [7]. Methanol extract of *Morus alba* L. leaves inhibited liver cancer cell line Hep G2 by inhibition of cell division in G2/M phase [8]. Cucurbitacin B, a triterpenoid from *Trichosanthes cucumerina* L., also inhibited breast cancer cell division in G2/M phase [9].

3.2. Induction of cancer cell apoptosis

This mechanism includes some minor mechanisms which stimulate anticancer genes, induction of caspase enzymes, induction of free radical formation, inhibition or induction of enzymes relating to histone protein, and the formation of spingosine or ceramide [4]. Dehydrocostus lactone from the root of *Saussurea lappa* induced the apoptosis of liver cancer cells Hep G2 and PLC/PRF/5 via p53 protein [10]. Water extract of the seed from *Sapindus rarak* Candolle. induced lung cancer cells A549 apoptosis through the induction of the caspase enzyme [11], while methanol extract of *Derris scandens* Benth. induced apoptosis of colon cancer cells SE480 by increased caspase-3 activity and down-regulated Bcl-2 and up-regulated Bax protein of SW480 cells; it also significantly induced cell necrosis determined by the release of LDH [12]. Alpha-mangostin separated from the fruit rind of mangosteen also upregulated Bax and down-regulated Bcl-2 proteins in rat liver tissue [13]. Methanol extract from stem bark of *Myristica fragrans* Houtt. promoted the apoptosis of lymphoblast Jurkat by controlling the SIRT1 gene [14]. G1 b, a glycosphingolipid from *Murdannia loriformis* (Hassk.) R.S.Rao & Kammathy, inhibited breast, lung, colon, and liver cell lines [15].

3.3. Immune stimulation

Methanol extract from the leaves of *Moringa oleifera* Lam. exhibited immune stimulation effect both cell-mediated immunity and humoral immunity by induction of neutrophil production and stimulation of macrophages in animals damaged by the toxicity of anticancer drugs [16].

In Thai traditional medicine, there are some medicinal formulas composed of several plants in different ratios. These formulas are traditionally used for a long time usually for the treatment of cancers in patients with late stage cancers, patients who cannot improve after treatment with chemotherapy, radiation or surgery, patients with cancers in several organs or patients with incurable diseases [4]. The sources of anticancer herbal formulas usually come from local traditional doctors or priests in the temples (in Thai, temple is called as "Wat"), with the normal method of preparation being the decoction of plant materials with water [4]. A herbal remedy from Wat Tha-it (Tha-it temple), Ang Thong province, Thailand, composed of several plant materials including *Gelonium multiflorum* A. Juss., *Erycibe elliptimba* Merrill & Chun, *Balanophora abbreviate* Blume, *Smilax china* L., *Smilax glabra* Wall. ex Roxb., and *Millingtonia hortensis* Linn. was reported to significantly promote synergistic effects on doxorubicin in the treatment of A549 cancer cells by the inhibition of cell divisions in the G2/M phase [4, 17]. Another herbal remedy is from a Thai herbal nursing home, Wat Khampramong, Sakon Nakhon province, comprises of several plant materials such as *Rhinacanthus nasutus* (L.) Kurz, *Acanthus ebracteatus* Wall., *Smilax glabra* Wall. ex Roxb.,

Artemisia annua L., *Angelica sinensis* (Oliv.) Diels, *Salacia chinensis* L., and *Orthosiphon aristatus* Miq [18]. This herbal remedy can inhibit the growth of some cancer cell lines such as breast adenocarcinoma MDA-MB 231, synovial sarcoma SW982, hepatocellular carcinoma HepG2, cervical adenocarcinoma HeLa, and lung carcinoma A549 [18].

4. Some potential Thai medicinal plants with anticancer effects

4.1. *Coscinium fenestratum* (Gaertn.) Colebr

NAG-1 or nonsteroidal anti-inflammatory drug (NSAID)-activated gene was identified in COX-negative cells by PCR-based subtractive hybridization from an NSAID-induced library as a divergent member of the TGF- β superfamily [19]. The overexpression of NAG-1 in cancer cells results in growth arrest and an increase in apoptosis, suggesting that NAG-1 has antitumorigenic activity [20]. NAG-1 expression is also upregulated by a number of dietary compounds, medicinal plants, and anticancer drugs [21–25]. *Coscinium fenestratum* is one of the medicinal plants that promoted antiproliferative effects on colon cancer cell lines with mechanisms related to NAG-1 [20].

Coscinium fenestratum (Gaertn.) Colebr. is a large climber with yellow wood and sap, known in the Thai language as Hamm or Khamin khrua. The genus *Coscinium* belongs to the tribe Coscinieae of the family Menispermaceae. This genus comprises two species, which are *Coscinium blumeanum* Miers. and *C. fenestratum* (Gaertn.) Colebr. Both of them are stout woody climbers growing in the tropical rain forest regions of Asia [26]. *Coscinium* species are characterized by the axillary flowers, extra-axillary or cauliflorous in racemiform, or peduncled subumbellate aggregate, of 20–50 cm in length. The inflorescences are axillary or cauliflorous with 6–12 florets. Male flowers are sessile or with pedicels, up to 1 mm. Sepals are broadly elliptic to obovate with the inner 3–6 spreading, yellow, and 1.5–2 mm long. Stamens are 6 with 1 mm long. The Sepals of female flower are as in male flowers. Staminodes are 6 and claviform with 1 mm long. Drupes are subglobose, tomentellous, brown to orange or yellowish, 2.8–3 cm diameter. Pericarp is drying woody. Seeds are whitish and subglobose with the enveloping condyle. The leaves are subpeltate or ovate, large, hard-coriaceous, palmately nerved, reticulate, and densely hairy beneath [26]. Physical characteristic of the *Coscinium fenestratum* stem (cross section) is shown in **Figure 2**.

The stem decoction and maceration extracts of *Coscinium fenestratum* have been traditionally used in the Northeastern part of Thailand for the treatment of various diseases such as cancer, diabetes mellitus, and arthritis [27]. The ethanolic extract of the stems from *C. fenestratum* and its dichloromethane and aqueous fractions showed antiproliferative activity on human colorectal cancer cells (HCT-116) determined by a cell growth assay. Berberine, one of the major alkaloids in the stems of *C. fenestratum*, also promoted an antiproliferative effect [20]. The mechanisms of action of the extracts from *C. fenestratum* were reported as the activation of proapoptotic proteins and ppar γ [20]. It was also reported that berberine facilitated the apoptosis of cancer cells, and the molecular targets for its activity are NAG-1 and AFT3 [24]. The chemical structure of Berberine is shown in **Figure 3**.

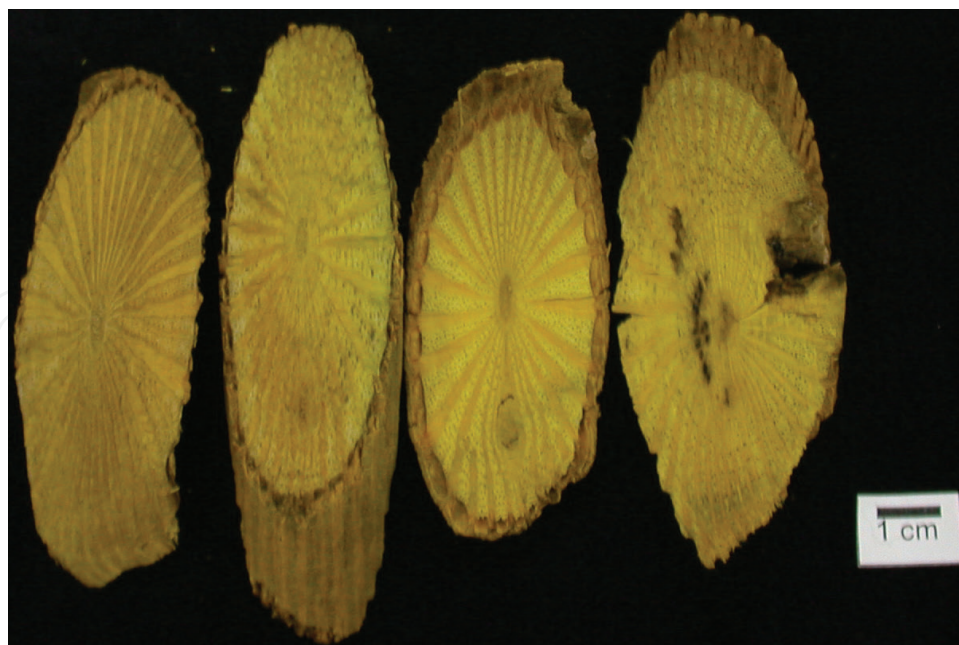


Figure 2. Physical characteristic of *Coscinium fenestratum* stem purchased from Nongkhai province, Thailand (cross section $\times 1$).

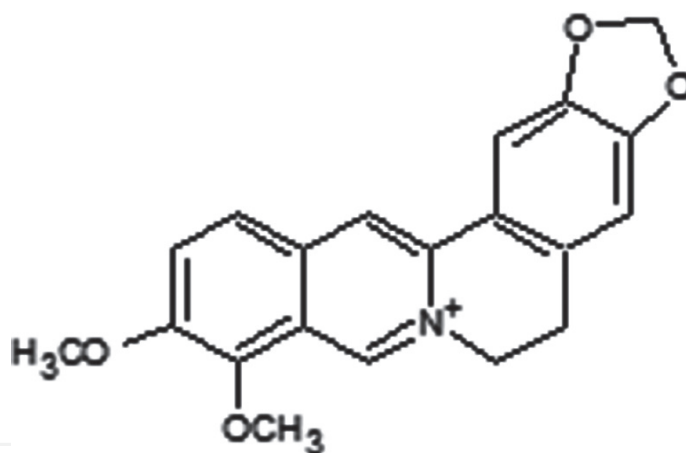


Figure 3. Chemical structure of Berberine.

4.2. *Azadirachta* plants

Oxidative stress is considered to be of some importance for many ailments and pathologies; including cardiovascular diseases, cancers, rheumatoid arthritis, and Alzheimer's disease [28]. Polyphenolic compounds have been reported to have important anticancer and chemopreventive effects [29]. Phenolic acids such as gallic acid, ellagic acid, and ferulic acid induce apoptosis in cancer cells, activated caspase, prevented cancer formation, and suppress the angiogenesis of cancer [29–32]. Flavonoids such as quercetin and kaempferol also promote apoptosis, inhibit oncogenes, and generated cell cycle arrest [29, 33–35].

Suttajit et al. [36] studied the antioxidant activities of extracts from many Thai medicinal plants using a ABTS-metmyoglobin assay and reported some plants with high antioxidant activities; including *Uncaria gambier* Roxb., *Piper betle* Linn., *Camellia sinensis* (L.) Kuntze., *Azadirachta indica* A. Juss. var. *siamensis* Valetton., *Curcuma zedoaria* Roxb., *Syzygium aromaticum* (L.) Merr. & Perry and *Tamarindus indica* Linn. When focusing on Thai medicinal plants, the Siamese neem tree (*Azadirachta indica* A. Juss. var. *siamensis* Valetton.) is an interesting plant that showed high antioxidant activity in the screening test [36, 37]. Moreover, there are reports about its antioxidant potential based on the antioxidant content as the butylated hydroxyanisole (BHA) equivalent of Thai indigenous vegetable extracts. From this report, the Siamese neem tree leaf extract appeared to be a high potency antioxidant, containing more than 100 mg BHA equivalent in 100 g fresh weight.

Azadirachta plants comprise of three different plant species; *Azadirachta indica* A. Juss or *A. indica* A. Juss var. *indica* (neem), *Azadirachta indica* A. Juss. var. *siamensis* Valetton (Siamese neem tree), and *Azadirachta excelsa* (Jack) Jacobs. (marrango tree). The Siamese neem tree leaves are wider, longer, and thicker than the leaves of neem, while the marrango tree has the widest, longest, and thickest leaves. The margin of the leaflet of Siamese neem tree is crenate to entire, while the margin of neem is serrate and that of marrango tree is entire to undulate. The colors of the leaflet blade of the Siamese neem tree, neem, and marrango tree are green, light green, and dark shiny green, respectively [38, 39]. The physical characteristics of Siamese neem tree, neem, and marrango tree leaves are shown in **Figure 4**.

The leaves and flowers of Siamese neem tree and neem have been traditionally used as element tonics and antipyretic and gastric secretion stimulating agents, while the stem bark of all *Azadirachta* plants is used to treat amoebic dysentery and diarrhea [40, 41]. There also reports suggesting that polysaccharides and limonoids found in neem bark, leaves, and seed oil reduce tumors and cancers and showed effectiveness against lymphocytic leukemia [42–44]. Moreover, the young leaves and flowers of the Siamese neem tree are popularly consumed as vegetables [39].

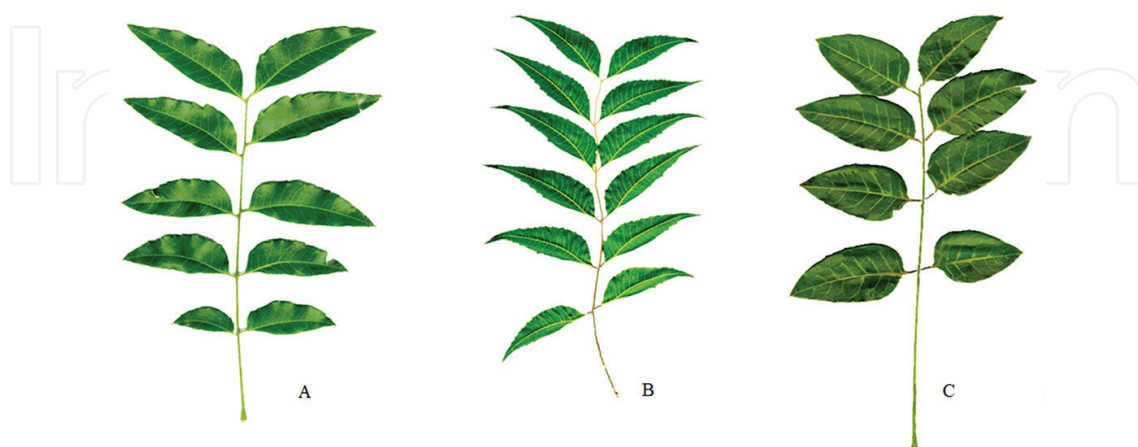


Figure 4. Physical characteristics of *Azadirachta* plants; A = Siamese neem tree (*Azadirachta indica* var. *siamensis*), B = neem (*Azadirachta indica*), C = marrango tree (*Azadirachta excelsa*).

For the antioxidant effect, *Azadirachta* plants were reported to promote *in vitro* activities tested by various methods. Extracts from the leaves of *A. indica*, *A. indica* var. *siamensis*, and *A. excelsa* were reported to promote *in vitro* antioxidant effects determined by a DPPH scavenging assay, Fremy's salt assay, ESR detection of POBN spin adducts, and an oxygen consumption assay [45, 46]. The leaf's aqueous and flower ethanol extracts from the Siamese neem tree provide antioxidant activity on lipid peroxidation formation induced by UV-irradiation of a Chago K-1 bronchogenic cell culture at a concentration of 100 $\mu\text{g/ml}$ determined by the thiobarbituric acid reactive substances (TBARS) method [47].

Cloning and expression analysis of genes involving flavonoid biosynthesis showed that Siamese neem tree leaves total RNA contained nucleotide sequences related to enzymes F3'H, FLS, DFR, and F3'5'H, which could be responsible for the biosynthesis of the antioxidant flavonoids [48]. Some flavonoids that were separated from Siamese neem tree and neem leaves and flowers are kaempferol, myricetin, quercetin, and rutin [39, 49–51]. The chemical structures of some flavonoids found in *Azadirachta* plants are shown in Figure 5.

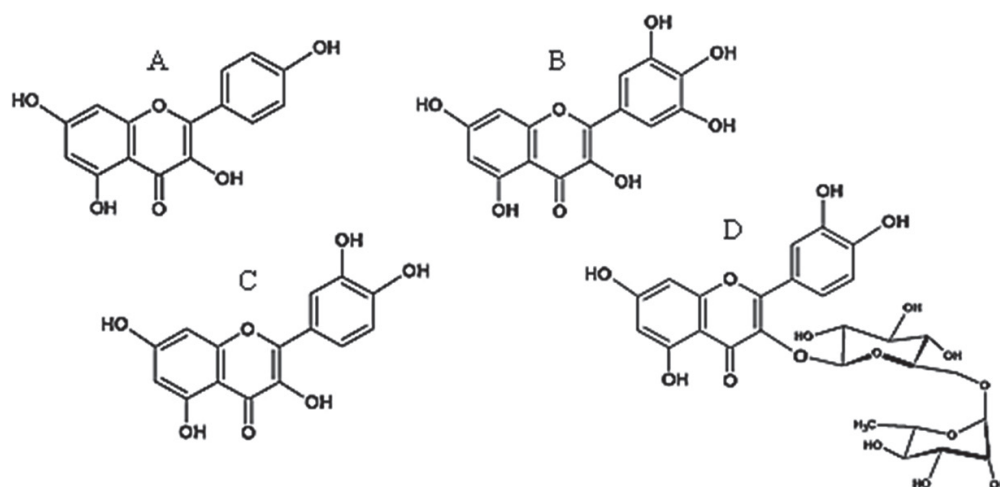


Figure 5. Chemical structures of some flavonoids found in *Azadirachta* plants. A = kaempferol, B = myricetin, C = quercetin, D = rutin.

4.3. *Russula* mushrooms

It is well established that many compounds separated from mushrooms can be used as immuno-modulators or as biological response modifiers [52]. Several mushroom species in Basidiomycetes have been reported to possess anti-tumor activity [53, 54].

Many phytochemical compounds have been reported in various mushrooms, and they can be classified into two main groups: high molecular weight compounds such as beta-glucan and other polysaccharides [55] and low molecular weight compounds including polyphenolics, flavonoids, and terpenoids [52]. Polyphenolics such as caffeic acid, chlorogenic acid, ferulic acid, and gallic acid and flavonoids such as myricetin and catechin were found in *Agaricusbisporus*, *Boletus edulis*, *Calocybe gambosa*, and *Cantharellus cibarius* [56]. Triterpenoids

were found in *Agaricus bisporus*, *Ganoderma lucidum*, and *Russula lepida*. Moreover, aristolane sesquiterpenoids were also found in *Russula lepida* [57]. Polysaccharides were found in *Agaricus bisporus*, *Agaricus brasiliensis*, *Ganoderma lucidum*, and *Phellinus linteus* [58]. Some polysaccharides such as beta-glucan are reported to promote immunomodulatory effects via CR3, the leukocytemembrane receptor for β -glucans [59]. The mechanisms of the action of the mushrooms to promote anticancer effects have been reported as NF- κ B inhibitors, protein kinase inhibitors, protein and DNA alkylating agents, modulators of G1/S and G2/M phases, inhibitors of MAPK protein kinase signaling pathways, aromatase and sulfatase inhibitors, matrix metalloproteinases inhibitors, cyclooxygenase inhibitors, DNA topoisomerases, and DNA polymerase inhibitors and anti-angiogenic substances [52].

A previous study reported the presence of 1147 mushroom species in the Northeast part of Thailand. They are composed of 647 consumed mushroom species, 222 trade mushroom species, and 400 poisonous mushroom species [60]. Thirty-seven species of these mushrooms are used in traditional medicine [60]. However, there are still some mushrooms in Thailand, especially in the Northeastern part of the country, that have never been studied for their biological properties and phytochemical compounds.

The *Russula* mushroom's shape resembles an umbrella. There have a clear cap and stem, with the gills underneath the cap. The cap is thin and has an underlying radius arranged around the center. The mushroom has no ring and no latex in the cap. The mushroom is fresh, soft, fragile, and perishable [61]. There are around 750 worldwide species of *Russula* [62, 63]. The distribution of the *Russula* species shows that they are present in several countries, including the United States of America, Sweden, France, Norway, Madagascar, Italy, Belgium, Taiwan, China, Japan, and Thailand [64]. In Thailand, *Russula* mushrooms have been found in 17 provinces in the Northeastern region of Thailand [65]. Numerous *Russula* mushrooms have been consumed as food such as *R. monspeliensis*, *R. virescens*, *R. alboareolata*, *R. medullata*, and *R. helios* [65, 66]. Various *Russula* mushrooms have been traditionally used for the treatments of various diseases such as *R. cyanoantha* and *R. nobilis*, which are used for the treatment of fever; *R. luteotacta*, which is used for wound healing; and *R. delica* and *R. parazurea*, which are used for the treatment of gastritis and high blood pressure, while *R. acrifolia* is used for treatments of skin cancer [36]. Moreover, some *Russula* mushrooms have also been traditionally used for tonic purposes such as *R. cyanoxantha*, *R. nobilis*, *R. delica*, *R. parazurea*, *R. acrifolia*, and *R. luteotacta* [67]. In addition, *Russula luteotacta* has been used as a sleep promoting agent [67]. Physical characteristics of some *Russula* mushrooms found in Thailand are shown in **Figure 6**.

Ten *Russula* mushroom collected from northeastern part of Thailand: *R. crustosa*, *R. delica*, *R. monspeliensis*, *R. velenovskyi*, *R. virescens*, *R. lepida*, *R. alboareolata*, *R. paludosa*, *R. medullata*, and *R. helios* were tested for their *in vitro* antioxidant activities using a photochemiluminescence assay for both lipid-soluble and water-soluble antioxidant capacities. *R. medullata* extract exhibited the highest antioxidant effects in both lipid-soluble and water-soluble models with antioxidant capacities of 1.1658 nmol of trolox equivalence and 1.323 nmol of ascorbic acid equivalence, respectively [68].

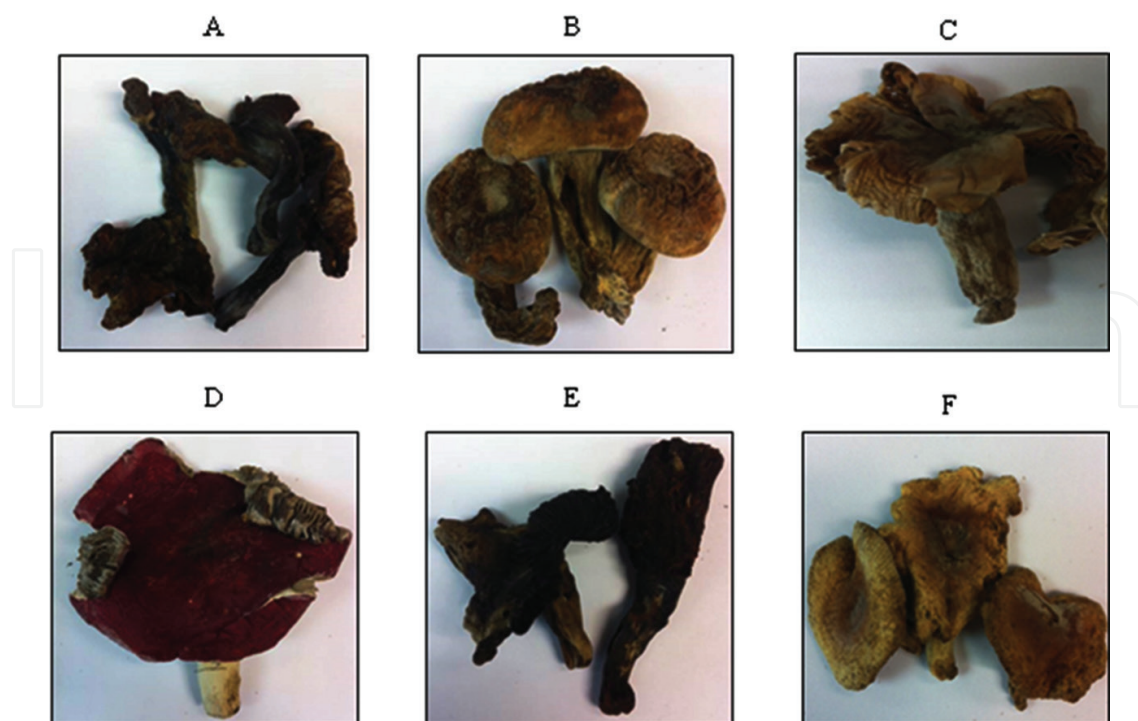


Figure 6. Physical characteristics of some *Russula* mushrooms found in Thailand; A = *Russula crustosa* Peck, B = *Russula delicata* Fries, C = *Russula monspeliensis* Sarnari, D = *Russula velenovskyi* Melzer & Zvára, E = *Russula virescens* (Schaeff) Fries, F = *Russula alboareolata* Hongo.

Some chemical constituents have been reported from *Russula* mushrooms including phenolic acids such as *o*-hydroxy-benzoic acid, chlorogenic acid, ferulic acid, caffeic acid, protocatechuic acid, and coumaric acid and flavonoids such as quercetin, chrysin, and catechin [69–71]. Some terpenoids were also found in *Russula* mushrooms including aristolane and marasmane [57, 72]. The chemical structures of the constituents found in *Russula* mushrooms are shown in **Figure 7**.

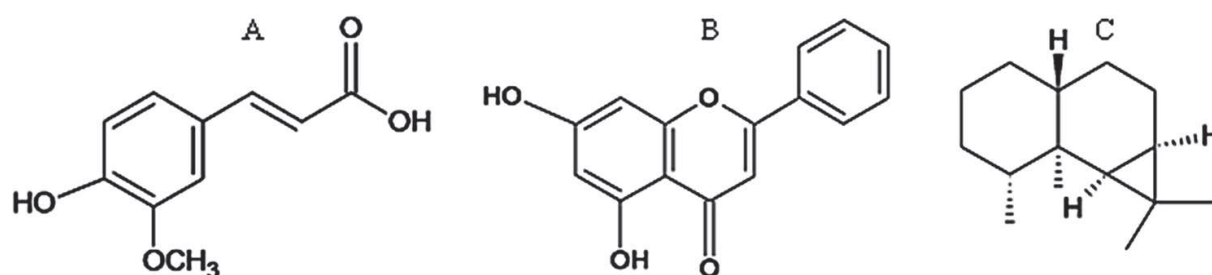


Figure 7. Chemical structures of some flavonoids found in *Russula* mushrooms. A = ferulic acid, B = chrysin, C = aristolane.

5. Conclusion

Natural products have been main sources of drug discoveries including the development of active compounds or formulas for the treatment of cancers. Even though it has become

difficult to discover or synthesize new active components, with the knowledge and intelligence regarding traditional medicine, there are still several ethnomedical herbal formulas and regional plants that could be studied and developed for further medicinal utilizations. Herbal remedies from Wat Tha-it and Wat Khampramong, Thailand, are examples of the efforts to develop anticancer therapies from traditional knowledge. Both remedies can inhibit the growth of various cancer cell lines. The stem extract and active compound, Berberine from the Thai medicinal plant *Coscinium fenestratum*, significantly promoted anti-proliferative activity on human colorectal cancer cells with the mechanism of action via NAG-1 and AFT3. Plants in the genus *Azadirachta* have been traditionally used as a tonic. They promote significant antioxidant activities, which could support the body's systems and prevent oxidative stress, which is one of the causes of carcinogenesis. *Russula* is the local mushroom species in the Northeastern part of Thailand. They promote significant antioxidant effects in both lipid-soluble and water-soluble models. These plants and natural products have the potential to be sources of anticancer compounds or active extracts for the treatments of cancer. However, standardization and quality control of the extract or active compounds should be performed before studying the toxicity, *in vivo* biological activity tests, and further clinical studies in the future.

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References

- [1] Attasara P, Buasom R (2009). Hospital-based cancer registry. National Cancer Institute. Department of Medical Services. Ministry of Public Health. [Access on January 10, 2017] http://www.nci.go.th/th/File_download/Nci%20Cancer%20Registry/hospital%20based%20cancer%20registry.pdf

- [2] Cooper GM, Sunderland MA (2000). The development and causes of cancer. The cell: a molecular approach. 2nd edition. Sinauer Associates.
- [3] Jiratchariyakul W (2015). Anticancer substances in medicinal plants. M and M Laser Printing Co. Ltd. Bangkok (book in Thai).
- [4] Kummalue T (2012). Anticancer mechanisms of medicinal plants, a medical research. Faculty of Medicine Siriraj Hospital, Mahidol University. Bangkok (book in Thai).
- [5] Evans WC (2009). Trease and Evans Pharmacognosy. 16th edition. Elsevier. London.
- [6] Ondee S, Sithisarn P, Ruangwises N, Rojsanga P (2015). Anti-proliferative activity on colorectal cancer cells of thirty Thai edible plants. Proceeding in the 1st international conference on pharmacy education and research network of ASEAN, Bangkok, Thailand, December, 2-4.
- [7] Shibata MA, Iinuma M, Morimoto J, Kurose H, Akamatsu K, Okuno Y, Akao Y, Otsuki Y (2011). α -Mangostin extracted from the pericarp of the mangosteen (*Garcinia mangostana* Linn) reduces tumor growth and lymph node metastasis in an immunocompetent xenograft model of metastatic mammary cancer carrying a p53 mutation. BMC Med. 9(69):1-18.
- [8] Naowaratwattana W, De-eknamkul W, De Meija EG (2010). Phenolic containing organic extracts of mulberry (*Morusalba* L.) leaves inhibit Hep G2 hepatoma cell through G2/M phase arrest, induction of apoptosis, and inhibition of topoisomerase II alpha activity. J Med Food. 13(5):1045-56.
- [9] Dakeng S, Duangmano S, Jiratchariyakul W, U-pratya Y, Bogler O, Patmasiriwat P (2012). Inhibition of Wnt signaling by cucurbitacin B in breast cancer cells: reduction of Wnt-associated proteins and reduced translocation of galectin-3-mediated β -catenin to the nucleus. J Cell Biochem. 113(1):49-60.
- [10] Hsu YL, Wu LY, Kuo PL (2009). Dehydrocostus lactone, a medicinal plant-derived sesquiterpene lactone, induces apoptosis coupled to endoplasmic reticulum stress in liver cancer cells. J Pharmacol Exp Ther. 329(2):808-19.
- [11] Kummalue T, Sujiwattanarat P, Jiratchariyakul W (2011). Apoptosis inducibility of Sapindusrorak water extract on A549 human lung cancer cell line. J Med Plant Res. 5(7):1087-94.
- [12] Kaewkon W, Khamprasert N, Limpeanchob N (2011). Derris scandens Benth extract induced necrosis rather than apoptosis of SW480 colon cancer cell. Thai J Pharmacol. 33(2):118-21.
- [13] Moongkarndi P, Jaisupa N, Kosem N, Konlata J, Samer J, Pattanapanyasat K, Rodpai E (2015). Effect of purified α -mangostin from mangosteen pericarp on cytotoxicity, cell cycle arrest and apoptotic gene expression in human cancer cells. World J Pharm Sci. 3(8):1473-84.
- [14] Chirathaworn C, Kongcharoensuntorn W, Dechdougchan T, Lowanitchapat A, Sa-nguanmoo P, Poovorawan Y (2007). Myristica fragrans Hoult. methanolic extract

- induces apoptosis in a human leukemia cell line through SIRT1 mRNA downregulation. *J Med Assoc Thai.* 90(11):2422-8.
- [15] Jiratgariyakul W, Okabe H, Moongkarndi P, Frahm AW (1998). Cytotoxic glycosphingolipid from *Murdannia loriformis* (Hassk.) Rolla Rao et Kammathy. *Thai J Phytopharm.* 5(1):10-20.
- [16] Sudha P, Syed M, Sunil D, Gowda C (2010). Immunomodulatory activity of methanolic leaf extract of *Moringa oleifera* in animals. *Indian J Physiol Pharmacol.* 54(2):133-40.
- [17] Srisapoomi T, Jiratchariyakul W, O-partkiattikul N, Kummalue T (2008). Effects of two Thai herbal remedies on the sensitivity of chemotherapeutic agents in human cancer cells. *Asian J Trad Med.* 3(4):144-52.
- [18] Soonthornchareonnon N, Sireeratawong S, Wiwat C, Ruangwises N, Wongnopphavich A, Jaijoy K (2011). Research and development of anti-cancer formula from Wat Khampramong. National Research Council of Thailand. Faculty of Pharmacy, Mahidol University, Bangkok.
- [19] Baek SJ, Kim KS, Nixon JB, Wilson LC, Eling TE (2001). Cyclooxygenase inhibitors regulate the expression of a TGF-beta superfamily member that has proapoptotic and antitumorigenic activities. *Mol Pharmacol* 59:901-8.
- [20] Rojsanga P, Sukhthankar M, Krisanapan C, Gritsanapun W, Lawson DB, Baek SJ (2010). In vitro anti-proliferative activity of alcoholic stem extract of *Coscinium fenestratum* in human colorectal cancer cells. *Exp Ther Med.* 1:181-6.
- [21] Baek SJ, Kim JS, Jackson FR, Eling TE, McEntee MF, Lee SH (2004). Epicatechin gallate induced expression of NA G-1 is associated with growth inhibition and apoptosis in colon cancer cells. *Carcinogenesis.* 25:2425-32.
- [22] Baek SJ, Wilson LC, Eling TE (2002). Resveratrol enhances the expression of non-steroidal anti-inflammatory drug-activated gene (NA G-1) by increasing the expression of p53. *Carcinogenesis.* 23: 425-34.
- [23] Martinez JM, Sali T, Okazaki R, Anna C, Hollingshead M, Hose C, Monks A, Walker NJ, Baek SJ, Eling TE (2006). Drug-induced expression of nonsteroidal anti-inflammatory drug-activated gene/macrophage inhibitory cytokine-1/prostate-derived factor, a putative tumor suppressor, inhibits tumor growth. *J Pharmacol Exp Ther.* 318:899-906.
- [24] Rojsanga P, Sukhthankar M, Baek SJ (2007). Berberine, a natural isoquinoline alkaloid, induces NAG-1 and AFT3 expression in human colorectal cancer cells. *Cancer Lett.* 258(2): 230-40.
- [25] Lee SH, Cekanova M, Baek SJ (2008). Multiple mechanisms are involved in 6-gingerol-induced cell growth arrest and apoptosis in human colorectal cancer cells. *Mol Carcinog.* 47:197-208.
- [26] Forman LL (1991). *Menispermaceae.* Bangkok: The Forest Herbarium.300-65.

- [27] Wattanathorn J, Uabundit N, Itarat W, Mucimapura S, Laopatarakasem P, Sripanidkulchai B (2006). Neurotoxicity of *Coscinium fenestratum* stem, a medicinal plant used in traditional medicine. *Food Chem Toxicol.* 44:1327-33.
- [28] Cross EC (1987). Oxygen radicals and human disease. *Ann Intern Med.* 107: 526-45.
- [29] Carocho M, Ferreira ICFR (2013). The role of phenolic compounds in the fight against cancer-a review. *Anticancer Agents Medicinal Chem.* 13:1236-58.
- [30] Ji B, Hsu W, Yang J, Hsia T, Lu C, Chiang J, Yang J, Lin C, Lin J, Suen L, Wood WG, Chung J (2009). Gallic acid induces apoptosis via caspase-3 and mitochondrion-dependent pathways in vitro and suppresses lung xenograft tumor growth in vivo. *J Agric Food Chem.* 57:7596-7604.
- [31] Kim S, Gaber MW, Zawaski JA, Zhang F, Richardson M, Zhang XA, Yang Y (2009). The inhibition of glioma growth in vitro and in vivo by a chitosan/ellagic acid composite biomaterial. *Biomaterials.* 30:4743-51.
- [32] Baskaran N, Manoharan S, Balakrishnan S, Pugalendhi P (2010). Chemopreventive potential of ferulic acid in 7,12-dimethylbenz[a]anthracene-induced mammary carcinogenesis in Sprague-Dawley rats. *Eur J Pharmacol.* 637:22-9.
- [33] Nair H, Rao KVK, Aalinkeel R, Mahajan S, Chawda R, Schwartz SA (2004). Inhibition of prostate cancer cell colony formation by the flavonoid quercetin correlates with modulation of specific regulatory genes. *Clin Diagn Lab Immunol.* 11:63-9.
- [34] Yuan Z, Chen L, Fan L, Tang M, Yang G, Yang H, Du X, Wang G, Yao W, Zhao Q, Ye B, Wang R, Diao P, Zhang W, Wu H, Zhao X, Wei Y (2006). Liposomal quercetin efficiently suppresses growth of solid tumors in murine models. *Clin Cancer Res.* 12:3193-9.
- [35] Zhang H, Zhang M, Yu L, Zhao Y, He N, Yang X (2012). Antitumor activities of quercetin and quercetin-50,8-disulfonate in human colon and breast cancer cell lines. *Food Chem Toxicol.* 50:1589-99.
- [36] Suttajit S, Khansuwan U, Suttajit M (2002). Antioxidative activity of Thai medicinal herbs. *Thai J Pharm Sci.* 26(suppl.):32.
- [37] Trakoontivakorn G, Saksitpitak J (2000). Antioxidative potential of Thai indigenous vegetable extracts. *Journal of Food Research and Product Development.* Kasetsart University. 30(3):164-76.
- [38] Sombatsiri K, Ermel K, Schmutterer H (1995). Other Meliaceae plants containing ingredients for integrated pest management and further purpose. In: Schmutterer H. The neem tree *Azadirachta indica* A. Juss. and other meliaceae plants. Germany: VCH.
- [39] Sithisarn P, Gritsanapan W (2008). Siamese neem tree: A plant from kitchen to antioxidative health supplement. *Advances in Phytotherapy Research.* Research Signpost. Kerala. India.
- [40] Clayton T, Soralump P, Chaukul W, Temsiririrkkul R (1996). Medicinal plants in Thailand volume 1. Bangkok: Amarin Printing.

- [41] Te-Chato S, Rungnoi O (2000). Induction of somatic embryogenesis from leaves of Sadao Chang (*Azadirachta excelsa* (Jack) Jacobs). *Sci Horticult.* 86:311-21.
- [42] Arivazhagan S, Balasenthil S, Nagini S (2000). Garlic and neem extracts enhance hepatic glutathione-dependent enzymes during N-methyl-N-nitro-N-nitrosoguanidine (MNNG)-induced gastric carcinogenesis in rats. *Phytother Res.* 14:291-3.
- [43] Akudugu J, Gade G, Bohm L (2001). Cytotoxicity of azadirachtin A in human glioblastoma cell lines. *Life Sci.* 68:1153-60.
- [44] Subapriya R, Nagini S (2003). Ethanolic neem leaf extracts protects against N-methyl-N-nitro-N-nitrosoguanidine-induced gastric carcinogenesis in Wistar rats. *Asian Pac J Cancer Prev.* 4:215-23.
- [45] Sithisarn P, Gritsanapan W, Supabphol R (2004). Free radical scavenging activity of three *Azadirachta* plants. Proceeding in the 21st Annual Research meeting in Pharmaceutical Sciences, Bangkok, Thailand, December, 23-24, 2004.
- [46] Sithisarn P, Carlsen CU, Andersen ML, Gritsanapan W, Skibsted LH (2007). Antioxidative effects of leaves from *Azadirachta* species of different provenience. *Food Chem.* 104: 1539-49.
- [47] Sithisarn P, Supabphol R, Gritsanapan W (2005). Antioxidant activity of Siamese neem tree. *J Ethnopharmacol.* 99: 109-12.
- [48] Sithisarn P, Suksangpanomrung M, Gritsanapan W (2007). Gene expression of enzymes related to biosynthesis of antioxidative flavonoids in Siamese neem tree leaves. *Planta Med.* 73: 222.
- [49] Pankadamani KS, Seshadri TR (1952). Survey of anthoxanthins. *Proc Indian Acad Sci Ser A* 36: 157-69.
- [50] Nakov N, Labode O, Akahtaedzhiev K (1982). Study of the flavonoid composition of *Azadirachta indica*. *Farmatsiya (Sofia).* 32:24-8.
- [51] Siddiqui S, Mahmood T, Siddiqui BS, Faizi S (1985). Studies in the nonterpenoidal constituents of *Azadirachta indica*. *Pak J Sci Ind Res.* 28(1):1-4.
- [52] Zaidman BZ, Yassin M, Mahajna J, Wasser SP (2005). Medicinal mushroom modulators of molecular targets as cancer therapeutics. *Appl Microbiol Biotechnol.* 67: 453-68.
- [53] Mizuno T (1995). Bioactive biomolecules of mushrooms: food function and medicinal effect of mushroom fungi. *Food Rev Int.* 11:7-21.
- [54] Wasser SP (2002). Medicinal mushrooms as a source of antitumor and immunomodulating polysaccharides. *Appl Microbiol Biotechnol.* 60:258-274.
- [55] Jaruntorn B, Chanida H (2010). Spatial distribution of Beta glucan containing wild mushroom communities in subtropical dry forest, Thailand. *Fungal Divers.* 46(1): 29-42.
- [56] Palacios I, Lozano M, Moro C, D'Arrigo M, Rostagno MA, Martínez JA, García-Lafuente A, Guillamón E, Villares A (2011). Antioxidant properties of phenolic compounds occurring in edible mushrooms. *Food Chem.* 128:674-8.

- [57] Jian-Wen T, Ze-Jun D, Ji-Kai L (2000). New terpenoids from Basidiomycetes *Russula lepida*. *Helv Chim Acta*. 83:3191-7.
- [58] Kozarski M, Klaus A, Niksic M, Jakovljevic D, Helsper JPF, Van Griensven LJLD (2011). Antioxidative and immunomodulating activities of polysaccharide extracts of the medicinal mushrooms *Agaricusbisporus*, *Agaricusbrasiliensis*, *Ganodermalucidum* and *Phellinusluteus*. *Food Chem*. 129(4):1667-75.
- [59] Xia Y, Vetvicka V, Yan J, Hanikyrova M, Mayadas T, Ross GD (1999). The beta-glucan-binding lectin site of mouse CR3 (CD11b/CD18) and its function in generating a primed state of the receptor that mediates cytotoxic activation in response to iC3b-opsonized target cells. *J Immunol*. 162:2281-90.
- [60] The Bureau of Thai Indigenous Medicine, Department for Development of Thai Traditional and Alternative Medicine. Ministry of Public Health (2011). Mushrooms are health food from folk medicine. The Bureau of Thai Indigenous Medicine, Department for Development of Thai Traditional and Alternative Medicine. Ministry of Public Health. Nonthaburi. (book in Thai).
- [61] Sonoamuang N (2010). Wild mushrooms of Thailand: biodiversity and utilization. Department of Plant Science and Natural Resources. Faculty of Agriculture, Khon Kaen University.
- [62] Joshi S, Bhatt RP, Stephenson SL (2012). The current status of the family Russulaceae in the Uttarakhand Himalaya, India. *Mycosphere*. 3(4):486-501.
- [63] Jain N, Pande V (2013). Diversity analysis of ectomycorrhizal genus *Russula* using RAPD markers. *Octa Jour Env Res*. 1(4):332-5.
- [64] Buyck B, Hofstetter V, Eberhardt U, Verbeken A, Kauff F (2008). Walking the thin line between *Russula* and *Lactarius*: the dilemma of *Russula* subsect. *Ochricompactae*. *Fungal Divers*. 28:15-40.
- [65] Manassila M, Sooksa-Nguan T, Boonkerd N, Rodtongb S, Teaumroonga N (2005). Phylogenetic diversity of wild edible *Russula* from Northeastern Thailand on the basis of internal transcribed spacer sequence. *Science Asia*. 31:323-8.
- [66] Quiñónez-Martínez M, Ruan-Soto F, AguilarMoreno IE, Garza-Ocañas F, LebgueKeleng T, Lavín-Murcio PA, Enríquez-Anchondo ID (2014). Knowledge and use of edible mushrooms in two municipalities of the Sierra Tarahumara, Chihuahua, Mexico. *J Ethnobiol Ethnomed*. 10(67):1-13.
- [67] Sanmee R, Dellb B, Lumyongc P, Izumorid K, Lumyong S (2003). Nutritive value of popular wild edible mushrooms from northern Thailand. *Food Chem*. 82: 527-32.
- [68] Jaengklang C, Jarikasem S, Sithisarn P, Klungsunya P (2015). Determination on antioxidant capacity and TLC analysis of ten Thai *Russula* mushroom extracts. *Isan J Pharm Sci*. 10:241-50.

- [69] Yaltirak T, Aslim B, Ozturk S, Alli H (2009). Antimicrobial and antioxidant activities of *Russula delica* Fr. *Food Chem Toxicol.* 47:2052-6.
- [70] Chen XH, Xia LX, Zhou HB, Qiu GZ (2010). Chemical composition and antioxidant activities of *Russula griseocarnosa* sp. nov. *J Agric Food Chem.* 58:6966-71.
- [71] Kalogeropoulos N, Yanni AE, Koutrotsios G, Aloupi M (2013). Bioactive microconstituents and antioxidant properties of wild edible mushrooms from the island of Lesbos, Greece. *Food Chem Toxicol.* 55:378-85.
- [72] Clericuzio M, Cassino C, Corana F, Vidari G (2012). Terpenoids from *Russula lepida* and *Russula amarissima* (Basidiomycota, Russulaceae). *Phytochemistry.* 84:154-9.

