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Review

Studies on hepatoprotective effects of Vietnamese medicinal plants

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Abstract. The liver is a major organ of the human and animal body, playing an important role in metabolism, detoxification and immunomodulation. When liver tissue is damaged, liver function will decrease, leading to liver diseases such as liver fibrosis, cirrhosis, and liver cancer. Medicinal plants and natural products have provided a source of new drug candidates for the prevention and treatment of liver diseases. This review is based on scientific publications on Vietnamese plants investigated for their hepatoprotective activity during the period from 1998 to 2022. 51 medicinal plants, 38 phytoconstituents, and 8 hepatoprotective herbal formulations of Vietnamese-origin have been reported to protect the liver from the harmful effects induced by hepatotoxins, typically CCl_4 or paracetamol. Each plant's information, including its botanical name, family, part of the plant used, local name, chemical constituents, extracts or fractions used, dosage, model used, parameters obtained, histopathology, and results of hepatoprotective studies, is displayed. Also discussed is the possible mechanism of action of these hepatoprotective plants.

Keywords: hepatoprotective plants, *in vivo*, liver failure, hepatitis.

Classification numbers: 1.2.1, 1.2.5, 1.4.4.

1. INTRODUCTION

The liver is a major organ of the human and animal bodies, playing an important role in metabolism, detoxification, and immunomodulation. As the liver is associated with nearly all metabolic processes that occur in the body, if the liver is damaged and its function decreases, our body will be seriously affected. Liver damage caused by acute or long-lasting exposure to hepatotoxins is associated with serious liver diseases such as hepatitis, fibrosis, cirrhosis, and liver cancer, which can lead to other bodily disorders and even death. Cirrhosis causes a large national economic burden of more than $14 million per year for patient treatment and $2 billion in indirect costs due to lost labour and reduced quality of life [1]. Liver cancer usually develops as a result of liver fibrosis or cirrhosis and is characterised by uncontrolled proliferation of

cancer cells in the liver. According to the study of the global burden of disease in 2020, chronic liver diseases accompanied by liver fibrosis and cirrhosis with high morbidity and mortality rates are the 6th leading cause of death in non-communicable diseases around the world [2].

In recent years, the rate of people suffering from liver diseases in Viet Nam has tended to increase rapidly. Viet Nam was in the top 5 countries with the highest overall rate of liver cancer in 2020, with 26,418 diagnosed incidences and an overall mortality rate from liver cancer of 23.0 cases per 100,000 residents [3]. Viral hepatitis and alcoholism are the leading causes of liver disease in Viet Nam. In addition, more rarely, cirrhosis can be induced by other causes such as drugs, chemicals, flukes, genetic metabolic disorders, autoimmune diseases, etc.

Ethnic medicinal plants play an important role in many traditional medicine cultures, such as China, Ayurveda, Thailand, Viet Nam, etc., for the prevention and treatment of liver diseases. An excessive number of herbals (~ 101 plants) [4], fruits (grapefruit, cranberries, cactus pear fruits, and grapes) [5], chemical constituents (extracted from fruits, plants, yeasts, and algae), resin (propolis) [5], and herbal formulations [4] have been reported to possess hepatoprotective activity. Currently, a number of medicinal plants are available, for example, *Solanum hainanense* Hance (Solanaceae), *Silybum marianum* (L.) Gaertn. (milk thistle), *Picrorhiza kurroa* Royle ex Benth (kutkin), *Curcuma longa* L. (turmeric), *Camellia sinensis* (L.) Kuntze (green tea), *Chelidonium majus* L. (greater celandine), *Glycyrrhiza glabra* L. (licorice), *Allium sativum* L. (garlic), and *Phyllanthus* sp., etc. in the form of plant extract, herbal tea, or pill capsules, have been marketed as functional foods, liver supplements, herbal liver tonics, and hepatic tonics. Consequently, the research and development of hepatoprotective drugs from ethnic medicinal plants are being promoted by scientists around the world. Vietnamese scientists have also long investigated and discovered plants and active ingredients with hepatoprotective activity from rich natural resources of 5,117 species and sub-species belonging to 1,823 genus, 360 families, and 8 phyla of vascular higher plants [6].

This review is based on scientific research conducted between 1998 and 2022 on plants of Vietnamese origin using *in vitro* and *in vivo* hepatoprotective assays. Briefly, it summarized the scientific data of 51 medicinal plants, 38 phytoconstituents, and 8 hepatoprotective herbal formulations that were published to possess hepatoprotective activity to protect the liver from the harmful effects of hepatotoxins, typically CCl_4, paracetamol, or other chemicals. Each plant's botanical name, family, part used, local name, chemical constituents, extracts or fractions used, dosage, model used, parameters obtained, histopathology, and results of hepatoprotective studies were reported. The possible mechanism of action of these hepatoprotective plants was also discussed.

2. MATERIALS AND METHODS

2.1. Data resources

This review is aimed at compiling data on hepatoprotective plants grown in Viet Nam or published in the form of project reports, theses, or publications from Vietnamese governmental and local data resources such as https://db.vista.gov.vn during the period from 1998 to 2022. Internet searches were used to collect published data in different international scientific journals through PubMed and Google Scholar search engines. Vietnamese traditional medicinal books [7] and Vietnam Pharmacopoeia V [8] were also used to collect information about the distribution and traditional uses of these plants.

2.2. Data collections

Multiple keyword combinations, including "hepatoprotective", "medicinal plants", "natural products", "herbal formulations", "hepatotoxins", "HepG2", "CCl_4", "paracetamol", "Vietnamese", "*in vitro*", "*in vivo*", and "Vietnam", were used to search the scientific literature. The selected documents need to describe the plants collected in Viet Nam and used in *in vitro* and *in vivo* assays such as carbon tetrachloride (CCl_4), paracetamol (PAR), etc. -induced liver injury on hepatic cell lines (HepG2, hepatocytes) or animal models to assess their hepatoprotective activity. The effectiveness of the herbal drug is compared to that of conventional drugs used to treat liver disease, specifically silymarin [9, 20]. As biomarkers for hepatocyte damage, the liver enzymes aspartate transaminase (AST), alanine transaminase (ALT), alkaline phosphatase (ALP), gamma glutamyl transferase (GGT), total bilirubin (BIL), total protein (TP), and albumin (Alb) were measured. The review also includes a comparison of the histopathological effects of standard hepatotoxic agents with those of plant extracts used as hepatoprotective agents.

3. GENERAL INFORMATION

The liver is the body's largest organ (1200 - 1800 g, or 2 % of the adult body weight). It is comprised of four major lobes and 50,000 - 100,000 lobules and is considered a functional unit. The liver receives blood from two distinct sources: nutrient-rich blood flow from the digestive system and the spleen through the portal vein (~75 %) and oxygen-rich blood flow through the hepatic artery (~25 %). Regarding microscopic structure, the liver is made up of two types of cells: liver parenchymal cells, also known as hepatocytes, which account for about 70 - 80 %, and non-parenchymal cells such as Kupffer cells, astrocytes, capillary endothelial cells, and dendritic cells [9].

The liver performs more than 500 distinct functions and is associated with nearly all metabolic processes in the body, such as metabolism, secretion, storage, and detoxification. It regulates and receives nutrients, metabolises substances (carbohydrates, lipids, and proteins), produces many important functional molecules (plasma proteins (albumin, globulin), clotting factors (fibrinogen, prothrombin), cholesterols, bile), regulates the levels of glucose, amino acids, and clotting factors in the blood, stores molecules (glycogen, iron, vitamins (A, D, K, E, and B12), and eliminates waste metabolised substances (endogenous) such as billirubin, poisonous ammonia and foreign substances (exogenous) including hepatotoxins (endotoxins, bacterial toxins, alcohol, free radicals, etc.) from the blood before they are transported to all organs, tissues, and cells in the body. To perform these functions, the liver produces many liver enzymes commonly known as AST, ALT, ALP, GGT, etc., which are proteins that accelerate chemical reactions in the body. When the levels of these liver enzymes are higher than normal, they may indicate acute or chronic liver diseases such as viral hepatitis, fibrosis, or liver cancer. Liver fibrosis is a pathological wound-healing process in which extracellular matrix protein (connective tissue), particularly collagen, persistently accumulates in the liver and replaces normal tissue. This tissue remodeling can silently progress and lead to cirrhosis, hepatocellular carcinoma, liver failure, and finally death.

Liver diseases may result from many causes, including viral infections (e.g., viral A-E hepatitis), genetic conditions (immunity problems, inherited diseases), and toxic chemical exposure [10]. When performing its functions, the liver is always at risk of acute or long-lasting exposure to hepatotoxic or harmful chemicals, including poisonous toxins or chlorinated compounds, drug overdose, as well as stimuli such as reactive oxygen species (ROS), alcohol,

heavy metals (such as iron, arsenic, cadmium, and copper), and tobacco smoking, which cause the hepatic cells to be damaged and die [11]. This makes the liver unable to perform its full functions, and it will lead to serious disturbances in other processes occurring in the body.

Typical liver disease symptoms include jaundice, easy bruising or bleeding, itchy skin, spider-like veins beneath the skin, loss of appetite, nausea, and diarrhea, etc. Over time, liver diseases may trigger more serious complications, for example, lower body edema or ascites (severe accumulation of fluid in the abdomen and legs); hepatic encephalopathy (accumulation of waste product ammonia in the brain, which destroys brain functions, causes confusion, fatigue, memory loss, and diminishes mental abilities); hepatorenal syndrome (a form of progressive kidney failure associated with liver damage); and portal hypertension. These liver diseases decline the patient's quality of life and cause a high rate of mortality. Consequently, the search for novel agents with hepatoprotective properties to repair liver damage and prevent liver failure is very important.

Description of biological activities related to the discovery of hepatoprotective medicines

Many medicinal plants have been reported to protect the liver from toxicity. To evaluate the hepatoprotective activity of the extracts and their compounds, many experimental assays or models that mimic the processes of liver toxicity and liver treatment are built up.

Hepatoprotective activity is generally understood as the protective effects on the activity of hepatocytes and the function and state of the liver against toxicity caused by hepatotoxins. Therefore, hepatoprotective activity can be evaluated by a combination of numerous related biological activities, such as antioxidant, anti-inflammatory, antiviral (anti-hepatitis), antifibrotic, (Supporting Information), and inhibition of hepatotoxin-induced liver toxicity. These assessments may be conducted *in vitro, ex vivo, or in vivo* (in experimental animals).

- ***Inhibition of hepatotoxin-induced liver toxicity*****:** These assays (*in vitro, ex vivo,* and *in vivo*) are used for the evaluation of hepatoprotective activity based on the use of chemicals or biological factors called hepatotoxins that cause damage to the cells, tissues, structure, and liver function. In the presence of hepatotoxins, the liver cells become damaged and die. Thus, compounds that protect the liver cells from injury or apoptosis by inhibiting hepatotoxin-induced liver toxicity and therefore supporting cell survival, reversing liver damage, and recovering liver function are considered to possess hepatoprotective activity.

- ***Hepatotoxins:*** There are many hepatotoxic agents of natural and human-made origin, such as heavy metals (cadmium, arsenic), biological factors (bacteria, viruses, and parasites), toxins (mycotoxins, endotoxins, plant toxins), and especially chemicals, which enter the body naturally through foods, canned food, drinking water, the environment, air, industrial factories, pesticides, toys, etc., or purposefully through the routine taking of different drugs or medicines. Even medicinal benefits, natural substances, and pharmaceuticals could become hepatotoxins when they are mishandled, abused, or improperly used (application route, dosage, time of use, patient's age, patient's status (pregnant women, children, with chronic diseases or kidney failure, etc.). More than 900 drugs, including paracetamol, antitubercular drugs, anticancer drugs, alcohols, etc., have been reported to be toxic to the liver [12] by inducing oxidative stress and liver cell damage. In the hepatoprotective experiment, hepatotoxins such as certain drugs [paracetamol (PAR)] and toxic compounds [CCl_4, thioacetamide, dimethylnitrosamine (DMN), d-galactosamine/ lipopolysaccharide (GALN/LPS), and alcohol] are frequently used to induce hepatotoxicity and acute liver failure in *vitro, ex vivo,* and *in vivo* models. While *in vitro* experiments are valuable tools for studying the hepatoprotective mechanism at the cellular level

of therapeutic agents, *ex vivo and in vivo* hepatoprotective activity assays provide additional information on the pathological and progressive pathways as well as the hepatoprotective mechanisms involved in tested samples [13].

- ***In vitro study model of hepatoprotective activity:*** In the *in vitro* hepatoprotective activity model, cells were exposed to hepatotoxic agents in order to evaluate the hepatoprotective effect and mechanism of action of compounds at the cellular and molecular levels. For hepatotoxic studies, fresh hepatocytes, cultured cells, and immortalized hepatocytes can be used [14]. Hepatocytes are damaged by hepatotoxins and then used to evaluate the hepatoprotective effects of the extracts and study agents. The effectiveness of the treatment was evaluated based on the activity of released intracellular enzymes, cell proliferation, cell morphology, synthesis of macromolecules, oxygen consumption, etc. [15]. Murine (Hepa1c1c7) hepatoma cells and human cancer liver cell lines, including HepG2, Hep3B, Huh7, and HepArg cells, are also used due to their experimental robustness, reproducibility, and infinite growth capacity. However, according to the assays induced by PAR, these cancer cell lines may present genetic instability and produce reactive metabolites independently, rendering them unsuitable for this *in vitro* study [13]. CCl_4 (500 μg/mL (1 %)) [16] or tert-butyl hydroperoxide (t-BHP) (2 mM/kg) [17] could be more appropriate hepatotoxins for *in vitro* hepatoprotective models.

The *in vitro* hepatoprotective model is commonly used for screening purposes because it is possible to screen a large number of samples in a short time with a small sample size and at a low cost. The results are more reproducible than the *in vivo* method. However, the primary hepatocytes cannot be maintained for a long time. And human cancer cells may differ in biochemical and metabolic patterns from normal cells [18]. In addition, the results of *in vitro* experiments may differ from those of *in vivo* systems.

- ***Ex-vivo study model of hepatoprotective activity:*** In *ex vivo* studies, fresh liver slices mimicking *in vivo* tissue are employed for hepatoprotective analyses. In liver slices, hepatocytes, Kuffer cells, and hepatic stellate cells have the ability to survive, the cells still interact, and the liver part maintains its essential activities. So the *ex vivo* models bridge *in vitro* and *in vivo* investigations to study drug metabolism and liver damage [18]. In the *ex vivo* study model, the number of experimental animals is significantly reduced, the cost is low, and the model can be performed on human tissues and organs. However, the low cell proliferation and short survival duration of liver slices, approximately 8 - 10 days, affect the *ex vivo* outcomes [18].

- ***In vivo study model of hepatoprotective activity:*** In an *in vivo* model, animals like mice and rats are used for experiments. A hepatotoxic agent in a toxic dose or repeated doses is given orally or intravenously to these animals to induce liver toxicity. The tested samples are administered along with, prior to, and/or after the toxin treatment [14]. Liver damage and recovery from damage are assessed by quantifying serum marker enzymes (ALT, AST, ALP, BIL, etc.), bile flow, and biochemical changes in the liver. Histopathology of the liver is also performed, as it reflects liver functions and evidence of liver cell damage [19]. Antioxidant liver enzymes such as superoxide dismutase (SOD), catalase (CAT), and glycogen phosphorylase (GP) are also monitored. These enzymes deactivate ROS and protect major molecules such as lipids, proteins, and DNA from oxidative damage. It is possible to screen and identify the mechanism of liver protection against harmful agents from plant extracts and isolated compounds using an *in vivo* model that closely resembles the human body. However, this model, which requires a large number of animals, is costly and time consuming.

Hepatotoxic agents commonly used in *in vivo* experiments are paracetamol (acetaminophen), CCl_4, ethanol, D-galactosamine, thioacetamide, *tert*-butyl hydroperoxide (t-BHP), etc.

+ Paracetamol (PAR) is a common and effective analgesic and antipyretic drug at normal doses, but its overdose can cause acute liver failure and liver necrosis due to the hepatotoxicity of its metabolite, N-acetyl-p-benzoquinone imine (NAPQI). Normally, NAPQI is removed by glutathione stored in the liver, but in overdose, reduced glutathione leads to the accumulation of NAPQI and hepatocellular necrosis. PAR overdose is also closely related to mitochondrial dysfunction when NAPQI has the ability to inhibit the electron transport chain, generate reactive oxygen free radicals, cause damage to mitochondria, and destroy liver cells. PAR at a dose of 400 - 500 mg/kg dissolved in water or mixed with 0.5 - 1 % CMC was the most commonly used oral dosage to induce liver failure in mice.

+ CCl_4 is the most commonly used hepatotoxic agent. Hepatocellular injury caused by CCl_4 involves the conversion of CCl_4 to trichloromethyl (CCl_3) or trichloroperoxyl ($CCl_3O_2^-$) radicals by the cytochrome P450 (CYP2E1, CYP2B, and CYP3A) oxygenase enzyme system of the endoplasmic reticulum, causing oxidative stress and cell membrane damage. Free radicals induce lipid peroxidation, leading to cell membrane damage and the increased release of pro-inflammatory cytokines such as TNF-α and IL-6. The total dose of CCl_4 administered was in the range of 0.2 - 2 mL/kg in acute liver damage by either subcutaneous or peritoneal injection with one-day treatment and in the range of 1.5 - 5 mL/kg in divided doses over one week for chronic (reversible) and 12 - 20 mL/kg for 5 - 12 weeks (irreversible) [20].

+ D-Galactosamine in combination with endotoxin such as lipopolysaccharide (LPS) (GAL/ET) is used to study apoptosis and necrosis in liver injury mediated by TNF-*α* [21].

+ t-BHP was treated intraperitoneally (i.p.) at a dose of 2 mM/kg, 100:1, dissolved in saline, to induce oxidative stress in mice [17].

+ Concanavalin A-based model is considered a relevant model of auto-immune-mediated liver injury [22].

+Trinitrotoluene, an explosive material, causes anaemia, aplasia, methemoglobinemia, and hepatocellular injury, which may lead to chronic liver dysfunction and cirrhosis in cases of prolonged exposure [23]. A dose of 10 mL (solution concentration of 10 mg/mL) (100 mg/kg body weight, daily, 6 times weekly for 6 weeks) was used to induce liver toxicity in male white mice [23].

In the *in vivo* experiments, the animals were tested for hepatoprotective activity with extracts and substances at different doses before or at the same time as the causative agent. The functions of the liver are assessed through liver biomarkers, and the methods to validate these biomarkers are called liver function tests. In general, liver function tests evaluate various marker enzymes that are related to liver functions. The hepatoprotective and toxic effects of the extracts were evaluated by biochemical parameters such as the activity of a number of plasma intracellular enzymes (ALT, AST,ALP, and GPT) for the metabolic function; total protein (TP), albumin, and globulin for the synthetic function; lactate dehydrogenase (LDH), glutathione (GSH), direct bilirubin, BIL, blood urea nitrogen (BUN) for the detoxifying function. ALT and AST levels are specifically raised when hepatocytesare damaged [1]. ALT and AST levels are normally maintained at 20 - 40 (UI/L), so an AST/ALT ratio greater than 1 may indicate a liver disorder. The BUN (normal range: 6 - 20 mg/dL) measures the amount of urea nitrogen produced by the liver in the blood. This parameter is primarily used to assess not only kidney function but also liver health [24]. Malondialdehyde (MDA) ($CH_2(CHO)_2$) is a biomarker of oxidative stress and serves as an indicator of lipid peroxidation in liver cells and liver injury [25].

This inhibition of hepatotoxin-induced liver toxicity assay (*in vitro* and *in vivo*) might be the most important experiment and can be used alone or in conjunction with the above-mentioned assays to elucidate the hepatoprotective mechanism or effects of medicinal plants. Based on the experimental results, many medicinal plants have been discovered to possess the hepatoprotective activity and can be further investigated for the development of drugs to treat liver diseases.

4. RESULTS AND DISCUSSION

Medicinal plants play a key role in the human health care system. The educated public and health care professionals have huge interests in the medicinal uses of herbs, but there is a great deal of confusion about their identification, effectiveness, therapeutic dosage, toxicity, standardization, and regulation. According to the World Health Organization, traditional medicine is popular in all regions of the world, and its use is rapidly expanding even in developed countries. In China, traditional herbal preparations account for 30 - 50 % of the total medicinal consumption, and the global market for herbal medicine now exceeds 60 billion USD annually. Consequently, western-trained physicians are currently more interested in the efficacy of traditional medicine for their patients.

Based on scientific publications of Vietnamese plants for their hepatoprotective activity during the period from 1998 to 2022, we have found 51 Vietnamese medicinal plants, belonging to 26 families, with used plant parts (leaves, roots, aerial parts, underground parts, seeds); 38 phytoconstituents and 8 herbal formulations with the ability to inhibit toxicity induced by hepatotoxins in experimental animals (*in vivo*), or on isolated hepatocytes (*in vitro, ex vivo*), presented in Table 1 *(Supporting Information - SI)*. Plant information including their botanical name, family, part of the plant used, Vietnamese name, chemical constituents, the extracts or fractions used, the dosage of extracts, the model used, parameters obtained, histopathology, and the results of hepatoprotective studies of each plant, was reported. The decreases and/or increases of these biomarkers in the treated animal groups when compared to those of the pathological group were expressed as symbols ↓ or ↑, respectively, with the values calculated as fold or % from $[ALT, AST]_{pathological\ group}$ / $[ALT, AST]_{treated\ group}$. In addition, the histopathological evaluation of several *in vivo* experiments was presented.

- Hepatoprotective activity of plant extracts

Table 1 - SI describes 51 medicinal *plants that belong to many different families,* such as *Aganope balansae* (Gagnep.) Phan (Vietnamese name Mạ mân, Fabaceae) [26], *Centlla asiatica* (L.) Urb. (Rau má, Apiaceae) [27], *Chloranthus japonicus* Roem. & Schult. (Sói nhật, Chloranthaceae) [28], *Colocasia esculenta* (L.) Schott. (Taro, Môn nước, Araceae) [29], *Curcuma longa* L. (Nghệ, Zingiberaceae) [30], *Desmodium triquetrum* L. (Mũi mác, Fabaceae) [31], *Eclipta prostrata* L. (cỏ Nhọ nồi, Asteraceae) [32], *Ganoderma lucidum* (Fr.) P.Karst. (Lingchi, Ganodermaceae) [33, 34], *Hovenia dulcis* Thunb. (Khúng khéng, Rhamnaceae) [35], *Ludisia discolor (Ker Gawl.) Blume (Lan gấm, Orchidaceae)* [36], *Moringa oleifera* Lam. (Chùm ngây, Moringaceae) [37], *Paramignya trimera* (Oliv.) Guill. (Xáo tam phân, Rutaceae) [38], *Nelumbo nucifera* (lotus, sen, Nelumbonaceae) [39], *Ixora dufii*cv. Super king (Trang to, Rubiaceae) [40], *Enydra fluctuans*Lour. (Rau ngổ, Asteraceae) [41], *Eriochloa procera* C.E. Hubb. (cỏ mật, Poaceae) [42], *Litchi chinensis* (Litchi seeds, hạt vải, Sapindaceae) [43], *Pandanus kaida* (dứa kaida, roots, Pandanaceae) [44], *Premna corymbosa* (Burm. F.) Rottl. & Willd. (Vọng cách, leaves, Verbenaceae) [45], *Helicteres hirsuta* Lour. (An xoa, Sterculiaceae)

[46], *Miliusa velutina* Hook. f. & Thomson (Cò sen, Annonaceae) [47], and *Morinda longissima* Y.Z.Ruan (Nhó đông, Fabaceae) [48].

In the *in vitro* model, hepatotoxins used to induce toxicity on human HepG2 cells comprised CCl_4 (2 mM for 2 h [49] or 40 mM for 2 h [50] or 500 μg/mL or CCl_4 (1 %)) [16] or H_2O_2 [51] or t-BHP (2 mM/kg) [17] (Table 1 - SI).

In the *in vivo* model, a variety of hepatotoxic agents (PAR, CCl_4, anti-tubercular drugs, cyclophosphamide, ethanol, D-galactosamine, thioacetamide, t-BHP, or concanavalin) have been used. The most commonly used ones are PAR and CCl_4. Liver damage and recovery from damage are assessed by quantifying serum marker enzymes (ALT, AST, ALP, etc.), bilirubin, bile flow, histopathological changes, and biochemical changes in the liver. Their hepatoprotective effects are evaluated and compared to those of positive controls such as silymarin (70 - 140 mg/kg daily) [28, 35, 36], silibinin (e.g., 100 μM) or glycyrrhizin (e.g., 200 μM) [52].

Tested samples in the form of plant extracts, fractions, or natural products, or compounds were prepared and expressed in *in vivo* experiments with a ratio of g/kg [weight of dry materials (DM) in gram/body weight (BW) in kilogram], of mg/kg [weight of dry extracts (DE)/BW in kilogram] or μM/kg [weight of pure compound (PC) in micromolar/BW in kilogram] and μg/mL [PC in microgram/mL diluted solvent]. The doses tested varied depending on the type of extract (alcohol, water, or fraction). For example, the alcoholic extract of *Ludisia discolor* (leaves), commonly known as "Jewel Orchid", collected in Co To, Tri Ton, An Giang [36] was tested at doses of 100 and 200 mg/kg body weight, while the hot aqueous extract of the roots of *Paramygnia trimera* (Rutaceae), collected in Ninh Van, Khanh Hoa, was tested at a dose of 10 g dry materials/kg body weight.

Most of these tested medicinal plants were reported to possess hepatoprotective effects by decreasing liver enzyme levels, reducing microstructural damage, and improving the liver tissue injury induced by hepatotoxins. Several of them were reported to have remarkable liver protective activity comparable to silymarin. For instance, the methanol extract (10 g/kg) of the roots of *Paramignya trimera* reduced serum AST and ALT levels, total cholesterol content, liver mass, and microstructural damage in liver injury induced by paracetamol in mice. These exhibited liver protective effects similar to silymarin (50 mg/kg body weight, daily) [38].

- Hepatoprotective activity of plant compositions

The chemical constituents of various medicinal plants, such as *Helicteres hirsuta, Phyllanthus* sp., *Cleome* sp., *Hedyotis diffusa, Physalis angulata*, *Moringa oleifera*, *Morinda longissima*, etc., have also been investigated for their hepatoprotective effects. Furthermore, the chemical composition of these medicinal plants, which consists mainly of polyphenolic substances such as flavonoids, coumarins, anthraquinones, triterpenoids, and steroids, demonstrated their hepatoprotective properties (Table 1 - SI).

Helicteres hirsuta Lour. (An xoa) is a traditional Vietnamese medicine for treating chronic liver diseases such as cirrhosis and liver cancer. The collagen deposition areas in the livers of CCl_4-induced liver fibrosis rats treated by methanol and ethanol extracts of *H. hirsuta* were only 3.44 and 5.12 %, respectively, significantly lower than the CCl_4-treated group (19.9 %). The ethanol extract has a definite advantage in the development of food or oral medications for hepatoprotective activity [46]. From this extract, two flavonoids, namely 3,4′,7,8-tetrahydroxyflavone (**1**) and kaempferol-3-*β*-D-(6-*O*-*trans*-*p*-coumaroyl) glucopyranoside (**2**),

were isolated and showed significant hepatoprotective activity against CCl_4-induced toxicity in the HepG2 cell line with an EC_{50} value of 92.20 μg/mL (quercetin: EC_{50} value of 59.57 μg/mL) [53].

Phyllanthus (Phyllanthaceae family) is a genus of over 600 species distributed throughout the tropical and subtropical regions of the world. Extracts of *Phyllanthus acidus* (leaves) [54], *Phyllanthus emblica (*fruit) [55, 56] contain various polyphenols and their glycosides such as quercetin (**3**), isoquercitrin (**9**), quercetin-3-*O*-*α*-L-rhamnoside, rutin, kaempferol-3-O-*β*-rutinoside, kaempferol-3-*O*-*β*-D-glucoside, myricetin-3-O-*β*-rutinoside, hesperitin, and vitamins C and E, which are strong antioxidants and may be responsible for the hepatoprotective activity of their extracts (for example, the leaf extract of *P. acidus* [54], fruit extract of *P. emblica*, etc.). The polyphenols effectively eradicate the toxic effects of reactive drug metabolites. They also improve the protective system by increasing protective antioxidant enzymes represented by SOD, glutathione (GST, GPx, GRs), peroxidase, CAT, etc., which remarkably reduce lipid peroxidation of the hepatocytic membrane and conversely elevate the levels of liver-injured biochemical markers such as ALT, AST, ALP, and BIL [57].

Phyllanthus emblica is commonly known as Indian gooseberry in English, amla in Hindi, or Me rừng, Mận rừng in Vietnamese [2]. Recent studies on the *P. emblica* fruits have also revealed that its ethanol crude extract possesses hepatoprotective activity [55, 56] and is effective in preventing or ameliorating the toxic effects of hepatotoxic agents such as ethanol, PAR, CCl_4, heavy metals, ochratoxins, hexachlorocyclohexane, anti-tubercular drugs, and iron overload [58]. The polyphenols from *P. emblica*, especially tannins and flavonoids, are key elements responsible for major bioactivities [59]. Three extracts of the *P. emblica* fruit extracts (methanol, 70 % ethanol, and 96 % ethanol extracts) increased the percentage of viable HepG2 cells and had the highest hepatoprotective effects against CCl_4-induced toxicity in HepG2 cells with IC_{50} values of 47.68, 60.14, and 56.56 μg/mL, respectively. The methanol extract had the highest liver protective effect, with an IC_{50} value of 47.68 μg/mL [60]. Silymarin, the positive reference control, exhibited a hepatoprotective effect against CCl_4 damage with an $IC_{50} > 100$ μg/mL.

The *Cleome* genus, belonging to the Cleomaceae family, comprises about 170 species. Two species found in Viet Nam, *Cleome viscosa* L. (Màn màn vàng) and *C. chelidonii* L.f. (Màn màn tím) were evaluated for hepatoprotective activity *in vitro and in vivo* [61]. After 72 h of treatment at 100 g/mL, methanol extracts of the stems of *C. chelidonii* and *C. viscosa* increased HepG2 cell viability by approximately 21.4 % and 30 %, respectively. Additionally, they both considerably reduced ALT and AST concentrations at doses of 30 mg/kg and 45 mg/kg as compared to untreated extracts, and their hepatoprotective activities were comparable to those of silymarin. Phytochemical study of these species led to the isolation of the main flavonoids, including visconoside A (**4**) and kaempferol 3-*O*-*β*-D-glucopyranoside 7-*O*-*α*-L-rhamnopyranoside (**5**), which at a concentration of 100 μM displayed significant hepatoprotective activity with prevention percentage values of 66.5 % and 74.2 %, respectively (quercetin: prevention value of 80.3 %). At a concentration of 100 μM, visconoside C (**6**), a flavonol glycoside from *C. viscosa* L., showed hepatoprotective activity against CCl_4-induced hepatotoxicity on HepG2 cells (34.3 %, compared with quercetin control) [49]. These results suggest that *C. chelidonii* and *C. viscosa* are good sources of natural hepatoprotective agents and contribute to understanding the biological activities of *Cleome* species in traditional Vietnamese medicine [61].

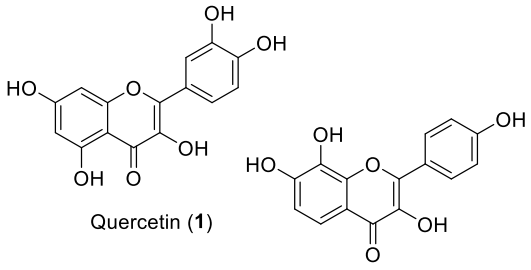
From the whole plant of *Physalis angulata* L., among four isolated phenolic glycosides, rutin (**7**) showed a protective effect with an EC_{50} of 4.56 μg/mL on the primary hepatocytes

freshly isolated from BALB/c mice exposed to 100 μm H_2O_2 to induce oxidative stress [62]. Quercitrin (**8**), a flavonol glycoside from this plant, and many medicinal plants such as *Hypericum perforatum* L., *H. patulum* Thunb., *Bauhinia microstachya* (Raddi) J.F.Macbr., *Rhododendron yedoense* Maxim. Ex Regeland *Alnus firma* had also been proven to exhibit antioxidant and hepatoprotective effects on acute liver hepatitis induced by CCl_4 injection [63].

In addition, the extract of *Moringa oleifera* Lam. (Chùm ngây, Moringaceae) reduced the level of malondialdehyde and increased the level of glutathione, an important endogenous antioxidant in the liver [37, 64]. Isoquercitrin (**9**), isolated from the leaves of this plant, prevented both lipid accumulation and GSH degeneration, and therefore increased the viability of HepG2 cells against CCl_4-induced damage.

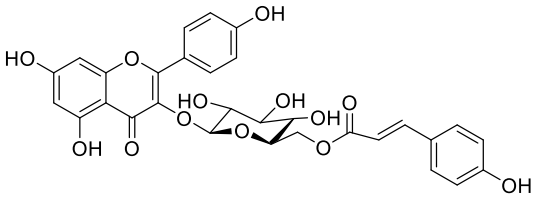
Several flavone glycosides such as ellagic acid 3,3′,4-trimethoxy 4′-*O*-*α*-L-rhamnopyranoside (**10**), 3,4,5-trimethoxyphenol *O*-*β*-D-glucopyranoside (**11**), and tricin (**12**) isolated from the stem barks of *Canararium bengalense* (Burseraceae) also exerted hepatoprotective effects in primary cultured hepatocytes induced hepatotoxicity by hydrogen peroxide (H_2O_2) [51].

Silymarin (**13**) is an isomer flavonolignan composed of the three isomers silibinin, silidianin, and silichristin, extracted from milk thistle (*Silybum marianum* L., Asteraceae, Cúc sữa). At a dose of 0.1 mg/g BW daily, it affected the P450 enzyme system of liver rats by increasing the P450 amount by 25 %, but reducing the activity of aniline hydroxylase and the cytochrome P450 reductase enzymes. Silymarin also had an antioxidant effect on the CCl_4-toxicated rats by activating the SOD and GPx enzymes; consequently, it increased the total antioxidant status by 37.2 % compared to the control group [65]. Silymarin may act as a toxin blocker by preventing the toxins from binding to hepatocyte cell membrane receptors and, therefore, is capable of reducing liver injury caused by chemicals like acetaminophen, CCl_4, radiation, iron overload, phenylhydrazine, alcohol, cold ischemia, and amatoxins from *Amanita phalloides* [66]. The whole extract from the seeds of this plant was found to prevent liver lipid peroxidation, changes in the phospholipid composition of the membranes, hepatic glutathione depletion, and improved functional markers of liver damage [67]. Silymarin (**14**) is able to support liver cells via multiple mechanisms, including inhibiting toxin penetration into hepatic cells by binding to cell membranes and boosting the activity of antioxidant enzymes such as SOD and CAT. The hepatoprotective activity of silymarin can be explained by its antioxidant properties derived from the phenolic nature of flavonolignans [57].



Quercetin (**1**)

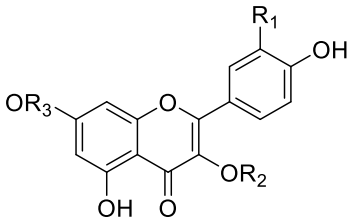
3,4',7,8-tetrahydroxyflavone (**2**)



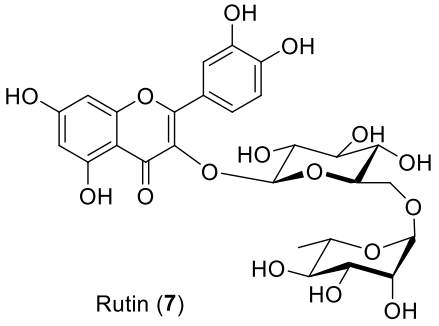
Kaempferol-3-*β*-D-(6-*O*-*trans*-*p*-coumaroyl)glucopyranoside (**3**)

Hedyotis diffusa Willd., whose Vietnamese name is "lưỡi rắn trắng", Rubiaceae family, contains many phytoconstituents, including iridoids, polyphenols, anthraquinones [68], tannins, and triterpenoids [69]. Its methanol extract at the dose of 400 mg/kg body weight showed hepatoprotective activity by effectively reducing the levels of AST ↓ 4.58-fold, ALT ↓ 2.83-fold, and MDA ↓ 9.93-fold and increasing the GSH level ↑ 2.89-fold in the serum of mice of the

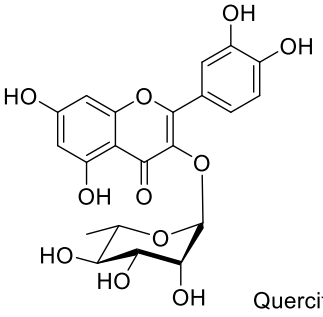
pathological group treated with CCl_4-induced liver damage (CCl_4 (25 % in olive oil), 2.5 mL/kgP mice) (Table 1 - SI). These values were similar to those in the serum of normal mice and the mice treated with silymarin [64].



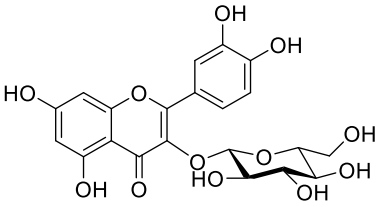
Compound	R_1	R_2	R_3
Visconoside A(**4**)	OH	OH	Glc-(1→3)-4-OAc-Rha
Kaempferol 3-*O*-*β*-D-glucopyranoside 7-*O*-*α*-L-rhamnopyranoside (**5**)	H	Glc	Rha
Visconoside C (**6**)	OH	Rha	[Rha(1→3)]Glc



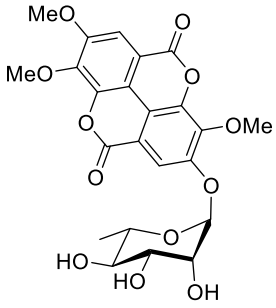
Rutin (**7**)



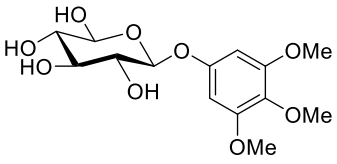
Quercitrin (**8**)



Isoquercitrin (**9**)



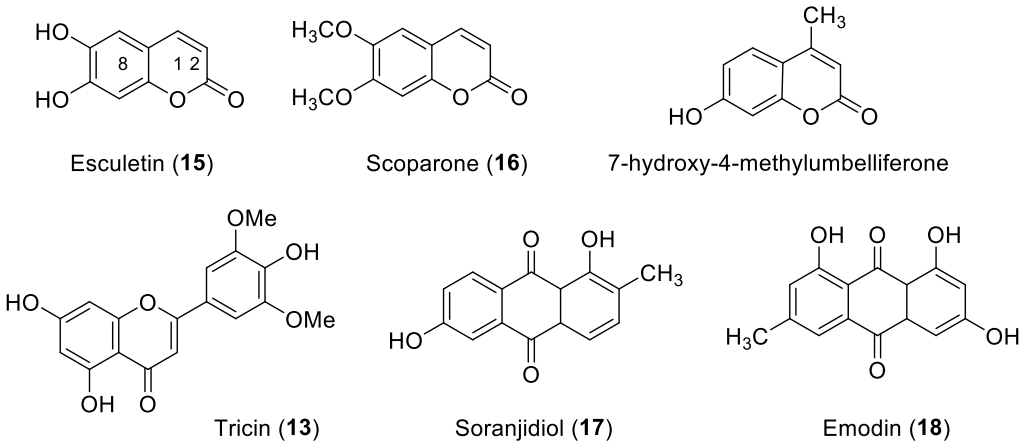
Ellagic acid 3,3',4-trimethoxy 4'-*O*-*α*-L-rhamnopyranoside (**11**)



3,4,5-trimethoxyphenol *O*–*β*-D-glucopyranoside (**12**)

A phytochemical study of *P. trimera* roots (Xáo tam phân) revealed the presence of a variety of coumarins and coumarin glycosides, including simple coumarins (ostruthin, ninhvanins A-B, 6-(6-hydroxy-3,7-dimethylocta-2,7-dienyl)-7-hydroxycoumarin, esculetin, and scopoletin) [70, 71] and biscoumarin monoterpene glycosides such as paratrimerins A-B and paratrimerins J-Y [72, 73]. Both the methanol extract and the water decoction of *P. trimera* roots showed hepatoprotective activities in PAR-induced liver damage mice. Boiling aqueous water extracts, given orally at a dose of 10 g/kg weight, reduced serum AST and ALT concentrations and reduced PAR-induced liver histopathological injury. The liver protective effect of the methanol extract was comparable to that of silymarin (50 mg/kg weight daily) [38]. Oral

administration of esculetin (**15**) (35 mg/kg) and scoparone (**16**) (35 mg/kg) significantly protected rats from CCl_4-induced liver toxicity by elevating the activities of antioxidant enzymes SOD and CAT and decreasing the levels of lipid peroxidation, MDA, GGT and LDH [74]. The results indicated that the number and location of hydroxyl groups on the benzene ring of the coumarin skeleton play an important role in the detoxification functions of coumarins and in the prevention of oxidative stress. According to Atmaca *et al*., esculetin with two hydroxyl groups at the *ortho* position exhibited higher protective activity against CCl_4-induced oxidative stress and hepatotoxicity than scoparone (6,7-dimethoxycoumarin) with two methoxyl groups and 7-hydroxy-4-methylumbelliferone with only one hydroxyl group and one methyl group [74].



In most living things, including humans, coumarin is primarily metabolized by liver enzymes such as CYP2A enzymes into 7-hydroxycoumarin, a nontoxic liver metabolite (Figure 1) [75]. Actually, the roots of *P. trimera* that were found in Ninh Van commune, Khanh Hoa province, and used to treat liver fibrosis contain mostly coumarins and biscoumarin glycosides [38]. Therefore, the hepatoprotective activity of coumarins, particularly the metabolites of different biscoumarin glycosides, merits further investigation.

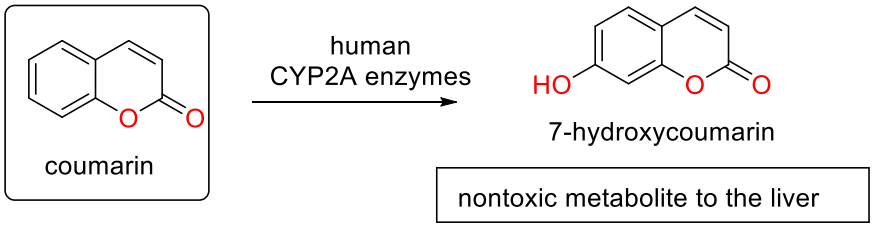


Figure 1. Coumarin fates in the human body to produce nontoxic metabolites in the liver.

It was discovered that a number of medicinal plants containing anthranoids have hepatoprotective properties. The stems and roots of *Morinda longissima* Y.Z.Ruan (Nhó Đông) have long been used by the ethnic Thai people of North Viet Nam (in the provinces of Son La and Lai Chau) to treat liver diseases such as jaundice, liver hepatitis, and cirrhosis. A phytochemical study of this plant extract showed the presence of 21 compounds, mainly anthraquinones and anthraquinone glycosides, including soranjidiol (**17**), rubiadin, rubiadin-3-methyl ether, morindone, morindone-5-methyl ether, morindone-6-methyl ether, lucidin-*ω*-methyl ether, damnacanthal, damnacanthol, lucidin-3-*O*-*β*-primeveroside, and morindone-6-*O*-*β*-

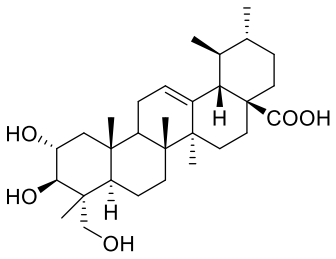
gentibioside, etc. [48, 76 - 79]. Ten different anthraquinone-rich fractions of the ethanolic extract of *M. longissima* roots inhibited 49.1 - 97.2 % and 58.6 - 98.1 % HBV replication in human HBV-infected Hep3B cells by reducing the HBV-DNA level after 48 and 72 h of incubation, respectively [48, 77]. Especially four components HCTN-213, -215, -216, and -219, at a dose of 50 µg/mL and after 72 h of post-treatment, reduced HBV-DNA levels by 90.68, 93.77, 98.11, and 90.48 %, respectively, in comparison to the untreated control group. These decreased HBV-DNA levels were even lower than those produced by lamivudine, the positive control (86.64 %, at a concentration of 50 µM/L in culture medium). The "molongosit" or anthraquinone-rich portion of *M. longissima* roots, served as the principal component of the food supplement "NHÓ ĐÔNG GAN VIỆT™", which is used to support the treatment of HBV and other liver diseases.

Several other free anthraquinones, such as emodin, aloe-emodin, or rhein, showed remarkable hepatoprotective activities by reducing the levels of MDA, ROS, and the liver enzymes ALT and AST and increasing the activity of superoxide dismutase (SOD) in rats. Rhubarb was found to have a significant effect on restraining the evolution of liver fibrosis and cirrhosis [80]. Soranjidiol (**14**) (10 mg/kg) extracted from the roots of *Morinda angustifolia* significantly inhibited the over-formation of MDA in liver damage induced by CCl_4 [81]. Emodin (**15**) (1,3,8-trihydroxy-6-methylanthraquinone) also showed hepatoprotective effects against CCl_4 intoxication as well as D-galactosamine (D-GalN)-induced liver damage [80].

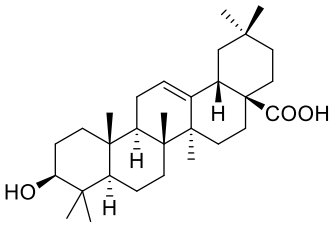
Asiatic acid (**16**), isolated from the aerial parts of *Centella asiatica* (Rau má), demonstrated significant liver-protective activity in mice treated with high paracetamol levels [27]. Oleanolic acid (**17**) from *Wedelia calendulaceae* (Sài đất, Asteraceae) was also discovered to have hepatoprotective properties, reducing liver MDA and serum ALT by 44.8 % and 26.2 %, respectively. These results demonstrated that oleanolic acid contributed to the plant's hepatoprotective activity [82].

Triterpenoids isolated from the seeds of *Combretum quadrangulare* (Trâm bầu, Combretaceace), including lupane-type compounds, 2α,6β-dihydoxybetulinic acid (**18**), 6-β-hydroxyhovenic acid **(19)**, and oleanane-type compounds, arjunolic acid (**20**) and 2α,2β,23-trihydroxyurs-12,18-dien-28-oic acid (**21**) exhibited hepatoprotective properties against D-GalN/TNF-α-induced cell death in primary cultured mouse hepatocytes with IC_{50} values of 60.9, 51.2, 58.4 and 51.0 µM, respectively (IC_{50} value of the control (silibinin) was 29.9 µM). Its MeOH extract (10, 100, and 200 µg/mL) also increased cell survival rates [52, 83].

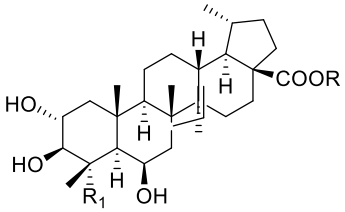
Vietnamese ginseng (*Panax vietnamensis* Ha et Grush, Araliaceae) was first discovered in 1973 in Viet Nam and has been used to treat a variety of diseases. The crude extract and total saponins of this ginseng significantly reduced the liver serum levels of ALT, AST, and GSTα (~50.0 %). The total saponins appeared to be more effective than the crude extract in protecting mice from CCl_4-induced hepatotoxicity [84]. Majonoside R_2 (MR_2) (**22**), the major triterpenoid saponin constituent of Vietnamese ginseng, was found to protect the liver from toxicity induced by D-GalN/TNF-α both *in vivo* and *in vitro*. Significant protection against hepatic apoptosis was observed at doses of 50 and 10 mg/kg in mice (*in vivo*), comparable to that of silymarin (100 mg/kg, p.o.). And at a dose of 200 µM, this compound significantly increased the viability of cultured hepatocyte cells to 89.8 %. MR_2 could inhibit DNA fragmentation and apoptosis induced by TNF-α [85]. Other dammarane-type triterpene saponins isolated from this plant, such as majonoside R_2 (**3**), pseudo-ginsenoside RT_4 (**23**), vinaginsenosides R_1 (**24**), R_2 (**25**), Rh_4 (**26**), and a known sapogenin, protopanaxatriol oxide II (**27**), were found to have hepatoprotective effects on D-galactosamine (D-GalN)/TNF-α-induced cell death in primary cultured mouse hepatocytes, with IC_{50} values of 82.4, 74.8, 47.0, 63.2, 97.0, and 74.0 µM, respectively [86].



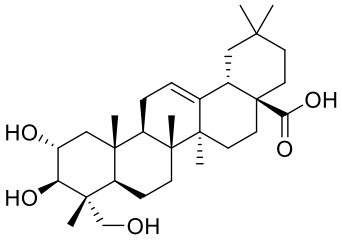
Asiatic acid (**19**)



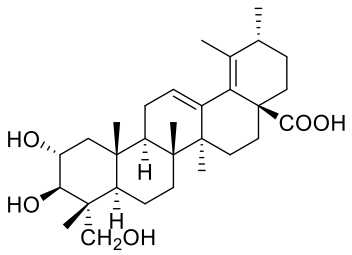
Oleanolic acid (**20**)



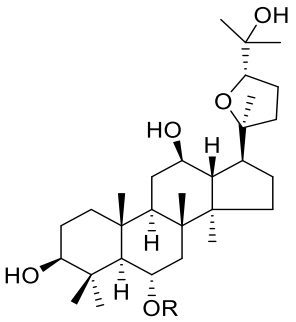
R_1= CH_3, R=H - 2α, 6β-dihydroxybetulinic acid (**21**)
R_1=CH_2OH, R=H - 6β-hydroxhovenic acid (**22**)



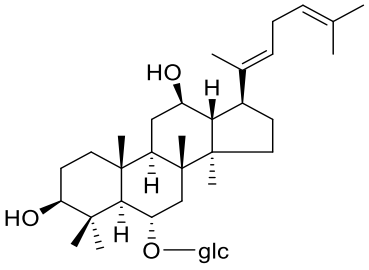
Arjunolic acid (**23**)



2α,3β,23-trihydroxyurs-12,18-dien-28-oic acid (**24**)



R= glc^2-xyl majonoside R_2 (**25**)
R= glc pseudoginsenoside RT_4 (**26**)
R= glc^2-rha vinaginsenoside R_1 (**27**)
^{6}Ac
R = glc^2-xyl vinaginsenoside R_2 (**28**)
^{6}Ac
R = H protopanaxatriol oxide II (**29**)



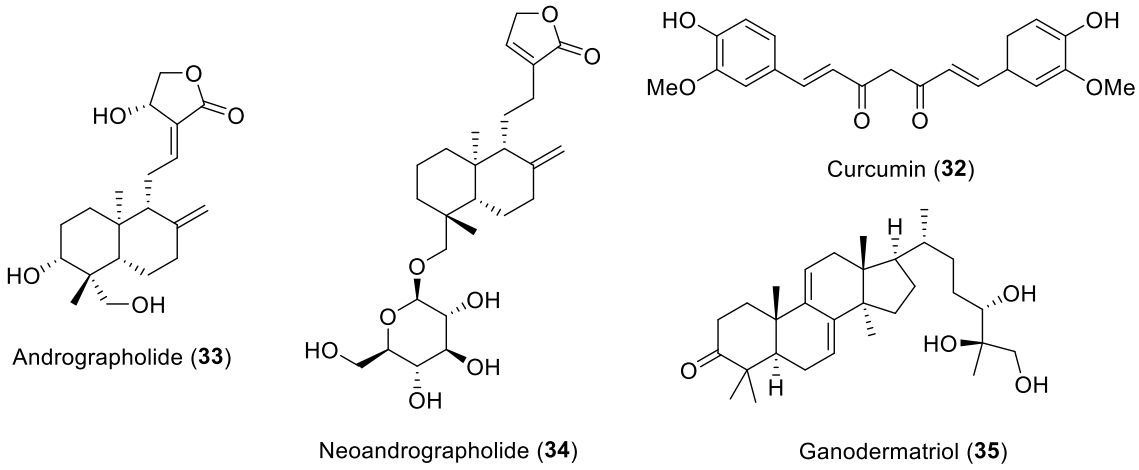
vinaginsenoside Rh_4 (**30**)

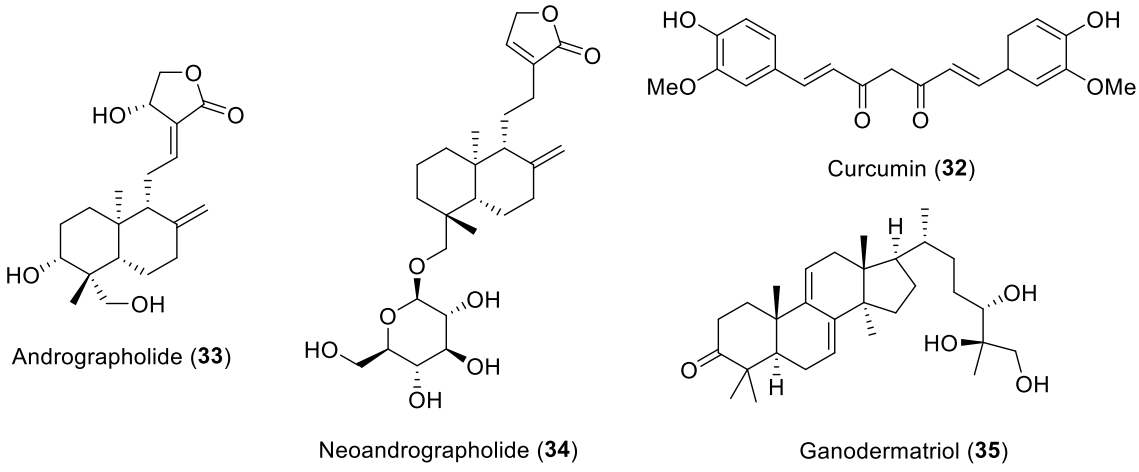
glc: β-D-glucopyranosyl
ara(p): α-L-arabinopyranosyl
ara(f): α-L-arabinofuranosyl
rha: α-L-rhamnopyranosyl
xyl: β-D-xylopyranosyl

Natural curcumin (**28**) isolated from *Curcuma longa* (Zingiberaceae) was found to inhibit CCl_4- and PAR-induced liver damage [30, 87]. Curcumin at doses of 100 or 200 mg/kg/day (intraperitoneal) also showed significant hepatoprotective activity against liver damage caused by aflatoxin B1, λ-cyhalothrin, mercury, and other heavy metals in adult rats [88]. It is not only an antioxidant but also a hepatoprotective compound due to its inhibitory activity of neutralization of ROS (superoxide, peroxyl, and hydroxyl radicals) [46] and RNS (nitric oxide and peroxynitrite) species and against lipid peroxidation [89]. Curcumin ameliorates antioxidant enzymes, including SOD, CAT and glutathione (GSH, GST, GRx, GPx), [88], which inhibit ROS production and prevent OS in liver diseases. Its ability to participate in various cellular and molecular mechanisms like inhibition of pro-inflammatory cytokines and hepatic stellate cells'

activation leads to its remarkable protective and therapeutic effects on oxidative process-associated liver diseases [90]. Curcumin also showed a protective effect on the primary hepatocytes freshly isolated from BALB/c mice exposed to 100 μM H_2O_2 to induce oxidative stress with an EC_{50} of 4.56 μg/mL [62].

Diterpene lactones such as andrographolide (**29**) and neoandrographolide (**30**) (from *Andrographide paniculata*, Xuyên tâm liên) showed hepatoprotective effects against liver injury induced by paracetamolin mice. At an oral dose of 250 mg/kg for 8 consecutive days, the lactone extract significantly showed a protective effect on the hepatotoxicity induced by PAR in mice by reducing the serum AST and ALT levels and the histopathological liver injury [91]. Andrographolide (5 mg/kg) showed no toxic effects on major organs in the healthy mice but could reduce the levels of ALT, AST, and TGF-β_1 as well as block the TGF-β_1/Smad2 and TLR4/NF-κB p50 inflammatory pathways in mice with liver fibrosis induced by CCl_4. This compound was also considered a potential therapeutic strategy for liver fibrosis [92].





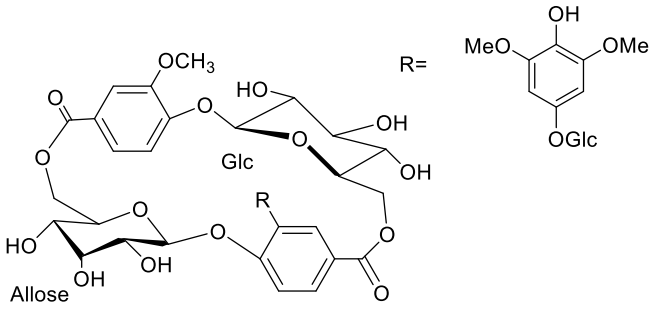
Several mushrooms such as *Phellinus linteus* (Nấm thượng hoàng), *Ophiocordyceps sinensis* (Đông trùng hạ thảo) or *Ganoderma lucidum* (Lingzhi) also showed hepatoprotective activities.

Phellinus linteus, a medicinal mushroom, has been used in oriental countries for a long time to prevent and treat many diseases. When given orally at a dose of 100 mg/kg, the crude polysaccharide from this mushroom reduced the rise in hepatic MDA and increased the level of endogenous hepatic glutathione in mice with cyclophosphamide-induced liver damage [79].

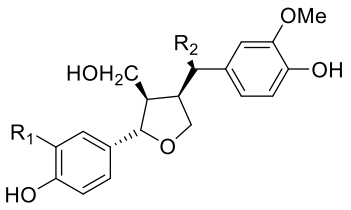
Exopolysaccharides extracted from *Ophiocordyceps sinensis* (0.83 g/kgP or 1.66 g/kgP) showed a significant hepatoprotective effect against CCl_4-induced hepatotoxicity in rats by increasing GSH, decreasing MDA levels, and decreasing serum AST and ALT activities. The improvement in liver tissue demonstrated that EPS significantly lessened the damage that CCl4 caused to the liver [70].

Vietnamese *Ganoderma lucidum* is naturally harvested in the forest, and the fruiting body is developed on the dead ironwood tree. The ethanolic extract of *G. lucidium* demonstrated hepatoprotective activity against cyclophosphamide-induced hepatotoxicity. Supplementation with *G. lucidium* for eight consecutive days after CP injection significantly reduced liver injury compared to CP-treated mice that did not receive *G. lucidium* supplementation. At 230 mg/kgP, hepatoprotective activity was determined by the depletion of MDA and restored by a decrease in endogenous hepatic GSH antioxidant content [93]. Ganodermanotriol (**35**), a sterol found in *G. lucidum*, protected Hepa1c1c7 murine hepatic cells *in vitro* and exhibited hepatoprotective activity *in vivo* in mice against t-BHP-mediated oxidative stimuli by lowering the levels of liver enzymes (ALT, AST), MDA, and glutathione [17].

Macrocyclic glycoside, namely 3,5-dimethoxy-4-hydroxyphenyl-1-*O*-*β*-D-glucopyranoside **(31**) isolated from the wood of *Heliciopsis lobata* (Merr.) Sleumer (cây Đũng), at a concentration of 100 g/mL, was able to reduce the toxic effects of CCl_4 on HepG2 cells and significantly protect cell viability up to 52.25 ± 4.36 % through its radical scavenging capability and limited lipid peroxidative activities [50].



3,5-dimethoxy-4-hydroxyphenyl-1-O-*β*-D-glucopyranoside (**36**)



R_1=OH, R_2 = H Taxiresinol (**37**)
R_1=OMe, R_2=OH (7'R)-7'-Hydroxylariciresinol (**38**)

Taxus yunnanensis Cheng et. I. K. Fu (Thông đỏ, Taxaceae) has been used for a long time for the treatment of kidney problems, diabetic ailments, and various diseases. Two tetrahydrofuran-type lignans from this plant, namely taxiresinol (**32**) and (7′-R)-7′-hydroxylariciresinol (**33**) were found to possess hepatoprotective activity in mice model induced liver toxicity by D-GalN/LPS. These two lignans also protected against TNF-α-mediated hepatocyte apoptosis and therefore increased cell viability [94].

- Hepatoprotective activity of several herbal formulations

Numerous medicinal plants, their formulations, and their derived drugs are being used for the treatment of liver disorders in ethnomedical practice and in traditional medicines in countries

all over the world [95]. Higher plants were reported to be useful in the treatment of liver diseases [1], and many herbal compounds have been demonstrated to possess anti-fibrotic effects or compounds with anti-fibrotic activity (Table 1) and are commonly used in Asian countries to treat liver diseases.

In Viet Nam, many medicinal plants and folk remedies, including *Phyllanthus* sp., *Glycyrrhiza* sp. [66], and *Solanum* sp., are used for the treatment of liver disorders. They are developed as liver supplements, such as:

Megatec Plus F300^{TM} contains extracts of *Solanum hainanense* (100 mg), *Phyllanthus urinaria* L. (80 mg), *Cardus marianus* L. (120 mg), etc. [96]; Tonka^{TM} contains extracts of *Paeonia lactiflora* (420 mg), *Glycyrrhiza uralensis* Fisch. (420 mg), *Phyllanthus urinaria* L. (840 mg), *Codonopsis pilosula* (Franch.) Nannf. (420 mg), etc. [97].

Liverlife^{TM} is a herbal product mixed from different medicinal plants with hepatoprotective activities, including *Phyllanthus urinaria* (Fabaceae, Diệp hạ châu đắng), *Solanum procumbens* (Solanaceae, cà gai leo), *Celastrus hindsii* (Celastraceae, xạ đen), *Panax pseudoginseng* (Araliaceae, tam thất), and silymarin, which have been reported to reduce liver weight index, MDA, AST, ALT, and bilirubin levels in mice's blood in the liverlife-treated group. Liverlife showed good detoxification and hepatoprotective effects and reduced the harmful effects of CCl_4 in an experimental model [98].

The "Lophandanum" extract, a combination of aqueous extracts from two medicinal plants, *Lophatherum gracile* and *Pandanus tectortoius*, at a low dose of 1.5 g/kgP (p.o.) significantly prevented mice's livers from the toxic effects of CCl_4. The extract also showed highly protective activity for the liver tissue structure in comparison to that of untreated groups. However, liver biochemical indices were not influenced [99].

A formula combined from extracts of *Phyllanthus amarus* (Fabaceae, Diệp hạ châu, leaves) (100 mg), *Adenosma bracteosum* (Fabaceae, Nhân trần tía) (80 mg), *Centella asiatica* (Rau má) (40 mg) and *Curcuma longa* (Nghệ, rhizome) (30 mg) at doses of 0.5 - 1.0 g/kg had hepatoprotective effects by reducing ALT and AST levels in plasma and MDA contents in liver mice induced toxicity by CCl_4 [100].

The "AH capsule" is a product made up of twelve plants, including *Strobilanthes cussia (*Chàm tía, leaves, 90 g), *Adenosma caeruleum* (Nhân trần, 20 g), *Atractylodes macrocephata* (Bạch truật, rhizome, 12 g), *Codonopsis pilosula* (Đăng sâm, roots, 12 g), *Astragalus membranaceous* (Hoàng kỳ, roots, 12 g), *Coix lachrymal-jobi* (Ý dĩ, seeds, 12 g), *Citrin aurantia* (Chỉ xác, Fructus, 6 g), *Silvae miltiorrhizae* (Đan sâm, 12 g), *Carthamus tinctorius* (Hồng hoa, 4 g), *Angelica sinensis* (Đương quy, Radix, 12 g), *Alismatis* sp. (Trạch tả, rhizome, 12 g), and *Glycyrrhiza uralensis* (Cam thảo, 6 g). It exhibited protective effects against acute liver injury induced by hepatoxins, either CCl_4 (0.5 mL/kg in olive oil) or PAR (400 mg/kg) in mice [101].

In 2005, V. M. Hung reported that the "Protectiv" capsule with constituents from extracts of *Spirulina platensis* (150 mg) and *Adenosma brateosum* (150 mg) at doses of 250 and 500 mg/kgP/day showed hepatoprotective effects against liver toxicity induced by anti-tuberculosis drugs isoniazid (INH) and rifampicin [102]. "Livcol", a product in granule form prepared as a mixture of medicinal plants including *Phyllanthus urinaria* (Diệp hạ châu), *Lonicera japonica* (Kim ngân hoa), *Chrysanthemum indicum* (Cúc hoa), *Pericarpium Citri Reticulatae* (Trần bì), *Morus alba* (Lá dâu), *Rhemannia gluticosa* (Sinh địa hoàng), *Cassia obtusifolia* (Thảo quyết minh), and *Atractylodes macrocephala* (Bạch truật) has been used for the treatment of chronic

viral hepatitis B and C. A dose of 1 g/kgP/day of "Livcol" demonstrated impressive protective effects against CCl_4-induced liver damage [103].

5. CONCLUSIONS AND FUTURE DIRECTION

The hepatoprotective efficacy of Vietnamese plants tested *in vitro* and *in vivo* in scholarly publications from 1998 to 2022 was evaluated. The paper reviewed 51 medicinal plants, 38 phytoconstituents, and eight hepatoprotective herbal formulations that have been published to protect the liver from the damaging effects of hepatotoxins, most commonly CCl_4 or paracetamol. Their information was shown, including their botanical name, family, part of the plant used, local name, chemical components, the extracts or fractions used, the dosage of extracts, the model used, parameters obtained, histopathology, and the results of studies on each plant's ability to protect the liver. Additionally, it was discussed how these hepatoprotective herbs might work. A number of valuable medicinal plants, including *Paramignya trimera* (Xáo tam phân, stems and roots), *Morinda longissima* (Nhó đông, stems and roots), *Moringa oleifera* (Chùm ngây, leaves), *Helicteres hirsuta* (An xoa, aerial part), and *Cleome* sp. (*Màn màn*, stems, and leaves), all merit further research for the development of herbal liver medications. Numerous naturally occurring bioactive compounds, particularly coumarins, flavonoids, and anthranoids, together with herbal formulations that had various pharmacological effects, were examined and showed promising hepatoprotective capabilities. They might be candidates for further research and development of medicines that could be used to treat liver disorders.

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Declaration of competing interest. The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

***Abbreviation*:** alanine transaminase (ALT), aspartate transaminase (AST), body weight (BW), alkaline phosphatase (ALP), catalase (CAT), effective concentration at 50 % (EC_{50}), gamma-glutamyl transferase (GGT), GSH (glutathione), Glutathion-*S*-transferase (GST), Gluthathion-S-transferase alpha (GSTα), glutathione peroxidase (GPx), glutathione reductase (GRx), hepatitis B virus (HBV), hepatitis C virus (HCV), inhibitory concentration at 50 % (IC_{50}), malondialdehyde (MDA), oxidative stress (OS), paracetamol (PAR) superoxide dismutase (SOD), per oral (p.o.), tert-butyl hydroperoxide (t-BHP), transforming growth factor beta 1 (TGF-β1), Toll-like receptor4/nuclear factor-kB (TLR4/NF-κB).

REFERENCES

1. Joshi D., Keane G., Brind A. - Hepatology at a glance, Wiley Blackwell, 2015, pp. 20-62.
2. Institute for Health Metrics and Evaluation (IHME), GBD 2015: Protocol: global burden of diseases, injuries, and risk factors, http://www.healthdata.org/, 2015 (accessed on 30th Dec. 2022).
3. WCRF, Liver cancer statistics - Liver cancer is the 6th most common cancer worldwide. World Cancer Research Fund International (WCRF), https://www.wcrf.org/cancer-trends/liver-cancer-statistics/, 2020 (accessed on 30th Dec. 2022).

4. Ali S. A., Sharief N., Mohamed Y. -Hepatoprotective Activity of Some Medicinal Plants in Sudan, Evid. Based Complement Alternat. Med. (2196315) (2019) 1-16. https://doi.org/10.1155/2019/2196315.
5. Santillán E. M., Bujaidar E. M., González I. Á., Martínez M. T. S., Salinas J. G., Bautista M., González Á. M., Rubio M. G. L. Y. G., Faisal J. L. A., Morales G., José A - Review of natural products with hepatoprotective effects, World J. Gastroenterol. **20** (40) (2014) 14787-14804.
6. Institute of Medical Materials - Checklist of medicinal plants in Vietnam, Publishing House of Science and Technology, Ha Noi, 2016, pp. 8-16 (in Vietnamese).
7. Loi D. T. - Vietnamese medicinal plants and herbs. Medical Publishing House, Hanoi, 2006, pp. 10-930 (in Vietnamese).
8. Ministry of Health -Vietnamese Pharmacopoeia V. Medical Publishing House, 2017, pp. 1063-1395 (in Vietnamese).
9. Schon H. T., Bartneck M., Borkham-Kamphorst E., Nattermann J., Lammers T., Tacke F., Weiskirchen R. - Pharmacological intervention in hepatic stellate cell activation and hepatic fibrosis, Front. Pharmacol. **7** (33) (2016) 1-22. https://doi.org/10.3389/fphar.2016.00033.
10. Cleverland-Clinic, https://my.clevelandclinic.org/health/diseases/17179-liver-disease, 2022 (accessed on 31st Dec. 2022).
11. Kumar R., Teo E. K., How C. H., Wong T. Y., Ang T. L. - A practical clinical approach to liver fibrosis, Singapore Med. J. **59** (12) (2018) 628-633. https://doi.org/10.11622/smedj.2018145.
12. Bethesda(MD) - LiverTox: Clinical and Research Information on Drug-Induced Liver Injury, National Institute of Diabetes and Digestive and Kidney Diseases. https://www.ncbi.nlm.nih.gov/books/NBK547852/ 2012 (accessed on 31st Dec. 2022).
13. Maes M., Vinken M., Jaeschke H. - Experimental models of hepatotoxicity related to acute liver failure, Toxicol. Appl. Pharmacol., **290** (2016) 86-97. https://doi.org/10.1016/j.taap.2015.11.016.
14. Ahmad F., Tabassum N. - Experimental models used for the study of antihepatotoxic agents, J. Acute Dis., **1** (2) (2012) 85-89. https://doi.org/10.1016/S2221-6189(13)60021-9.
15. Kumar E., Susmitha K., Swathy B., Ramu E., Venkatesh B. - A review on liver disorders and screening models of hepatoprotective agents, Int. J. Allied. Med. Sci. Clin. Res. **2** (2) (2014) 136-150.
16. Men T. T., Yen N. D. H., Trang D. T. X. - Study of toxicity and hepatoprotective activity of methanol extracts of some plant leaves on HepG2 cell line, J. Sci. - Dong Thap Uni. **33** (2018) 86-89 (in Vietnamese).
17. Ha D. T., Oh J., Khoi N. M., Dao T. T., Dung L. V., Que T. N. D., Lee S. M., Jang T. S., Jeong G.-S., Na M. - *In vitro* and in vivo hepatoprotective effect of ganodermanontriol against t-BHP-induced oxidative stress, J. Ethnopharmacol. **150** (2013) 875-885. https://doi.org/10.1016/j.jep.2013.09.039.
18. Kothari P., Andhale A., Waghmare S. - A review: herbal medicines and screening models for hepatoprotective agents, World J. Pharmaceut. Res. **10** (8) (2021) 1373-1386.

19. Korver S., Bowen J., Pearson K., Gonzalez R., French N P. K., Jenkins R, Goldring C. - The application of cytokeratin-18 as a biomarker for drug-induced liver injury, Arch. Toxicol. **95** (11) (2021) 3435-3448. https://doi.org/10.1007/s00204-021-03121-0. .
20. Maqbool M., Dar M. A., Rasool S., Bashir R., Khan M. - Hepatotoxicity and Hepatoprotective agents: A Mini review, PharmaTutor **7** (9) (2019) 34-40. https://doi.org/10.29161/PT.v7.i9.2019.34.
21. Jaeschke H., Fisher M. A., Lawson J. A., Simmons C. A., Farhood A., Jones D. A. - Activation of caspase 3 (CPP32)-like proteases is essential for TNF-alpha-induced hepatic parenchymal cell apoptosis and neutrophil-mediated necrosis in a murine endotoxin shock model, J. Immunol. **160** (1998) 3480-3486. PMID: 9531309.
22. Wang H. X., Liu M., Weng S. Y., Li J. J., Xie C., He H. L., Guan W., Yuan Y. S., Gao J. - Immune mechanisms of Concanavalin A model of autoimmune hepatitis, World J. Gastroenterol. **18** (2) (2012) 119-125. https://doi.org/10.3748/wjg.v18.i2.119.
23. Thai N. P., Trung L. V., Hai N. K., Huynh L. - Protective efficacy of *Solanum hainanense* Hance during hepatotoxicity in male mice with prolonged and small oral doses of Trinitrotoluene, J. Occup. Health **40** (1998) 276-278.
24. Constable P. D., Hinchcliff K. W., Done S. H., Grünberg W. - 9-Diseases of the Liver, W. B., Saunders Elsevier, Missouri, 2017, pp. 622-656.
25. Mas-Bargues C., Escrivá C., Dromant M., Borrás C., Viña J. - Lipid peroxidation as measured by chromatographic determination of malondialdehyde. Human plasma reference values in health and disease, Arch. Biochem. Biophys. **709** (2021) 108941. https://doi.org/10.1016/j.abb.2021.108941.
26. Toan T. Q. - Hepatoprotective effects of *Aganope belansae* (Gagnep.), J. Vietnam Med., **5** (2013) 457-461 (in Vietnamese).
27. Nhu V. T. Q., Ha L. T. T., Thao T. T. P., Loc T. V. - Hepatoprotective activity of asiatic acid isolated from *Centella asiatica* (L.) Urban, J. Chem. **54** (5) (2016) 540-541 (in Vietnamese). https://doi.org/10.15625/0866-7144.2016-00361.
28. Oanh D. T., Ky P. T., Thong N. T., Anh P. T. V. - Evaluation of hepatoprotective and antioxidant activities of *Chloranthus japonicus in vivo*, J. Pharm. **462** (36) (2014) 26-29 (in Vietnamese).
29. Huy N. Q., Thuong L. Q., Huy H. X., Hung T. Q., Huong P. T. M., Viet T. Q., Hoa, Le Thi Phuong - Hepatoprotective activities of *Colocasia esculenta* (L.) Schott in mice model with liver injury induced by Paracetamol, J. Sci. Med. Pharm. Sci. **36** (3) (2020) 66-75 (in Vietnamese). https://doi.org/10.25073/2588-1132/vnumps.4244.
30. Tung B. T., Hai N. T., Loi V. D., Thu D. K. - Hepatoprotective effects of phytosome curcumin in mice model induced liver toxicity by paracetamol, J. Pharm. **485** (2016) 22-26 (in Vietnamese).
31. Thu N. T. A., Thong N. T., Anh P. T. V., Thu N. B. - Effect of Mui mac (*Desmodium triquetum*) in mouse liver injury induced by Paracetamol, Vietnam J. Sci. Technol. **177** (01) (2018) 203-207 (in Vietnamese).
32. Yen L. T. H., Anh P. T. V. - Hepatoprotective activity of *Eclipat prostrata* L., Asteraceae extract in paracetamol - induced toxicity in mice, J. Pharmacol. Sci. **431** (2012) 13-16 (in Vietnamese).

33. Duc T. V., Tung T. T., Ha N. T. T. - Hepatoprotective and antioxidant effect of aqueous extracts of Hong chi da lat (*Ganoderma lucidum*) dl1 on mice with liver injury model induced by paracetamol, Med. Res. J. **126** (2) (2020) 34-41 (in Vietnamese).
34. Hung T. M., Diep L. T., Thoai N. D., Cuong H. D. - Hepatoprotective effect of Lingzhi extract on rifampicin and isoniazid-induced hepatotoxicity, J. Med. Materials **12** (5) (2007) 151-155 (in Vietnamese).
35. Nhung B. T. Q., Cuc H. T., Huyen N. T. T., Cuong L. N. - Hepatoprotective activity of *Hovenia dulcis* Thunb. on paracetamol induced liver toxicity in mice, J. Vietnam Med. **486** (1&2) (2020) 181-185 (in Vietnamese).
36. Nguyen C. K., Do T. H. T., Nguyen L. A. T. - Antioxidant and hepatoprotective activity of *Lusidia discolo*r extract in An Giang, J. Agric. Sci. Tech. Vietnam **04** (2021) 131-139 (in Vietnamese).
37. Huong N. T. T., Minh N. H., Nhan N. L., Trieu L. H., Dan N. T. N. - Experimental study on the invigoration and hepatoprotective effects of *Moringa oleifera* leaves, J. Med. HCMC **20** (6) (2016) 222-227 (in Vietnamese).
38. Cuong N. M., Huong T. T., Khanh, Pham Ngoc, Ha V. T., Cuc N. T., Thao D. T. - Hepatoprotective activity of *Paramignya trimera* roots on mice model with liver injury induced by paracetamol, Vietnam J. Sci. Technol. **54** (1) (2016) 37-45 https://doi.org/10.15625/0866-708X/54/1/5666.
39. Hong N. N., Ngan T. T. K. - Phytochemical, free radical scavenging effects and antioxidant activity of lotus leaves (*Nelumbo nucifera* Gaertn.), J. Agr. Agricult. Dev. **23** (2018) 74-80 (in Vietnamese).
40. Dinh P. K., Tuan N. T., Trang D. T. X. - Antioxidant, anti-inflammatory and hepatoprotective activities on carbon tetrachloride - induced hepatic damage in mice of *Ixora duffii* leaf extract, J. Biol. **41** (1) (2019) 117-128. https://doi.org/10.15625/0866-7160/v41n1.12734.
41. Duy H. A., Giau L. T. N., Linh B. M. - Evaluation of acute toxicity and hepatoprotective activity of extract of *Enhydra fluctuans* Lour. (Asteraceae) in mice, J. Med. Pharm. Can Tho Uni. **9** (2017) 41-47.
42. Loan L. T. K., Bang B. T., Thu L. T. V., Thanh V. T. N., Thong N. T., Chi D. T. K. - The hepatoprotective effects of *Eriochloa procera* (Retz.) C. Hubb., J. Med. Materials **12** (3+4) (2007) 111-114 (in Vietnamese).
43. Anh P. T., Nga H. T., Tuyen N. M. - Antioxidant and hepatoprotective activities of the ethanol extract of Litchi seeds, J. Pharm. **457** (2014) 22-25 (in Vietnamese).
44. Hoa P. T., Na V. T. H. - Antioxidant and hepatoprotective effects of water extract from the roots of pineapple Kaida (*Pandanus kaida* Kurz) in mice model, J. Pharm. **447** (2013) 21-24 (in Vietnamese).
45. Hang N. T. B., Ky P. T., Thong N. T., Anh P. T. V. - Analgesic effects and antioxidant activities of Vong cach plant (*Premma integrifolia* (L.) Verbenaceae) in term of liver-protective effects on paracetamol - poisoned mice, J. Pharm. **405** (2010) 26-29 (in Vietnamese).
46. Thang H. D., Hien T. T. T., Viet N. D., Anh H. L. T., Do T. T., Vinh L. B., Yang S. Y., Dan G., Anh L. T. - Hepatoprotective effects of extract of *Helicteres hirsuta* Lour. on

liver fibrosis induced by carbon tetrachloride in rats, Appl. Sci. **11** (18) (2021) 8758-8768. https://doi.org/10.3390/app11188758.

47. Trang D. T. X., Hieu B. L. T., Linh T. C., Danh L. T., Tuan N. T. - Antioxidant and hepatoprotective potentials of *Miliusa velutina* stem bark extract, Sci. Tech. Dev. J. Nat. Sci. **4** (3) (2020) 633-642.
48. Toan N. L., Chung Đ. T., Huong T. T., Tram N. C. T., Son N. T., Khanh P. N., Hung N. T., Dong L. C., Son H. A., Cuong N. M. - Inhibition of hepatitis B virus replication by products from the roots of the *Morinda longgissima in vitro*, Vietnam Med. J. **1** (2016) 32-36 (in Vietnamese).
49. Phat N. T., Luan T. C., Hung V. C., Tuoi D. T. H., Dung L. T., Tri M. D., Minh P. N. - Flavonoids with hepatoprotective activity from the leaves of *Cleome viscosa* L., Nat. Prod. Res. **31** (22) (2017) 2587-2592. https://doi.org/10.1080/14786419.2017.1283497 (in Vietnamese).
50. Trung B. V., Thao D. T., Anh D. H., Kiem P. V., Viet P. H. - Antioxidant and hepatoprotective activity of phenyl glycosides isolated from *Heliciopsis lobata*, Nat. Prod. Commun. **15** (7) (2020) 1-7. https://doi.org/10.1177/1934578X20946255.
51. Hoang L. T., Ha D. T., Minh C. T. A., Kim T. H., Kiem P. V., Thuan N. D., Na M. - Constituents from the stem barks of *Canarium bengalense* with cytoprotective activity against hydrogen peroxide-induced hepatotoxicity, Arch. Pharm. Res. **35** (1) (2012) 87-92. https://doi.org/10.1007/s12272-012-0110-2.
52. Banskota A. H., Tezuka Y., Adnyana I. K., Xiong Q., Hase K., Tran K. Q., Tanak K., Saiki I., Kadota S. - Hepatoprotective effect of *Combretum quadrangulare* and its constitutents, Biol. Pharm. Bull. **23** (4) (2000) 456-460. https://doi.org/10.1248/bpb.23.456.
53. Tien H. L. N., Win P. H. X., Tri L. V. - Hepatoprotective activity of secondary metabolites from *Helicteres hirsuta* collected in Gia Lai, J. Sci. Tech. Uni. Sci., Hue Uni. **2** (2021) 65-74 (in Vietnamese).
54. Tram N. C. T., Son N. T., Nga N. T., Phuong V. T. T., Cuc N. T., Phuong D. T., Truan G., Cuong N. M., Thao D. T. - The hepatoprotective activity of a new derivative kaempferol glycsoide from the leaves of Vietnamese *Phyllanthus acidus* (L.) Skeels, Med. Chem. Res. **26** (2017) 2057-2064. https://doi.org/10.1007/s00044-017-1914-x.
55. Duong N. T., Uyen N. T., Vinh P. D., Son N. T., Ha L. V., Ha D. T., Thuy P. T. - Hepatoprotective effect of standardized dry extract from *Phyllanthus emblica* L. fruits on hepatotoxicity model induced by carbon-tetracloride in mice, J. Med. Materials **24** (2) (2019) 108-112 (in Vietnamese).
56. Jose J., Kuttan R. - Hepatoprotective activity of *Emblica officinalis* and Chyavanaprash, J. Ethnopharmacol. **72** (1-2) (2000) 135-140. https://doi.org/10.1016/s0378-8741(00)00219-1.
57. Madrigal S. E., Madrigal B. E., Álvarez G. I., Sumaya M. M. T., Gutiérrez S. J., Bautista M., Morales G. Á., García L., González R. M., Morales G. J. A. *et. al*. - Review of natural products with hepatoprotective effects, World J. Gastroenterol. **20** (40) (2014) 14787-14804. https://doi.org/10.3748/wjg.v20.i40.14787.
58. Thilakchand K., Mathai R., Simon P., Ravi R., Baliga-Rao M., Baliga M. - Hepatoprotective properties of the Indian gooseberry (*Emblica officinalis* Gaertn): a review, Food Funct. **4** (10) (2013) 1431-1441. https://doi.org/10.1039/c3fo60237k.

59. Yadav S., Singh M., Singh P., Kumar V. - Traditional knowledge to clinical trials: A review on therapeutic actions of *Emblica officinalis*, Biomed. Pharmacother. **93** (2017) 1292-1302. https://doi.org/10.1016/j.biopha.2017.07.065.
60. Cuong N. M., Khanh P. N., Huong T. T., Ha V. T., Anh H. T. N., Thao D. T., Cuc N. T., Cuong T. D. - Antioxidant, lipid peroxidation inhibitory activities, and hepatoprotective effect of extracts of *Phyllanthus emblica* L. fruits, Vietnam J. Sci. Technol. **60** (3) (2022) 410-423. https://doi.org/10.15625/2525-2518/16110.
61. Minh P. N., Tuoi D. T. H., Tuyen N. L. T., Tuan N. T., Luan N. Q., Duc T. T., Hien N. Q., Chi H. B. L., Ky N. D. X., Dat B. T., Tri M. D., Phat N. T. - Hepatoprotection and phytochemistry of the Vietnamese herbs *Cleome chelidonii* and *Cleome viscosa* stems, Hindawi J. Chem. **2021** (2021) 1-8. https://doi.org/10.1155/2021/5578667.
62. Anh H. L. T., Dung D. T., Tuan D. T., Hung T. Q., Yen P. H., Quang T. H., Nhiem N. X., Minh C. V., Yen D. T. H., Kiem P. V. - Hepatoprotective effects of phenolic glycosides from the methanol extract of *Physalis angulata*, Vietnam J. Sci. Technol. **55** (2) (2017) 161-167. https://doi.org/10.15625/0866-708X/55/2/8527.
63. Bang B. T., Huong N. T. T., Tien T. M., Bich L. K., Anh H. V. - Quercitrin - flavon glycosid isoated from *Hypericum patulum* Thunb. ex Murray with hepatoprotective activity, J. Pharm. **378** (2007) 40-42 (in Vietnamese).
64. Dinh P. K., Huyen V. T. M., My T. D. H., Diem L. T., Linh T. C., Tuan N. T., Trang D. T. X. - Study on some biological activities of methanol extract of white snake's tongue - *Hedyotis diffusa* Willd., J. Sci. Can Tho Uni. (Topics: Natural Science) **56** (2) (2020) 103-114 (in Vietnamese).
65. Nga N. T. T., Tuyen D. T., Viet D. V., Dao N. T. N. - The liver protection and antioxidant effects of silymarin extracted from milk thistle *Silybum marianum* (L.) Gaertn., J. Biol. **28** (3) (2006) 88-92. https://doi.org/10.15625/0866-7160/v28n3.5332.
66. Dhiman R. K., Chawla, Y.K. - Herbal medicines for liver diseases, Dig. Dis. Sci. **50** (2005) 1807-1812. https://doi.org/10.1007/s10620-005-2942-9.
67. Dehmlow C., Erhard J., de Groot H. - Inhibition of Kupffer cell functions as an explanation for the hepatoprotective properties of silibinin, Hepatology **23** (4) (1996) 749-754. https://doi.org/10.1053/jhep.1996.v23.pm0008666328.
68. Dung L. K., Sung T. V., Dien P. G. - Two anthraquinones isolated from *Hedyotis corymbosa* and *Hedyotis diffusa*, Vietnam J. Chem., **40** (3) (2002) 66-68.
69. Chen R., He J., Tong X., Tang L., Liu M. - The *Hedyotis diffusa* Willd. (Rubiaceae): a Review on phytochemistry, pharmacology, quality control and pharmacokinetics, Molecules **21** (6) (2016) 710. https://doi.org/10.3390/molecules21060710. .
70. Huong T. T., Ha V. T., Cuong T. D., Son N. T., Toan T. Q., Anh H. T. N., Tram N. T. T., Woo S. H., Kim Y. H., Cuong N. M. - New constituents from the roots and stems of *Paramignya trimera*, Nat. Prod. Commun. **14** (6) (2019) 1-5. https://doi.org/10.1177/1934578x19861015
71. Cuong N. M., Duc H. V., Tai N. V., Khanh P. N., Ha V. T., Huong T. T., Nhut N. D. - Initial study of chemical compositions of *Paramignya trimera* (Rutaceae), Vietnam J. Chem., **51** (3) (2013) 292-296.
72. Quan K. T., Park H. B., Yuk H., Lee S. J., Na M. - Paratrimerins J–Y, Dimeric coumarins isolated from the stems of *Paramignya trimera*, J. Nat. Prod. **84** (2) (2021) 310-326. https://doi.org/10.1021/acs.jnatprod.0c00978.

73. Cuong N. M., Huong T. T., Khanh P. N., Tai N. V., Ha V. T., Son N. T., Tai B. H., Kim Y. H. - Paratrimerins A and B, two new dimeric monoterpene-linked coumarin glycosides from the roots and stems of *Paramignya trimera*, Chem. Pharm. Bull. **63** (11) (2015) 945-949. https://doi.org/10.1248/cpb.c15-00336.

74. Atmaca M., Bilgin H. M., Obay B. D., Diken H., Kelle M., Kale E. T. - The hepatoprotective effect of coumarin and coumarin derivates on carbon tetrachloride-induced hepatic injury by antioxidative activities in rats, J. Physiol. Biochem. **67** (4) (2011) 569-576. https://doi.org/10.1007/s13105-011-0103-5.

75. Vassallo J. D., Hicks S. M., Daston G. P., Lehman-McKeeman, Lois D. - Metabolic detoxification determines species differences in coumarin-induced hepatotoxicity, Toxicol. Sci. **80** (2) (2004) 249-257. https://doi.org/10.1093/toxsci/kfh162.

76. Cuong N. M., Huong T. T., Khanh P. N., Tai N. V., Van D. T., Ha V. T., Phuong N. T., Ha L. M., Long P. Q. - Anthraquinon compounds from the roots of *Morinda longissima*, J. Drug Qual. Cont. **3A** (2014) 3-8 (in Vietnamese).

77. Chung D. T., Huong T. T., Tram N. C. T., Van D. T., Ha V. T., Hung N. T., Dong L. C., Son H. A., Lenon G. B., Cuong N. M., Toan N. L. - Anti-hepatitis B virus activity of components from the roots of *Morinda longissima in vitro* model, J. Milli. Pharmaco-Med. **1** (2016) 85-93 (in Vietnamese).

78. Cuong N. M., Huong T. T., Son N. T., Cuong T. D., Van D. T., Khanh P. N., Ha V. T., Tram N. C. T., Long P. Q., Kim Y. H. - Morinlongosides A-C, two new naphthalene glycoside and a new iridoid glycoside from the roots of *Morinda longissima*, Chem. Pharm. Bull. **64** (8) (2016) 1230-1234. https://doi.org/10.1248/cpb.c15-01039.

79. Cuong N. M., Long P. Q., Sơn N. T., Van D. T., Khanh P. N., Cuong T. D., Ha V. T., Tram N. C. T., Viet D. Q., Huong T. T. - Phenylethanoid glucoside and anthraquinone compounds from *Morinda longissima* Y. Z. Ruan roots, Vietnam J. Chem. **54** (2) (2016) 133-138.

80. Zhao Y. L., Wang J. B., Zhou G. D., Shan L. M., Xiao X. H. - Investigations of free anthraquinones from Rhubarb against α-naphthylisothiocyanate-induced cholestatic liver injury in rats, Basic Clin. Pharmacol. Toxicol. **104** (6) (2009) 463-469. https://doi.org/10.1111/j.1742-7843.2009.00389.x.

81. Chen R. R., Liu J., Chen Z., Cai W. J., Li X. F., Lu C. L. - Anthraquinones extract from *Morinda angustifolia* Roxb. root alleviates hepatic injury induced by carbon tetrachloride through Inhibition of hepatic oxidative stress, Evid. Based Complement Alternat. Med. **20** (2020) 1-9. https://doi.org/10.1155/2020/9861571.

82. Hoa N. T. X., Thanh P. V. - Liver protective effects of oleanolic acid isolated from *Wedelia calendulacea* (L.), J. Med. Materials **5** (2003) 146-149 (in Vietnamese).

83. Adnyana I. K., Tezuka Y., Banskota A. H., Tran K. Q., Kadota S. - Three new triterpenes from the seeds of *Combretum quadrangulare* and their hepatoprotective activity, J. Nat. Prod. **64** (2001) 360-363. https://doi.org/10.1021/np000486x.

84. Nguyen T. D., Villard P. H., Barlatier A., Elsisi A. E., Jouve E., Duc N. M., Sauze C., Durand A., Lacarelle B. - *Panax vietnamensis* protects mice against carbon tetrachloride-induced hepatotoxicity without any modification of CYP2E1 gene expression, Planta Med. **66** (8) (2000) 714-719. https://doi.org/10.1055/s-2000-9603.

85. Quan L. T., Adnyana I. K., Tezuka Y., Harimaya Y., Saiki I., Kurashige Y., Qui T. K., Kadota S. - Hepatoprotective effect of majonoside R2, the major saponin from Vietnamese ginseng (*Panax vietnamensis*), Planta Med., **68** (2002) 402-406. https://doi.org/10.1055/s-2002-32069.

86. Quan L. T., Adnyana I. K., Tezuka Y., Nagaoka T., Qui T. K., Kadota S. - Triterpene saponins from Vietnamese ginseng (*Panax vietnamensis*) and their hepatocytoprotective activity, J. Nat. Prod. **64** (2000) 456-461. https://doi.org/10.1021/np000393f.

87. Thanh V. T. N., Mai N. T. T. - Hepatoprotective effect of curcuminoid *in vivo*. J. Med. Res. **48** (2) (2007) 22-27.

88. Farzaei M. H., Zobeiri M., Parvizi F., El-Senduny F. F., Marmouzi I., Coy-Barrera E., Naseri R. - Curcumin in liver diseases: a systematic review of the cellular mechanisms of oxidative stress and clinical perspective, Nutrients **10** (2018) 855-883. https://doi.org/ 10.3390/nu10070855.

89. Sreejayan R. M. N. - Nitric oxide scavenging by curcuminoids, J. Pharm. Pharmacol. **49** (1) (1997) 105-107. https://doi.org/10.1111/j.2042-7158.1997.tb06761.x.

90. Ak T., Gülçin I. - Antioxidant and radical scavenging properties of curcumin, Chem. Biol. Interact. **174** (1) (2008) 27-37. https://doi.org/10.1016/j.cbi.2008.05.003.

91. Thanh V. T. N., Diep T. T., Tranh D. D., Anh P. T. V. - Evaluation on the protection effect and the antioxidant activity of the lactonne compound from Xuyen tam lien (XTL) in the liver damage induced by paracetamol in mice experiment, J. Res. Med. **80** (3C) (2012) 104-109.

92. Lin L., Li R., Cai M., Huang J., Huang W., Guo Y., Yang L., Yang G., Lan T., Zhu K. - Andrographolide ameliorates liver fibrosis in mice: involvement of TLR4/NF-κB and TGF-β1/Smad2 signaling pathways, Oxid. Med. Cell. Longev. (2018) 1-11. https://doi.org 10.1155/2018/7808656.

93. Ngoc P. H., Son H. L., Trung P. V. - Hepatoprotective activity of *Ganoderma lucidium* (Curtis) P. Karst against cyclophosphamide-induced liver injury in mice, Cogent. Biol., **2** (1267421) (2016) 1-9. https://doi.org/10.1080/23312025.2016.1267421.

94. Nguyen N. T., Banskota A. H., Tezuka Y., Le Tran Q., Nobukawa T., Kurashige Y., Sasahara M., Kadota S. - Hepatoprotective effect of taxiresinol and (7'R)-7'-hydroxylariciresinol on D-galactosamine and lipopolysaccharide-induced liver injury in mice, Planta Med. **70** (1) (2004) 29-33. https://doi.org/10.1055/s-2004-815451.

95. Sun T. Y., Li J. S., Chen C. - Effects of blending wheatgrass juice on enhancing phenolic compounds and antioxidant activities of traditional kombucha beverage, J. Food Drug Anal. **23** (4) (2015) 709-718. https://doi.org/10.1016/j.jfda.2015.01.009.

96. Ethnic medicine, - Megatec Plus F300 (https://www.thuocdantoc.org/megatec-plus-f300.html) 2022 (accessed 5th Aug 2022).

97. Tonka, https://nhatnhat.com/tonka.html 2022 (accessed on 30th Dec. 2022).

98. Long N. V., Anh V. T., Hung T. T. - Evaluation of the hepatoprotective effects of liverlife, Vietnam Med. J. **2** (2016) 173-177 (in Vietnamese).

99. Lien N. T., Hang N. T., Huong N. T. K., Hung T. V., Son D. C. - Initial study of hepatoprotective effects of lophandanum extract *in vivo*, J. Pharm. **461** (2014) 34-39 (in Vietnamese).

100. Duc N. M., Huong N. T. T., Phuong H. T. Y., Thong L. H. -Hepatoprotective effects of a combined formula from extracts of *Phyllanthus amarus, Adenosma bracteosum, Centella asiatica* and *Curcuma longa*, J. Med. Materials **12** (3+4) (2007) 115-120 (in Vietnamese).

101. Thanh V. T. N., Thanh D. K., Yen P. T. C. - In vivo experiments of liverprotective effects and acute toxicity of AH herbal product, J. Vietnam Med. **12** (2006) 173-180 (in Vietnamese).

102. Hung V. M. - Hepatoprotective effect of Protecliv *in vivo.* Exp. Med. **10** (2005) 17-20 (in Vietnamese).

103. Hien T. L. V., Truong C. Q., Hoa T. T. P., Phuong D. T. B., Phong T. T. - Hepatoprotective effects of livcol granule in the liver of CCl_4-induced poisoning mice, Res. J. Vietnam Trad. Med. **17** (2007) 25-32 (in Vietnamese).