



Original Article

Evaluation of the Hepatoprotective Effect of *Phyllanthus niruri* in Paracetamol and Isoniazid-Induced Hepatotoxicity in Albino Wistar Rats

Deepshikha Sarswat¹, Md. Quamuddin², Saurabh Nimesh², Anshul Pandey³, Abhimanyu Anand⁴ and Raj Kumar Mandal^{5,*}

¹Research Scholar, Metro College of Health Science and Research, Greater Noida, Knowledge park-3, Uttar Pradesh-201310, India.

²Department of Pharmacology, Metro College of Health Science and Research, Greater Noida, Uttar Pradesh-201310, India.

³Department of Pharmacy, Shakuni Choudhary College of Health and Sciences, Tarapur, Munger, Bihar – 813201, India.

⁴Department of Pharmacology, Global Research Institute of Pharmacy, Nachraun, Yamuna Nagar, Haryana-135133, India.

⁵Chitkara University, Chandigarh-Patiala National Highway, Rajpura, Punjab 140401, India.

ARTICLE INFO

Received 16 September 2026

Revised 17 October 2026

Available Online 20 October 2026

Keywords:

Silymarin

Phyllanthus niruri

Hepatoprotective activity

Isoniazid-induced hepatotoxicity

ABSTRACT

The liver performs several vital physiological and biochemical functions, including metabolism, detoxification, protein synthesis, nutrient regulation and bile secretion. Exposure to hepatotoxic drugs, chemicals, alcohol and infections can impair hepatic function and cause liver injury. Paracetamol (PCM) overdose and isoniazid (INH) therapy are important causes of drug-induced hepatotoxicity, mainly through oxidative stress and the formation of reactive metabolites. The present study evaluated the hepatoprotective activity of the ethanolic leaf extract of *Phyllanthus niruri* against PCM- and INH-induced hepatotoxicity in experimental rats. In the PCM model, hepatic injury was induced by oral administration of PCM (750 mg/kg) on the 8th and 16th days of the 21-day experimental period. In the INH model, isoniazid (50 mg/kg) was administered orally for 28 days, followed by treatment with the plant extract for 15 days.

The extract was evaluated at two dose levels and its effects were compared with the standard hepatoprotective drug, silymarin. Treatment with the ethanolic leaf extract significantly improved the altered biochemical parameters in a dose-dependent manner. It reduced elevated serum liver enzymes and increased serum albumin and total protein levels. The higher dose showed greater protective activity, with significant differences observed compared with the toxic control groups.

Histopathological findings further supported the biochemical results, showing improvement in hepatic architecture following extract treatment. Overall, the findings indicate that the ethanolic leaf extract of *P. niruri* possesses significant hepatoprotective activity against PCM and INH induced liver injury. The observed effect may be related to its antioxidant and anti-inflammatory phytoconstituents. Further studies are required to identify the active constituents and clarify their mechanisms of action.

This is an Open Access journal, and articles are distributed under the terms of the [Creative Commons Attribution 4.0 International License](https://creativecommons.org/licenses/by/4.0/), which permits unrestricted use, distribution, and reproduction in any medium, provided you give appropriate credit to the original author[s] and the source.

Introduction

Orally administered anti-infectives with poor solubility
Nature has always been a valuable symbol of

harmonious coexistence. Natural products derived from animals, plants and minerals have long served as the foundation for the treatment of human diseases [1]. Among these natural resources, plants have played a

particularly important role in healthcare and medicine. Even before recorded history, early humans identified and utilized plants for food, shelter, clothing and fuel, gradually recognizing their medicinal properties. The need to relieve pain, reduce discomfort and improve health encouraged humans to explore nature for therapeutic and health-promoting substances. The use of medicinal plants later developed into one of the earliest forms of science in civilizations such as Greece, China, India and Egypt. In ancient Persia, plants were widely employed as disinfectants, medicines and aromatic agents. Similarly, the ancient Greek medical community possessed considerable knowledge regarding the therapeutic value of plants. Hippocrates, regarded as the father of medicine and his student Aristotle employed medicinal herbs for the treatment of various illnesses. Later, the Greek scientist Theophrastus established the School of Medicinal Plants. In the first century AD, the physician and surgeon Pedanius Dioscorides compiled the influential encyclopedia *De Materia Medica*, which systematically documented nearly 600 medicinal plants and their therapeutic uses. The term medicinal plant refers to different types of plants possessing therapeutic properties. These plants are a valuable source of bioactive compounds that can be utilized in the development and synthesis of drugs. Medicinal plants are not only used for the treatment of diseases in humans and plants but also serve as an important source of substances that help maintain overall health and proper body function. Medicinal plants contain a wide variety of phytoconstituents, commonly known as phytomolecules, phytochemicals, or bioactive nutrients. These organic compounds are naturally produced by plants as protective mechanisms against environmental stress, herbivorous animals and microbial pathogen. Numerous natural bioactive compounds have been isolated from different medicinal plant species due to their traditional therapeutic uses and health-promoting properties. Consequently, more than 25 percent of modern clinical drugs are derived from medicinal plants. The use of herbal products for the prevention and treatment of diseases is an ancient therapeutic practice that has played a major role in the evolution of modern medicine. Many contemporary pharmacological drug classes have originated from phytochemical constituents present in medicinal plants [3].

*Corresponding author: Mr. Raj Kumar Mandal, Chitkara University, Chandigarh-Patiala National Highway, Rajpura, Punjab 140401, India.

<https://doi.org/10.31531/jprst.1000213>

Several important drugs, including ephedrine, Taxol, morphine, reserpine, quinine, aspirin, atropine, digoxin, codeine and tubocurarine, were discovered through the study of medicinal plants and the traditional knowledge of indigenous communities. Medicinal plants such as black garlic, milk thistle, ginger, olive oil, ginseng, ginkgo, seeds, pomegranate and St. John's wort have gained considerable attention and are increasingly being investigated by modern researchers and healthcare professionals. The continued interest in plant-based medicines is expected to grow further, partly due to the high cost associated with the development of synthetic patentable drugs. In recent years, medicinal plants have regained significant attention within the scientific community and pharmaceutical industry. This renewed interest is largely driven by the growing demand for natural and alternative healthcare products, along with the potential discovery of novel therapeutic compounds. Advanced research techniques, including high-throughput screening and modern analytical methods, have facilitated the identification and characterization of numerous bioactive constituents from medicinal plants, leading to the development of new drugs and nutraceutical products [4].

Evolution of Plant-Based Medicines

The evolution of plant-based medicine is one of the earliest and most important developments in human healthcare. Since ancient times, medicinal plants have been widely used for the prevention and treatment of diseases, forming the basis of both traditional and modern medicine. Early civilizations such as India, Egypt, Greece, China and Mesopotamia utilized herbs and plant extracts to maintain health and treat illnesses. Traditional medical systems including Ayurveda, Unani, Traditional Chinese Medicine and Siddha extensively relied on medicinal plants for their therapeutic benefits. The continued use of medicinal plants over centuries highlights their therapeutic value and enduring significance in global healthcare. The transformation of traditional herbal medicine into modern plant-based therapy was driven by advances in pharmacology, photochemistry and biotechnology. Scientists identified active phytochemicals responsible for the medicinal effects of plants, leading to the discovery of important drugs such as quinine, morphine, digoxin, artemisinin and Taxol. These discoveries highlighted the significant pharmaceutical value of medicinal plants and many modern drugs are now derived from plant-based compounds [5].

Conventional Medicine System

Medicinal herbs have been widely used worldwide for maintaining health and treating diseases. In recent years, research in traditional medicine has led to the development of safe, eco-friendly and affordable herbal medicines. Due to its rich herbal resources, the Indian subcontinent plays an important role in meeting the global demand for herbal products and preserving traditional medicinal knowledge. According to the World Health Organization, traditional medicine includes the knowledge, skills and practices developed by different cultures to maintain health and treat illnesses. It may be based on written records or passed down orally through generations. Many people still rely on traditional medicine for primary healthcare and when used outside its native culture, it is often called “complementary and alternative medicine”. Major traditional medicine systems include those of India, China and Africa [6].

Herbal Remedies in Liver Disease

Since ancient times, herbs have been utilized for the treatment of various diseases and disorders due to their beneficial therapeutic effects on the health of human. Owing to their healing and mystical significance, they were often used in rituals across many cultures. Ancient people Possessed considerable knowledge about the therapeutic as well as toxic effects of plants and utilized numerous medicinal herbs for healthcare purposes. Around 65 percent of patients in the Europe and US use herbal medicines for liver disorders due to their easy availability, biochemical diversity, pharmacological properties and lower side effects compared to synthetic drugs. The 21st century has witnessed a major shift toward the therapeutic evaluation of herbal products in liver disease models, integrating traditional medical knowledge with modern evidence-based approaches, standardization and randomized placebo-controlled clinical trials to establish their clinical effectiveness [7]. The management of liver disorders through modern medical approaches is increasingly limited by reduced effectiveness, adverse effects associated with prolonged use and high treatment costs, particularly in developing nations. As a result, there has been growing global interest in medicinal plants that are affordable, readily accessible and require less complex pharmaceutical processing as alternative therapeutic options. Therefore, greater attention has been directed toward traditional herbal remedies due to their high therapeutic potential, low toxicity and costeffectiveness. Several plants including, *Solanum nigrum*, *Aegle marmelos*, *Lepidium sativum*, *Flacourtia indica*, *Coccinia grandis*, *Annona squamosa*, *Cassia roxburghii*, *Silybum marianum* and *Wedelia calendulaceae*, have traditionally been used for the treatment of liver diseases [3].

Liver

The liver, the largest solid organ and gland in the human body. It is a wedge-shaped structure that performs several essential functions. It plays a major role in metabolism, nutrient processing, waste elimination, bile production, bilirubin synthesis, digestion and the metabolism of drugs. The liver starts developing in the human embryo during the 3rd-4th week of gestation and functions as the main organ for blood cell formation throughout prenatal life. During fetal development, the comparatively large size of the liver contributes to the early displacement of the developing intestines to an extra-coelomic position. In adults, the liver typically weighs around 1.4-1.8 kg. The liver is situated in the upper right region of the abdominal cavity, mainly within the right hypochondriac and epigastric areas. It lies beneath the diaphragm, above the stomach, right kidney and intestines and extends partially into the left hypochondriac region. The liver is largely shielded by the lower ribs on the right side and is held in place by folds of the peritoneum known as ligamentous attachments [8]. The liver is attached to the peritoneum at four main sites: the right and left triangular ligaments, the coronary ligament and the falciform ligament. Each cell type carries out specific roles that collectively support the normal structure and function of the liver. The liver is the largest internal organ in the human body and is composed of two major lobes, the right and left lobes, along with two smaller centrally located lobes known as the caudate and quadrate lobes. For clinical applications, the liver is more effectively divided into eight segments based on the distribution of blood vessels and the branching of the bile ducts. The organ receives blood through two major vessels, the portal vein and the hepatic artery, which together deliver nutrient-rich and oxygenated blood to the hepatic sinusoids before it is drained by the hepatic veins. The liver contains several specialized cell types namely cholangiocytes, stellate cells, Kupffer cells [4,5]. Kupffer cells are specialized macrophages located in the liver and account for nearly 8-12% of the total liver cell population. These cells are present within the hepatic sinusoids and serve as an important initial defense mechanism against antigens entering through the gastrointestinal tract. Cholangiocytes are considered the second most abundant cell type in the liver. They form the biliary network and line the inner surface of the bile ducts, where they help collect bile secreted by hepatocytes and transport it to the gallbladder. Various substances may lead to liver inflammation, cirrhosis, tissue degeneration and fibrosis and necrotic damage, ultimately impairing normal liver function. Additionally, the liver closely interacts with several

organs and has an important role in blood flow regulation [9].

Hepatotoxicity

The liver is one of the most important organs in the human body. It plays a crucial role in various metabolic activities essential for energy production, storage and immune function. Toxins and various foreign substances absorbed from the gastrointestinal tract are transported to the liver, where they are detoxified and eliminated through bile and urine. In addition, the liver serves as the primary site for drug metabolism, where medications are converted into active or inactive metabolites by enzymes containing cytochrome P-450. Liver disease affects millions of individuals globally and is responsible for nearly 2 million deaths each year. Its incidence has been rising significantly over time regardless of age, gender, geographical region, or ethnicity. Hepatotoxicity refers to liver injury caused by exposure to drugs, chemicals, or other harmful substances that adversely affect normal hepatic function. It is regarded as an important adverse reaction that can range from minor liver impairment to severe liver failure. Depending on the pattern of biochemical changes, liver damage may be categorized as hepatocellular, cholestatic, or mixed type [10]. Elevated levels of liver enzymes, particularly alanine aminotransferase and alkaline phosphatase, are common indicators of hepatic injury. Several factors such as age, gender, alcohol intake, genetic makeup, environmental conditions and existing liver disorders contribute to the development of hepatotoxicity. The liver is considered a major regulatory organ involved in the progression of hepatic injury and is highly susceptible to damage caused by ROS. Over the years,

various liver-derived in vitro models have been developed to evaluate the potential toxic effects of synthetic chemicals and pharmaceutical agents. Commonly employed experimental models include isolated microsomes, primary hepatocytes, perfused liver systems and immortalized cell lines. Among these, primary isolated hepatocytes and hepatic cell lines are presently the most widely utilized in vitro models for the assessment of hepatotoxicity. Many major classes of drugs exhibit hepatotoxic effects during preclinical investigations and in some cases, hepatic injury is later observed in humans during Phase III or Phase IV clinical trials despite not being predicted from earlier experimental studies. This form of liver injury is commonly linked to hepatocellular death through mechanisms such as necrosis and apoptosis [11]. Most liver disorders, including acute liver injury, drug-induced liver damage, metabolic liver disorders, viral hepatitis and advanced-stage liver diseases, are closely associated with hormonal regulation. Therefore, understanding the clinical features and underlying mechanisms of sexual dimorphism is essential for improving the diagnosis, prevention and treatment of these conditions. Lifestyle factors such as alcohol consumption, obesity and smoking can significantly influence hormone-related mechanisms involved in liver diseases. Hormones exert their regulatory effects on the liver mainly through classical and non-classical receptors present in various hepatic cell types. Additionally, exogenous sex and sex-related hormone therapies may offer promising therapeutic approaches for metabolic liver diseases, cirrhosis and liver cancer. Nevertheless, potential hormone-induced hepatotoxicity must be thoroughly evaluated in preclinical studies and carefully monitored during clinical use [12].

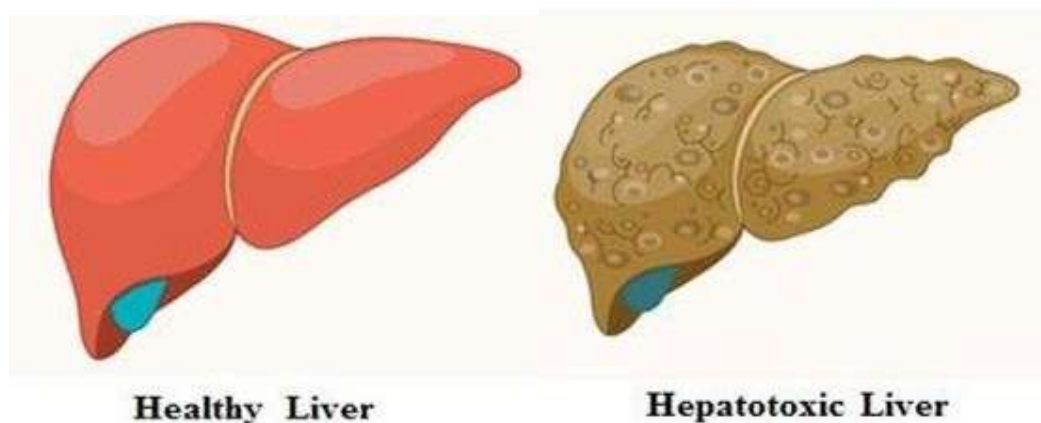


Figure 1: Healthy and Hepatotoxic liver.

Etiology

Various factors can disrupt normal liver function and lead to the development of liver diseases. Some of the major contributing factors are described below.

Alcohol: Alcohol consumption induces oxidative stress, which plays a major role in alcohol related hepatotoxicity. Alcohol intake promotes the formation of ROS such as hydroxyl radicals, superoxide anions and hydrogen peroxide. Accumulation of these reactive molecules in hepatocytes depletes glutathione levels and induces lipid peroxidation of cellular membranes along with oxidation of proteins and DNA, ultimately resulting in liver injury [2].

Viral Infections: Viruses frequently infect the liver and may cause temporary and mild hepatitis. Certain viruses specifically target hepatic tissue and are known as hepatotropic viruses. These viruses can produce clinically significant liver inflammation and, in some cases, progress to chronic viral hepatitis due to persistent infection. Hepatitis viruses such as hepatitis A, BC, D, E and G. Despite variations in their mode of transmission and clinical manifestations, each of these viruses has the ability to cause acute inflammation of the liver and may result in hepatitis [11].

Immune Disorder: Autoimmune disorders may involve hepatocytes or bile ducts and are typically characterized by elevated immunoglobulin levels along with the presence of autoantibodies. Examples of such conditions include autoimmune hepatitis, primary biliary cirrhosis and primary sclerosing cholangitis [4].

Biliary Tract Disease: Blockage of bile flow from the liver may lead to inflammation, fibrosis and ultimately cirrhosis. This obstruction can result from gallstones, tumors, or may occur secondary to surgical injury of the common bile duct. In developed countries, primary cancers of the liver and biliary tract are comparatively uncommon, whereas secondary metastatic tumors occur far more frequently than primary hepatic malignancies [2].

Drug Induced: Drug-induced liver injury (DILI), also referred to as drug-induced hepatotoxicity, is one of the leading reasons for the withdrawal of prescription and over-the counter medications from the market. It can be caused by various xenobiotic, including fungal toxins, herbal remedies and dietary supplements, which are increasingly recognized as important contributors. DILI generally represents an adverse hepatic reaction to drugs and often resembles the clinical features and underlying pathology of both acute and chronic liver

diseases. Paracetamol, also known as acetaminophen or APAP, is a commonly used over-the-counter drug with analgesic and antipyretic properties. However, accidental or intentional overdose of paracetamol remains a significant global health concern and is a major cause of hepatotoxicity, acute liver failure and irreversible liver damage that may require liver transplantation. Numerous studies have demonstrated that paracetamol-induced liver toxicity primarily occurs due to depletion of glutathione stores followed by the accumulation of toxic metabolites within hepatocytes. Despite a few well-established cases, the exact mechanisms through which various drugs and chemical compounds induce hepatic injury remain largely unclear. Therefore, this Special Issue aims to compile recent advances regarding the molecular pathways involved in hepatotoxicity, identify promising novel therapeutic targets and explore the development of innovative tools and models for enhanced drug screening and evaluation of pharmacological responses [8].

Pathophysiology

Hepatotoxicity may occur through two major mechanisms: intrinsic toxicity, which is dose dependent and predictable and idiosyncratic toxicity, which is unpredictable and occurs independently of dose. DILI can be classified according to the pattern of liver damage, clinical manifestations such as hepatocellular, cholestatic, or mixed injury, or the histological changes observed in liver tissue. Jaundice is considered the principal clinical sign of hepatotoxicity, although many affected individuals remain asymptomatic. In laboratory investigations, cholestatic liver injury is generally associated with elevated ALP levels, whereas hepatocellular injury is characterized by increased serum aminotransferase concentrations [10].

Oxidative stress: Oxidative stress and nitrosative stress generally occur due to an imbalance between the excessive generation of ROS and RNS and their inadequate removal, along with an impaired antioxidant defense response. Oxidative stress may arise from either exogenous or endogenous sources. Exogenous oxidative stress is commonly triggered by drugs and toxic agents, whereas endogenous oxidative stress is naturally associated with certain pathological conditions and diseases. Excessive or uncontrolled oxidative stress can be detrimental to the body and may lead to tissue and organ damage [7].

Mitochondrial Dysfunction: Mitochondria are semi-autonomous intracellular organelles that play a vital role in normal cellular functions. They serve not only as the

primary source of cellular energy production but also as important sites for numerous metabolic activities within the cell. Mitochondria can be damaged by several factors, including drugs, viruses, fat accumulation and carcinogens. Excessive injury impairs mitochondrial structure and function, resulting in mitochondrial swelling, loss of membrane integrity, reduced ATP production, increased ROS generation, mitochondrial DNA damage and defective mitophagy [8].

Lipid Peroxidation: Tissue injury caused by iron, oxygen and various hepatotoxins often involves the generation of reactive free radicals and the initiation of lipid peroxidation. Lipid peroxidation has gained considerable attention because many xenobiotics are known to induce oxidative degradation of hepatic lipids, leading to severe damage to biological membranes. Once initiated, lipid peroxidation can propagate through autocatalytic reactions that depend on oxygen and metal-catalyzed breakdown of lipid hydroperoxides. In the case of toxic aryl halides and allyl alcohol,

glutathione depletion alone may not be sufficient to explain the extensive lipid peroxidation observed [2].

Immune-Mediated Hepatic Injury: Some drugs and their metabolites combine with cellular proteins to form neoantigens that stimulate immune responses. This leads to activation of T lymphocytes, Kupffer cells and inflammatory cytokines, resulting in immune-mediated damage to hepatocytes. Such mechanisms play a significant role in idiosyncratic drug-induced liver injury [6].

Genetic factors: Genetic factors also play an important role in the development of hepatic injury. Polymorphisms in antioxidant defense genes, such as manganese superoxide dismutase (MnSOD), can impair the ability of hepatocytes to eliminate reactive oxygen species effectively. As a result, oxidative stress increases, making individuals more susceptible to hepatocellular damage [9].

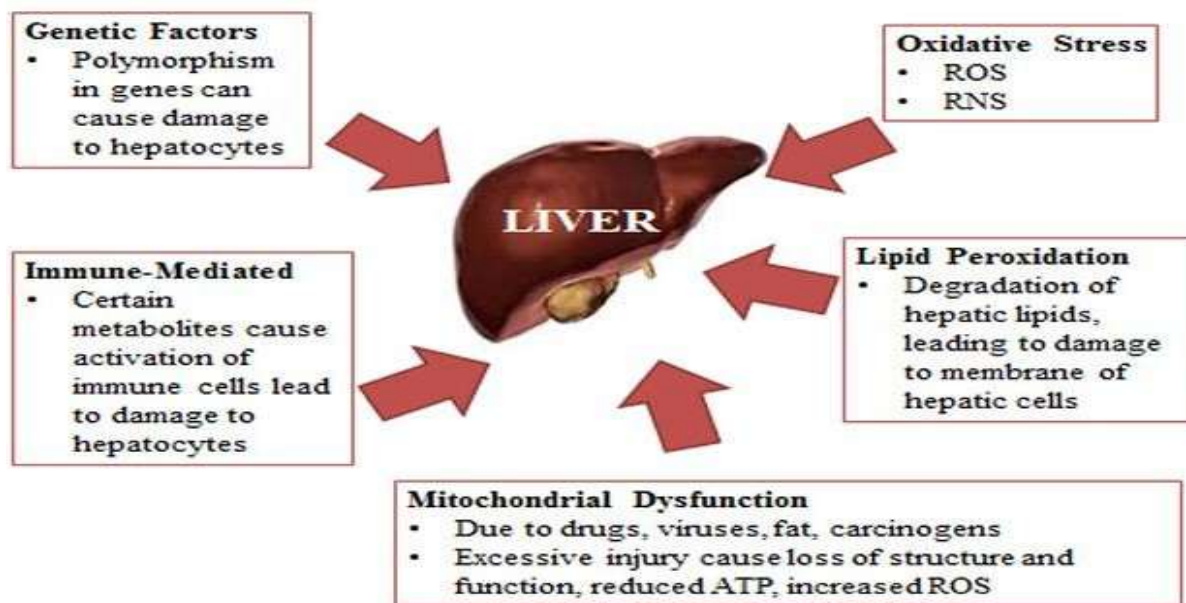


Figure 2: Diagrammatic representation of pathophysiology.

Symptoms

Symptoms related to hepatotoxicity may include

- ✓ Nausea
- ✓ Vomiting
- ✓ Tiredness or weakness
- ✓ Pruritus

- ✓ Intense abdominal pain
- ✓ Swelling of legs/feet
- ✓ Dark colored urine
- ✓ Light colored feces
- ✓ Sudden or unusual weight gain for a short interval of time
- ✓ Generalized itching
- ✓ Rashes on skin
- ✓ Continuous bleeding

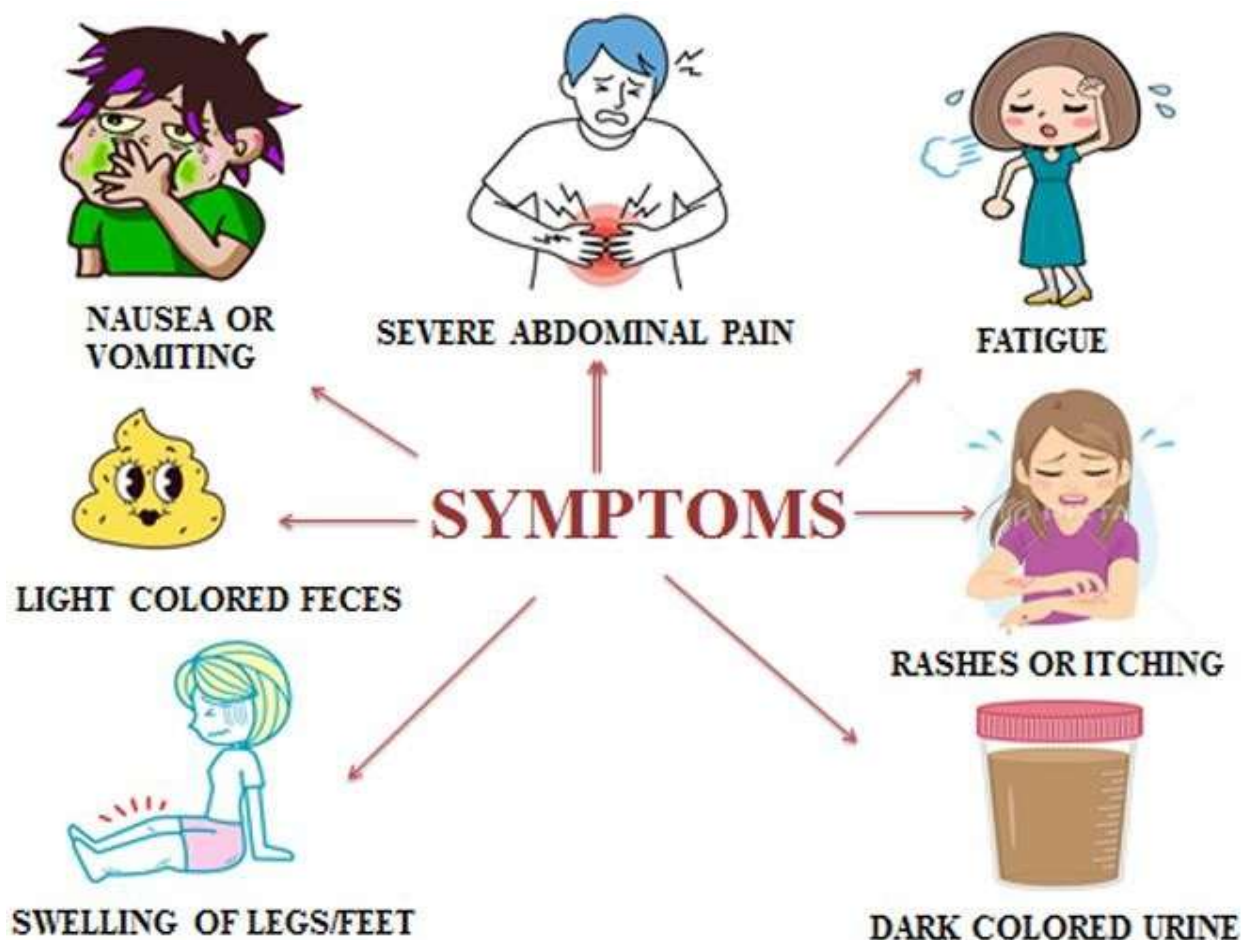


Figure 3: Diagrammatic Representation of Symptoms of Liver Damage.

Biochemical Parameters

A biomarker is described as a measurable characteristic that is assessed objectively and used to indicate normal biological functions, disease-related processes, or responses to therapeutic treatment Singh et al. [86]. Markers of hepatic cellular injury are generally enzymes released into the circulation when liver cells are damaged or experience necrosis. Frequently used indicators include ALP, AST, GGT, ALT and GLDH [11].

ALT and AST: are metabolic enzymes and increased serum levels of these enzymes reflect hepatocellular necrosis, injury and inflammatory changes in the liver. In cases of viral hepatitis, the elevation of AST is generally lower compared to ALT; however, both enzymes remain important clinical markers for identifying acute liver damage [13].

GGT: is commonly used as a conventional marker for hepatic disorders, biliary tract abnormalities and alcohol intake. Moreover, elevated GGT levels have also been associated with an increased risk of stroke, type II diabetes mellitus and coronary artery disease [7].

Bilirubin: is first carried to the liver in an insoluble form, where it undergoes conversion into a water-soluble state to facilitate its excretion. Increased total bilirubin (TBIL) levels suggest impairment in normal hepatic metabolic function [11].

Alkaline phosphatases: are classified into two main types: tissue-specific and tissue-nonspecific isoenzymes. Tissue-specific alkaline phosphatases are mainly found in the placenta, colon and germinal tissues, whereas tissue-nonspecific alkaline phosphatases, which play an important therapeutic role, are predominantly present in the liver, bone and kidney tissues. ALP is widely utilized as a diagnostic marker for cholestatic liver disorders. Approximately 75% of patients with either

intrahepatic or extrahepatic cholestasis exhibit ALP levels that are at least four times higher than the upper normal limit [14].

GLDH: may also serve as a valuable biomarker for differentiating liver injury from musclerelated causes of elevated ALT levels in individuals with muscular disorders. However, it is still uncertain whether the combined assessment of ALT and GLDH offers greater diagnostic accuracy compared to the commonly used combination of ALT and creatine kinase in clinical practice. Overall, serum GLDH is considered a promising and reliable indicator of mitochondrial damage and remains useful in translational studies involving acute hepatic injury [3].

Albumin: The liver is able to produce sufficient protein to keep albumin levels normal until about 50% of the liver tissue is damaged. Measuring plasma albumin helps evaluate the severity and duration of liver disease. However, in acute kidney disease, plasma albumin levels decrease, which reduces the usefulness of this measurement for assessment. They may serve as early markers of liver dysfunction, potentially enhancing the diagnosis and treatment of liver diseases. Antiviral therapy can help restore liver function and reduce cirrhosis-related decompensation, which may influence albumin binding capacity [15].

Diagnosis

Diagnosis is a process by which we identify the disease or medical condition by evaluating the person's signs, symptoms, medical history, physical examination and laboratory, or imaging findings. It helps determine the exact nature of the illness affecting the individual, enabling healthcare professionals to provide appropriate treatment and management [11].

✓ History and physical: The clinical manifestations of drug-induced hepatotoxicity are diverse and may often remain asymptomatic. Patient history commonly reveals the onset of signs and symptoms within 3–6 months after exposure to the causative drug; therefore, obtaining a detailed medication history is essential for accurate diagnosis [2].

✓ Laboratory examinations: This includes LFT which examines the serum enzyme levels, namely ALP, AST, Bilirubin, Albumin, GGT and many others.

✓ Radiological findings: Imaging or radiological findings include CT/PET, MRI and ultrasound to see the

liver structure and find out any kind abnormality to liver.

✓ Biopsy: It is the method in which a sample of tissue taken from the liver and examine to detect disease [16].

Management of Hepatotoxicity

The management of hepatotoxicity primarily varies according to the severity of liver damage, the causative agent involved and the patient's clinical manifestations. Timely diagnosis and appropriate therapeutic intervention are crucial to prevent the development of liver injury.

Medication Treatment

- N-acetylcysteine (NAC): It is a standard antidote against acetaminophen-induced hepatotoxicity. It restores glutathione levels, decreases oxidative stress and enhances mitochondrial activity. Recent research has also demonstrated its potential therapeutic benefits in cases of acute liver failure not caused by acetaminophen toxicity [14].

- Steroid Therapy: Glucocorticoids, including prednisolone, are commonly administered in cases of immune-mediated or hypersensitivity-related hepatotoxicity. These corticosteroids reduce inflammatory and immune reactions, especially in liver injuries that mimic autoimmune hepatitis. Nevertheless, their therapeutic use is still debated and should be tailored to individual patients.

- Ursodeoxycholic acid: is widely employed in the treatment of cholestatic hepatotoxicity. It enhances bile secretion, stabilizes liver cell membranes and alleviates cholestatic manifestations including itching and jaundice [17].

- Oxidative Stress-Targeted Therapy: is a key contributor to hepatocellular damage; consequently, antioxidants including silymarin, vitamin E and glutathione precursors have been explored for their hepatoprotective properties. Herbal antioxidants may also assist in minimizing lipid peroxidation and inflammation-induced liver injury. Silymarin exhibits antioxidant, anti-inflammatory and antifibrotic activities that contribute to its hepatoprotective effects. It helps stabilize hepatocyte membranes and enhances protein synthesis, thereby supporting regeneration of liver cells. Several studies have reported improvements in liver function parameters among patients with liver disorders; however, the findings remain inconsistent and further well-designed clinical trials are required to

establish its efficacy conclusively. The commonly recommended dosage of silymarin is 140 mg administered two to three times daily [14].

Withdrawal of Causative Agent

The primary and most crucial step in treating hepatotoxicity is the discontinuation of offending drug. In mild to moderate cases, liver function improves once the offending drug is stopped. Early cessation of the causative drug markedly decreases the risk of complications and death Tujios [18].

Supportive Care in Hepatotoxicity

Supportive management involves maintaining proper hydration, providing nutritional supplementation, regular assessment of liver enzyme levels and correcting electrolyte disturbances. Individuals with severe hepatotoxicity may need hospitalization and close observation for complications including coagulopathy, hepatic encephalopathy and acute liver failure [16].

Liver Transplant

Liver transplantation is considered the ultimate therapeutic option for fulminant hepatic failure when conservative treatment is unsuccessful. It is recommended for patients exhibiting progressive hepatic encephalopathy, marked coagulopathy and irreversible liver injury [19].

Innovative Strategies for Hepatotoxicity Management

Current advances in hepatotoxicity management are exploring novel approaches such as stem cell-based therapies, regulation of cytokine activity, mitochondrial protective compounds, gene-targeted modulation strategies and artificial liver support systems for severe liver injury. The treatment of hepatotoxicity involves a multidisciplinary strategy that includes discontinuation of the offending agent, supportive management, targeted therapeutic interventions and liver transplantation in severe or advanced cases. In addition, preventive measures play an essential role in reducing the risk of hepatotoxicity development [20].

Materials and Method

Procurement and Authentication

Fresh leaves of *P. niruri* were collected from them in the month of October.

Collection, Drying and Extraction

- The leaves of plant were washed with running water and then shade-dried for two weeks.
- The dried leaves were ground into a coarse powder, passed through a 20-mesh sieve and finally stored in a sealed, clear container to prevent contamination.
- Ethanol was used as the solvent for extraction. A total of 250 g of the powdered material was extracted using a Soxhlet apparatus for 14 hours at 40°C. The resulting extract was filtered through Whatman No. 42 filter paper and then concentrated using a rotary evaporator at 50°C.
- Afterward, the percentage yield was determined.

% Yield: $\frac{\text{Weight of extract}}{\text{Weight of empty china dish}} \times 100 / \frac{\text{Weight of Powder Drug}}$

Materials

The materials used in the present study included isoniazid, paracetamol, silymarin, ethanol and distilled water.

Experimental Animals

A total of 60 rats, including both males and females aged 8-12 weeks and weighing 180-200 g, were used to evaluate hepatoprotective activity. The animals were obtained from the Central Animal House of Pharmacy Institute, Greater Noida. The study was conducted with approval from the IAEC under protocol number IAEC/05/09/2026. The rats were acclimatized under standard laboratory conditions, including a temperature of 25 ± 2 °C, relative humidity of 45-55%, free access to food and water and a 12-hour light-dark cycle. All experimental procedures were carried out in accordance with CPCSEA ethical guidelines.

Preliminary Screening

Preliminary phytochemical screening of the ethanolic leaf extract of *P. niruri* (PNEE) was carried out to verify the presence of bioactive metabolites. These constituents are believed to be responsible for its beneficial effects. The extract was analyzed using

standard conventional methods to detect various secondary metabolites, including alkaloids, glycosides, terpenoids, steroids and saponins, in accordance with established protocols Shaikh and Matsyagandha [16].

Alkaloid's Tests

The PNEE was first diluted with a small amount of water to prepare the test solution, followed by further analysis [21].

Mayer's test: Potassium mercuric iodide was added to the PNEE and shaken gently. The appearance of a cream-colored precipitate indicated the presence of alkaloids [22].

Hager's test: The test solution was acidified with 2 ml of HCl, after which picric acid was added. The development of a yellow color confirmed the presence of alkaloids [22].

Dragendorff's test: The solution was acidified with 2 ml of HCl and then treated with potassium bismuth iodide and Dragendorff's reagent. The formation of a reddish-brown precipitate indicated the presence of alkaloids [22].

Tests for Flavonoids

Zinc-HCL reduction test: The addition of small amounts of hydrochloric acid and zinc dust to the test solution produced a magenta-red coloration, confirming the presence of flavonoids [23].

Shinoda's test: Magnesium ribbon was added to the PNEE test solution, followed by the addition of concentrated hydrochloric acid. The development of a pink to magenta-red color indicated the presence of flavonoids [23].

Test for polyphenols

a. When three drops of 5% lead acetate solution were added to 1 ml of PNEE, the formation of a yellow precipitate indicated the presence of phenolic compounds [24].

Saponin Test

a. Foam test: In a separate test tube, 2 mL of PNEE was combined with 20 mL of water. The solution was shaken vigorously for 15 minutes and the formation of stable foam confirmed the presence of Saponin [23].

Test for Glycosides

Libermann's test: About 1 mL of PNEE was mixed with acetic acid and chloroform and the solution was cooled before adding concentrated H₂SO₄. The appearance of a green color indicated the presence of aglycone, representing the steroidal portion of glycosides [23].

Test for Fatty Acids

A mixture of 0.5 mL of PNEE and 4 mL of ether was prepared and applied to filter paper. After allowing it to evaporate and dry, the development of transparency on the filter paper indicated the presence of fatty acids [24].

Test for Amino Acids

One milliliter of PNEE was treated with a few drops of ninhydrin reagent. The appearance of a purple color confirmed the presence of amino acids [25].

Tests for Terpenoids:

Libermann Burchard Test: Two milliliters of PNEE were treated with a few drops of acetic anhydride and mixed well. Subsequently, 1 mL of concentrated H₂SO₄ was carefully added. The formation of a red coloration in the lower layer of the test tube confirmed the presence of terpenoids [25].

Salkowski's test: About 2 mL of the extract (PNEE) was taken as the test solution and mixed thoroughly with a few drops of concentrated H₂SO₄. The appearance of a golden yellow color after a short time indicated the presence of terpenoids [25].

Tests for Proteins

One milliliter of PNEE was mixed thoroughly with 1 mL of 40% NaOH and a few drops of copper sulfate were added along the inner wall of the test tube. The appearance of a violet or pink color indicated the presence of proteins [25].

Tannins Test

Ferric chloride test: Small amounts of FeCl₃ solution were added to the PNEE sample and observed for a short time. The development of a dark blue color confirmed the presence of tannins [23].



Figure 4: Tests for Flavonoids



Figure 5: Tests for Glycosides



Figure 6: Tests for Carbohydrates



Figure 7: Tests for Proteins

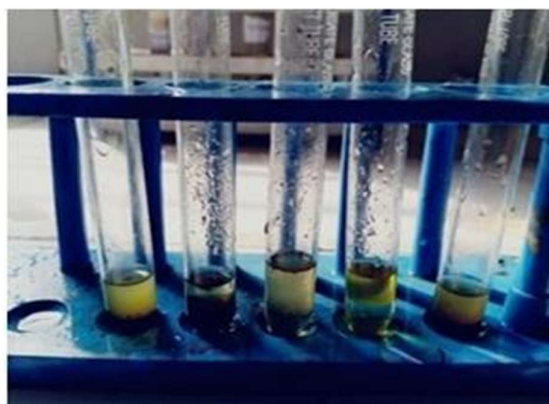


Figure 8: Tests for Alkaloids

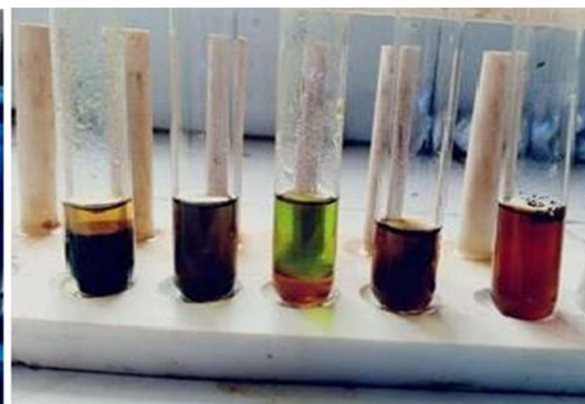


Figure 9: Tests for Terpenoids

In vivo Hepatoprotective Activity

Paracetamol-Induced Hepatotoxicity Model

Paracetamol is a commonly used analgesic and antipyretic, but at high doses it can lead to acute liver injury. Its administration in excessive amounts results in damage to centrilobular hepatic cells, marked by features such as nuclear pyknosis and an eosinophilic cytoplasm, which eventually progresses to extensive hepatic lesions. This toxicity is mainly attributed to the formation of N-acetyl-p-benzoquinone imine, a reactive metabolite that binds covalently to sulfhydryl groups of proteins. This interaction depletes GSH and promotes lipid peroxidation, ultimately leading to liver cell necrosis [26].

Experimental Procedure

Group I: Negative control, normal saline, (1ml/kg, i.p.) daily

Group II: Positive control, normal saline (1ml/kg, i.p.) daily + Paracetamol (750mg/kg, p.o.) on days 8 and 16

Group III: Standard, silymarin (50mg/kg, p.o.) daily + Paracetamol (750mg/kg, p.o.) on days 8 and 16

Group IV: Paracetamol (750mg/kg, p.o.) on days 8 and 16 + low dose of *P. niruri* (200 mg/kg, p.o.)

Group V: Paracetamol (750mg/kg, p.o.) on days 8 and 16 + high dose of *P. niruri* (400 mg/kg, p.o.)

The rats were randomly allocated into five groups, with each group consisting of six animals (n = 6). Group I received normal saline daily for 3 weeks. Group II was given normal saline daily along with paracetamol (750 mg/kg) on days 8 and 16 to induce liver damage. Group III received silymarin (50 mg/kg) daily along with paracetamol. The treatment groups were administered paracetamol (750 mg/kg) on days 8 and 16, along with *P. niruri* at doses of 200 mg/kg (low dose) and 400 mg/kg (high dose), respectively [27].

Evaluation of Biochemical Parameters

On the 22nd day, 24 hours after treatment, the rats were anesthetized with ketamine hydrochloride (100 mg/kg) and xylazine (15mg/kg) prior to sacrifice. Blood samples were obtained via cardiac puncture for the estimation of biochemical parameters. Blood collected in EDTA tubes was centrifuged at 4000 rpm for 20 minutes to separate the serum and the supernatant was carefully collected and stored for further evaluation [28].

Histopathology

After euthanizing the rats, the liver was excised and fixed in 10% formalin. A 1 g portion of liver tissue was accurately weighed, mixed with 100 mL of normal saline and homogenized at around 4°C using a cold-water bath. The resulting 1% liver homogenate was utilized for biochemical analyses [29].

Isoniazid-Induced Hepatotoxicity Model

Drug-induced liver injury is a significant adverse effect associated with anti-tubercular therapy, particularly regimens that include isoniazid (INH), rifampicin and pyrazinamide. While each of these drugs has hepatotoxic potential individually, their combined use can intensify liver damage. In this model, single INH used for liver toxicity at a dose of 50 mg/kg. INH is metabolized into monodactyl hydrazine, which is further converted by cytochrome P450 enzymes into toxic metabolites that contribute to hepatic injury [30].

Experimental Procedure

Group I: Negative control, normal saline (1ml/kg, p.o.)

Group II: Positive control, isoniazid (50mg/kg, p.o.)

Group III: Standard, isoniazid (50mg/kg, p.o.) + silymarin (50mg/kg, p.o.)

Group IV: Isoniazid + low dose of *P. niruri* (200 mg/kg, p.o.)

Group V: Isoniazid + high dose of *P. niruri* (400 mg/kg, p.o.)

Rats were randomly divided into five groups, with six rats in each group (n = 6). Group I (negative control) received a standard diet along with normal saline (1 mL/kg, p.o.) daily for 43 days. All groups, except Group I, were administered isoniazid orally at a dose of 50 mg/kg for 28 days. From day 29 onward, isoniazid administration was discontinued and the animals received oral treatment with normal saline (1 mL/kg), silymarin (50 mg/kg) and EEPN at doses of 200 mg/kg and 400 mg/kg. At the end of the experimental period, the animals were sacrificed and blood samples were collected for serum separation [31].

Evaluation of Biochemical Parameters

On the 44th day of the experiment, after overnight fasting, the rats were anesthetized by slow subcutaneous administration of ketamine/xylazine (50/5 mg/kg). Blood samples were collected from each animal through the retro-orbital venous plexus using microhematocrit capillary tubes into EDTA-containing tubes for plasma separation and hematological evaluation. The samples were centrifuged at 5000 rpm for 10 minutes. The

Citation: Sarswat D, Quamuddin M, Nimesh S, et al. Evaluation of the Hepatoprotective Effect of *Phyllanthus niruri* in Paracetamol and Isoniazid-Induced Hepatotoxicity in Albino Wistar Rat. *J Pharm Res Sci Technol* 2026; 10(2): 213. doi: [10.31531/jprst.1000213](https://doi.org/10.31531/jprst.1000213)

obtained serum was then analyzed for ALT, AST, ALP, albumin/globulin ratio and total bilirubin [32].

Histopathology Examination

All rats were sacrificed under anesthesia by cutting neck using a sharp blade, followed by dissection. The liver was immediately excised, weighed and rinsed with ice-cold saline to remove excess blood. Liver sample was preserved in 10% neutral-buffered formalin for histopathological analysis [33].

Statistical Analysis

The study data were expressed as Mean $\pm$ Standard Error of the Mean (SEM). Statistical comparisons

among the groups were performed using one-way analysis of variance (ANOVA) to determine statistical significance. A p-value less than 0.05 was considered statistically significant [34].

Results and Discussion

Estimation of Extracted Plant Material

The dried leaf material of *P. niruri* was extracted for 24 hours using a Soxhlet apparatus with 70% ethanol as the solvent. The resulting extract was then filtered and concentrated using a rotary evaporator, after which the percentage yield was calculated.



Figure 10: Soxhlet Assembly of *P. niruri* Extraction Process.

The percentage yield was calculated to be 8.9 % (w/w).



Figure 11: Rotary Vacuum Evaporation Process of *P. niruri* Ethanolic Extract.

Preliminary Phytochemical Analysis

Phytochemical Test Results of *P. niruri*

The leaves of *P. niruri* were analyzed through phytochemical screening using laboratory-grade reagents that indicate the presence of phytoconstituents by producing characteristic color changes or precipitates. As presented in the table below, the ethanolic leaf extract (PNEE) was found to contain

alkaloids, flavonoids, tannins, Saponin, amino acids, fatty acids and terpenoids.

These phytochemical identification tests may play an important role in isolating the bioactive constituents of the plant responsible for its pharmacological activities. *P. niruri* has demonstrated therapeutic potential against various diseases and exhibits several pharmacological properties, including antidiabetic, antioxidant, anti-inflammatory, wound healing, Nephroprotective, gastro protective and Reno protective activities.

Table 1: Results of Preliminary Phytochemical Screening.

Phytochemical tests	Tests result	Precipitate or color formed
Alkaloids	+	A reddish-brown color indicates the presence of alkaloids.
Flavonoids	+	Pink colored solution indicates the presence of flavonoids
Saponins	+	A soap formation indicates the presence of saponins
Tannins	+	Black color shows the presence of tannins
Phenols	+	White precipitate shows the presence of phenols
Glycosides	+	Green color indicates the presence of glycosides
Amino acids	+	Purple color shows presence of amino acids
Terpenoids	+	A reddish color presents terpenoids
Fatty acids	+	A transparent spot on filter paper shows fatty acids
Proteins	+	Violet color indicates proteins

Keywords: '+' indicates the presence of phytoconstituents, while '-' indicates the absence of phytoconstituents

In vivo Hepatoprotective Activity

Paracetamol-Induced Hepatotoxicity Model

Oral administration of paracetamol (750 mg/kg body weight) caused a significant elevation in hepatic marker enzymes such as ALT, ALP, AST and bilirubin levels when compared with the normal control group. Treatment with the ethanolic extract of *P. niruri* at doses of 200 mg/kg and 400 mg/kg significantly reduced the

elevated liver enzyme levels in paracetamol-induced hepatotoxicity. In comparison with the healthy control group, paracetamol administration markedly decreased albumin and total protein levels while increasing bilirubin levels. Administration of the plant extracts exhibited a protective effect against liver damage and improved chronic liver conditions by restoring or reducing the altered serum biochemical parameters toward normal levels [19].

Table 2: Effect of *P. niruri* on Paracetamol-Induced Hepatotoxicity in Rats (ALT, AST, ALP).

Groups	ALT (u/l)	AST (u/l)	ALP (u/l)
Normal Control	3.24 ± 0.23	74.6 ± 10.09	149.8 ± 9.60
Experimental Control	4.36 ± 0.36	161.4 ± 38.74	291.0 ± 6.44
Standard Control	2.30 ± 0.45***	48.4 ± 7.33***	149.6 ± 8.76***
Test (Low Dose)	3.38 ± 0.44**	89.56 ± 2.84**	208.8 ± 12.68**
Test (Low Dose)	3.28 ± 0.18***	50.6 ± 8.53***	152.8 ± 9.12***

The data were expressed as mean ± SEM for six animals in each group (n = 6). The highest response was observed with *P. niruri* at a dose of 400 mg/kg (***p < 0.001), followed by the standard treatment (***p < 0.001) and 200 mg/kg (**p < 0.01).

Statistical Analysis

Statistical evaluation was performed using one-way ANOVA. A significant elevation in serum ALT, AST and ALP levels was observed in the experimental control group when compared with the normal control group. Administration of the standard drug (p < 0.001***)

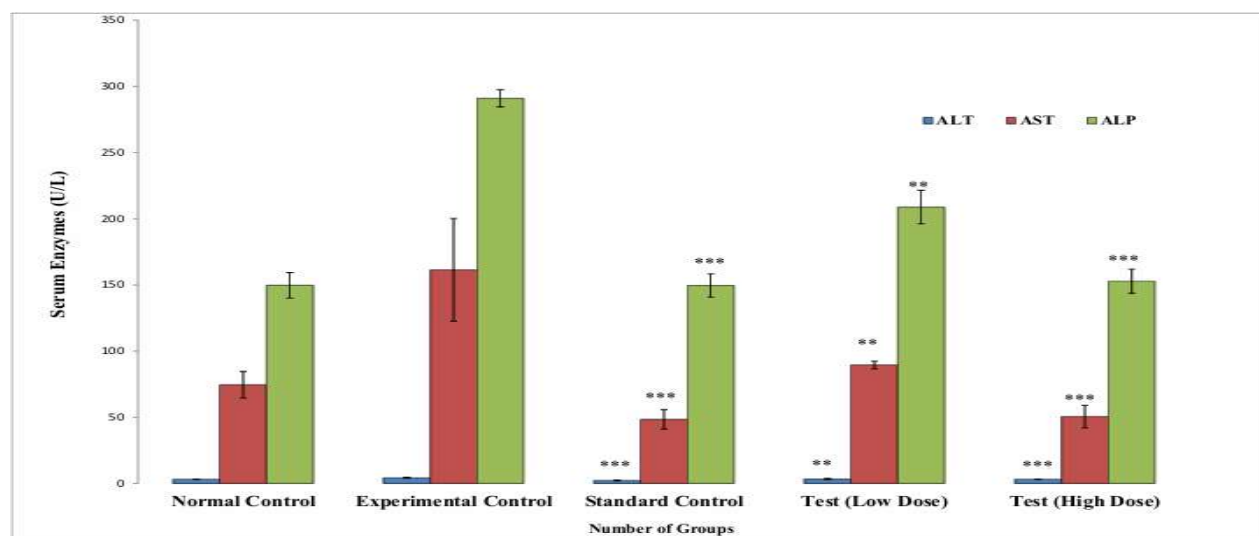


Figure 12: Effect of ethanolic leaf extract of *Phyllanthus niruri* on serum ALT, AST and ALP levels in paracetamol-induced hepatotoxicity in rats.

Histopathology

Liver tissues were subjected to histopathological analysis and the relevant images are displayed in Figure.

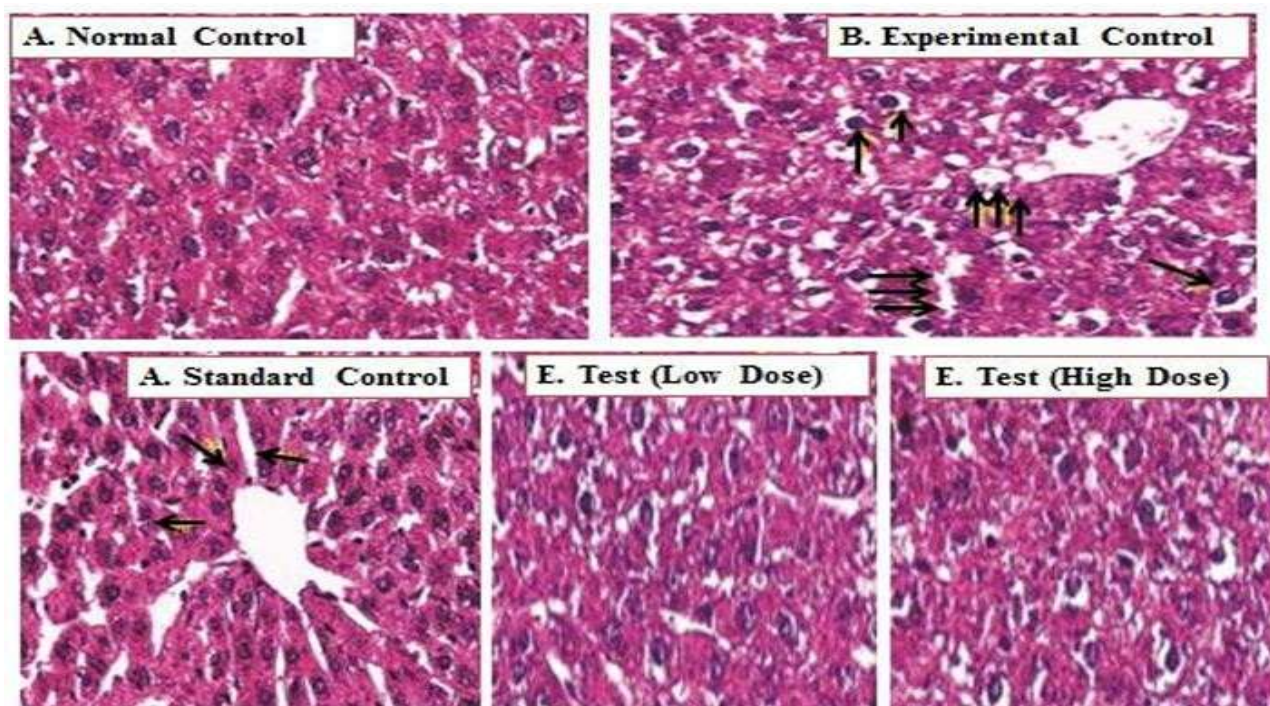


Figure 13: Histopathological examination of liver tissue sections

- Normal Control showed normal hepatic architecture with properly arranged hepatocytes, reflecting healthy liver morphology.
- Experimental Control, showed severe hepatic injury characterized by hepatocellular degeneration, cell infiltration, inflammatory cell infiltration, vacuolization, necrosis and disruption of normal architecture.

hepatic architecture, indicating pronounced hepatotoxicity.

- c) Standard Control represents considerable protection against hepatic damage, with nearly normal hepatocyte arrangement, reduced inflammation and restoration of normal liver structure.
- d) Test (Low Dose), demonstrated moderate histological improvement, including reduced cellular degeneration and partial recovery of normal hepatic architecture compared with the experimental control group.
- e) Test (High Dose), showed significant restoration of liver tissue, with almost normal hepatocellular architecture, minimal inflammatory changes and reduced necrosis, suggesting potent hepatoprotective activity.

Following 21 days of treatment, histopathological evaluation of liver sections from the experimental rats demonstrated restoration of normal hepatic architecture, reduced periportal inflammatory cell infiltration, decreased levels of liver injury markers such as ALT, ALP, AST and bilirubin and an increase in total protein content compared with the paracetamol-treated control group. These beneficial effects were observed after administration of the test drugs at doses of 200 mg/kg and 400 mg/kg [18].

Isoniazid-Induced Hepatotoxicity Model

Oral administration of isoniazid significantly elevated the levels of hepatic marker enzymes such as ALT, ALP, AST and bilirubin when compared with the normal control group. Treatment with the ethanolic extract of *P. niruri* at doses of 200 mg/kg and 400 mg/kg markedly reduced the elevated liver enzyme levels in isoniazid-induced hepatotoxicity. In comparison with the normal control group, oral administration of isoniazid significantly altered the levels of albumin, bilirubin and total protein. Treatment with the plant extracts exhibited a protective effect against liver damage and improved chronic liver conditions by restoring the altered serum biochemical parameters toward normal levels. In the isoniazid-induced hepatotoxicity model, the effect of the test drug *P. niruri* at doses of 200 mg/kg and 400 mg/kg was evaluated using various biochemical and hematological parameters. Treatment with the extract significantly reduced the elevated serum levels of liver enzymes such as ALT, ALP, AST and total bilirubin, while increasing total protein and albumin levels. These findings indicate that rats are susceptible to liver toxicity following administration of isoniazid and that *P. niruri* possesses protective effects against hepatic damage [22].

Table 3: Effect of *P. niruri* on Isoniazid-Induced Hepatotoxicity in Rats (ALP, AST, ALT)

Groups	ALP (u/l)	AST (u/l)	ALT (u/l)
Normal Control	56.40 ± 4.82	44.40 ± 5.01	20.64 ± 5.75
Experimental Control	118.36 ± 7.40	115.40 ± 6.35	116.76 ± 12.13
Standard	85.18 ± 12.96	52.64 ± 10.68	36.60 ± 10.14
Test (Low Dose)	100.88 ± 5.52	94.62 ± 4.26	61.30 ± 5.96
Test (High Dose)	94.20 ± 13.35	68.38 ± 12.78	47.11 ± 10.15

The data were expressed as mean ± SEM for six animals in each group (n = 6). The highest response was observed with *P. niruri* at a dose of 400 mg/kg (**p < 0.001), followed by the standard treatment (**p < 0.001) and 200 mg/kg (**p < 0.01).

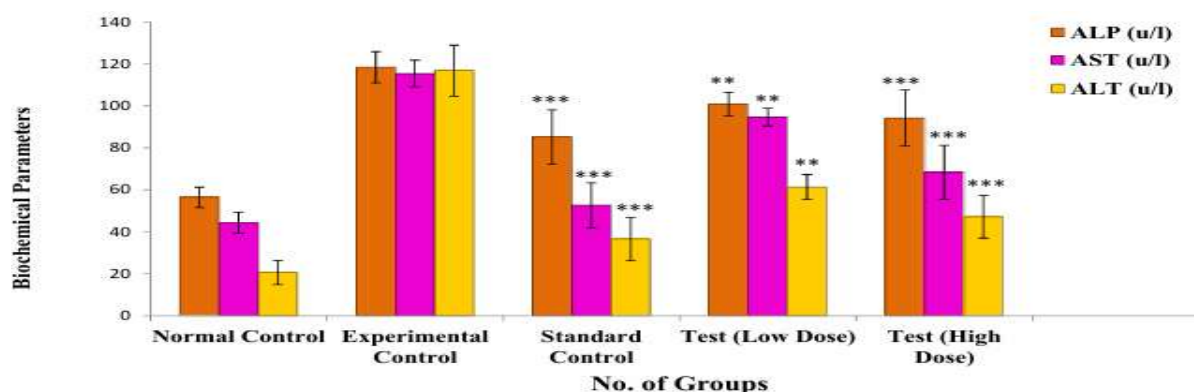


Figure 14: Graphical Representation of Effect of *P. niruri* on Isoniazid-Induced Hepatotoxicity in Rats (ALP, AST, ALT)

Statistical Analysis

Statistical analysis was conducted using one-way ANOVA. The experimental control group showed a significant increase in serum ALT, AST and ALP levels compared with the normal control group. Treatment with the standard drug ($p < 0.001$ *** and the test

extract significantly restored these elevated enzyme levels toward normal values ($p < 0.001$ *** and $p < 0.01$ ** respectively).

Histopathology

Liver tissues were subjected to histopathological analysis and the relevant images are displayed in Figure.

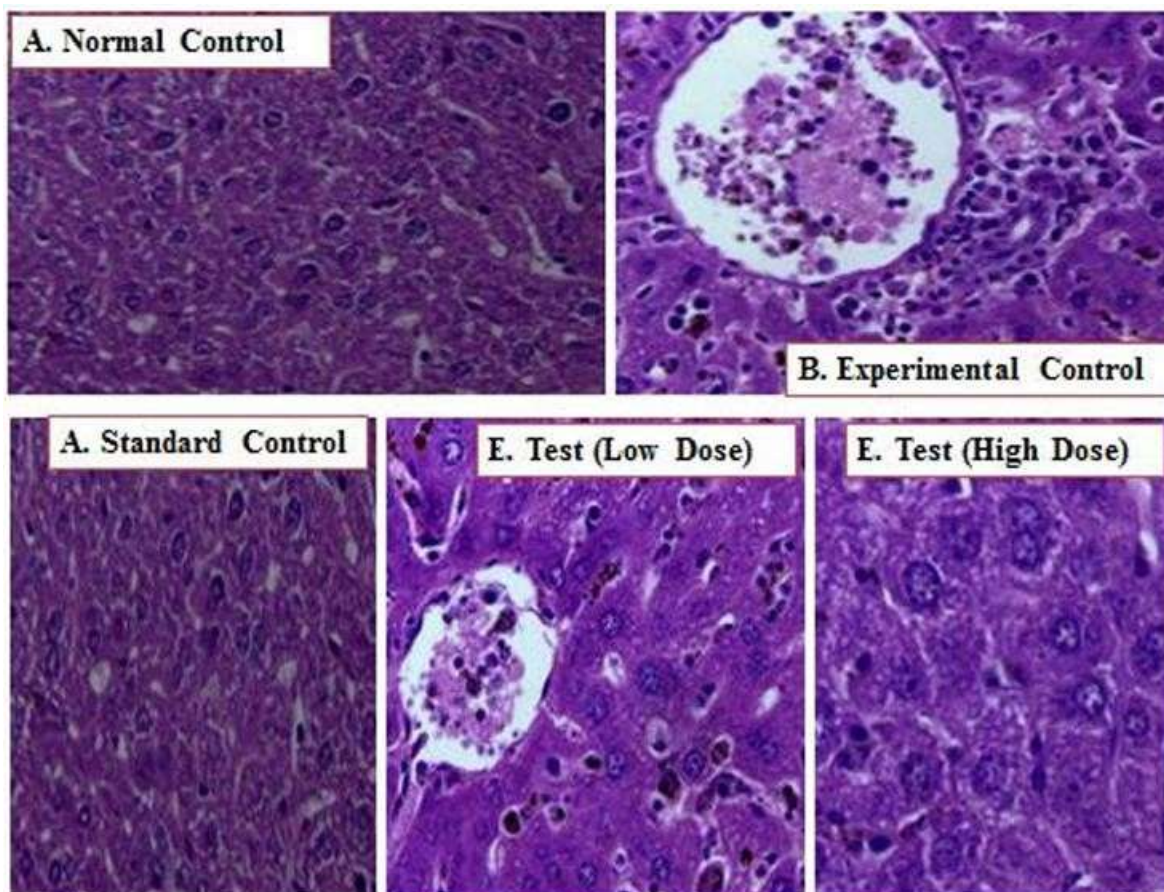


Figure 15: Histopathological evaluation of treated liver tissues

(A) Normal Control, showed normal hepatic architecture with well-organized hepatocytes, indicating healthy liver morphology. (B) Experimental Control, Liver tissue represented severe hepatic damage characterized by hepatocellular degeneration, inflammatory cell infiltration, vacuolization, necrosis and disruption of normal liver architecture, suggesting marked hepatotoxicity. (C) Standard Control, demonstrated significant protection against hepatic injury, with nearly normal hepatocyte arrangement, reduced inflammation and restoration of hepatic structure. (D) Test (Low Dose), showed moderate histopathological improvement, including decreased cellular degeneration and partial restoration of normal

hepatic architecture compared with the experimental control group. (E) Test (High Dose), showed substantial recovery of liver tissue with almost normal hepatocellular architecture, minimal inflammatory changes and reduced necrosis, indicating strong hepatoprotective activity. After 43 days of treatment, histopathological examination of liver sections from the experimental rats revealed restoration of normal hepatic architecture, decreased periportal inflammatory cell infiltration, reduced levels of liver injury markers including ALT, ALP, AST and bilirubin, along with an increase in total protein levels compared with the paracetamol-treated control group. These protective

effects were observed following administration of the test drugs at doses of 200 mg/kg and 400 mg/kg.

Discussion

Earlier investigations on *P. niruri* (*P. niruri*) have reported a wide range of pharmacological properties, which are mainly associated with the phytochemical constituents present in the plant. Preliminary qualitative phytochemical analysis revealed the presence of flavonoids, terpenoids, phenols, fatty acids, proteins, alkaloids, tannins, saponins, glycosides and amino acids. In this study, hepatoprotective activity was evaluated using Paracetamol and isoniazid-induced hepatotoxicity models. In the Paracetamol model, hepatotoxicity was produced by administering an oral dose of 750 mg/kg at 8th day and 16th day of 21 days activity. In the isoniazid-induced model, liver toxicity was induced through an oral administration of isoniazid at a dose of 50 mg/kg at initial 28 days followed by treatment for next 15 days. Rats treated with Paracetamol and isoniazid exhibited a significant increase in serum ALT, AST and ALP levels. At the end of the experimental period, the hematological parameters suggested enhanced therapeutic efficacy and improved liver function. Elevated serum levels of liver enzymes were observed initially. However, when compared with the control group, treatment with *P. niruri* significantly decreased the levels of liver enzymes and markedly increased albumin and total protein concentrations. The ethanolic leaf extract of *P. niruri* exhibited significant pharmacological activity by markedly improving hepatotoxic conditions, along with reducing cellular inflammation and necrotic damage. The extract of *P. niruri*, administration showed statistically significant hepatoprotective effects (** $p < 0.001$, * $p < 0.01$). The findings revealed that the hepatoprotective effect of the ethanolic extract of *P. niruri* leaves was comparable to that of the standard drug used in conventional therapy. Both low and high doses of the plant extract were evaluated against standard silymarin (100 mg/kg). The hepatoprotective potential observed in the ethanolic extract may be attributed to the presence of major bioactive constituents responsible for this activity. Therefore, the leaves of *P. niruri* may be considered a valuable natural remedy for the management and prevention of various liver disorders, contributing to improved human health.

Funding

None.

Conflict of Interest

None.

Author Contributions

All the authors contributed to the study.

References

1. Pratima H, Shiraguppi A, Joojagar P, Shah K, Cheeraladinni SS, Singh PS, et al. Phytochemical profile and hepatoprotective potentiality of *Phyllanthus* genus: a review. *J Pharm Pharmacol*. 2025;77(2):189-205. doi: 10.1093/jpp/rgae040.
2. Ezzat MI, Okba MM, Ahmed SH, El-Banna HA, Prince A, Mohamed SO, et al. In-depth hepatoprotective mechanistic study of *Phyllanthus niruri*: in vitro and in vivo studies and its chemical characterization. *PLoS One*. 2020;15(1):e0226185. doi: 10.1371/journal.pone.0226185.
3. Kaur N, Kaur B, Sirhindi G. Phytochemistry and pharmacology of *Phyllanthus niruri* L.: a review. *Phytother Res*. 2017;31(7):980-1004. doi: 10.1002/ptr.5825.
4. Bagalkotkar G, Sagineedu SR, Saad MS, Stanslas J. Phytochemicals from *Phyllanthus niruri* Linn. and their pharmacological properties: A review. *J Pharm Pharmacol*. 2006;58(12):1559-1570. doi: 10.1211/jpp.58.12.0001.
5. Harish R, Shivanandappa T. Antioxidant activity and hepatoprotective potential of *Phyllanthus niruri*. *Food Chem*. 2006;95(2):180-185. doi: 10.1016/j.foodchem.2004.11.049.
6. Bhattacharjee R, Sil PC. Protein isolate from the herb, *Phyllanthus niruri* L. (Euphorbiaceae), plays hepatoprotective role against carbon tetrachloride induced liver damage via its antioxidant properties. *Food Chem Toxicol*. 2007;45(5):817-826. doi: 10.1016/j.fct.2006.10.029.
7. Karri S, Girish C, Bammigatti C, Lakshmi NCP. Efficacy of *Phyllanthus niruri* on improving liver functions in patients with alcoholic hepatitis: a double-blind randomized controlled trial. *Indian J Pharmacol*. 2021;53(6):448-456. doi: 10.4103/ijp.IJP_540_20.
8. Ramachandran A, Jaeschke H. Mechanisms of acetaminophen hepatotoxicity and their translation to the human pathophysiology. *J Clin Transl Res*. 2017;3(Suppl 1):157-169. doi: 10.18053/jctres.03.2017S1.002.
9. Jaeschke H, Ramachandran A. Acetaminophen hepatotoxicity: paradigm for understanding mechanisms of drug-induced liver injury. *Annu Rev Pathol*. 2024;19:453-478. doi: 10.1146/annurev-pathmechdis-051122-094016.
10. Jaeschke H, Ramachandran A. Central mechanisms of acetaminophen hepatotoxicity: mitochondrial

- dysfunction by protein adducts and oxidant stress. *Drug Metab Dispos.* 2024;52(8):712-721. doi: 10.1124/dmd.123.001279.
11. Jaeschke H, Williams CD, Ramachandran A, Bajt ML. Acetaminophen hepatotoxicity and repair: the role of sterile inflammation and innate immunity. *Liver Int.* 2012;32(1):8-20. doi: 10.1111/j.1478-3231.2011.02501.x.
 12. Jaeschke H, McGill MR, Williams CD, Ramachandran A. Current issues with acetaminophen hepatotoxicity—a clinically relevant model to test the efficacy of natural products. *Life Sci.* 2011;88(17-18):737-745. doi: 10.1016/j.lfs.2011.01.025.
 13. Ramachandran A, Jaeschke H. Acetaminophen hepatotoxicity: a mitochondrial perspective. *Adv Pharmacol.* 2019;85:195-219. doi: 10.1016/bs.apha.2019.01.007.
 14. Ramachandran A, Jaeschke H. Acetaminophen toxicity: novel insights into mechanisms and future perspectives. *Gene Expr.* 2018;18(1):19-30. doi: 10.3727/105221617X15084371374138.
 15. Sarich TC, Zhou T, Adams SP, Bain AI, Wall RA, Wright JM. A model of isoniazid-induced hepatotoxicity in rabbits. *J Pharmacol Toxicol Methods.* 1995;34(2):109-116. doi: 10.1016/1056-8719(95)00044-I.
 16. Lee NYS, Khoo WKS, Adnan MA, Mahalingam TP, Fernandez AR, Jeevaratnam K. The pharmacological potential of *Phyllanthus niruri*. *J Pharm Pharmacol.* 2016;68(8):953-969. doi: 10.1111/jphp.12565.
 17. European Association for the Study of the Liver. EASL Clinical Practice Guidelines: drug-induced liver injury. *J Hepatol.* 2019;70(6):1222-1261. doi: 10.1016/j.jhep.2019.02.014.
 18. McGill MR, Jaeschke H. Biomarkers of drug-induced liver injury. *Adv Pharmacol.* 2019;85:221-239. doi: 10.1016/bs.apha.2019.02.001.
 19. McGill MR, Jaeschke H. Biomarkers of drug-induced liver injury: progress and utility in research, medicine and regulation. *Expert Rev Mol Diagn.* 2018;18(9):797-807. doi: 10.1080/14737159.2018.1508998.
 20. Meunier L, Larrey D. Drug-induced liver injury: biomarkers, requirements, candidates and validation. *Front Pharmacol.* 2019;10:1482. doi: 10.3389/fphar.2019.01482.
 21. Shaikh JR, Patil MK. Qualitative tests for preliminary phytochemical screening: an overview. *Int J Chem Stud.* 2020;8(2):603-608. doi: 10.22271/chemi.2020.v8.i2i.8834.
 22. McGill MR, Sharpe MR, Williams CD, Taha M, Curry SC, Jaeschke H. The mechanism underlying acetaminophen-induced hepatotoxicity in humans and mice involves mitochondrial damage and nuclear DNA fragmentation. *J Clin Invest.* 2012;122(4):1574-1583. doi: 10.1172/JCI59755.
 23. Brennan PN, Cartledge P, Manship T, Dillon JF. Guideline review: EASL clinical practice guidelines: drug-induced liver injury (DILI). *Frontline Gastroenterol.* 2022;13(4):332-336. doi: 10.1136/flgastro-2021-101886.
 24. Jaeschke H, McGill MR, Ramachandran A. Oxidant stress, mitochondria and cell death mechanisms in drug-induced liver injury: lessons learned from acetaminophen hepatotoxicity. *Drug Metab Rev.* 2012;44(1):88-106. doi: 10.3109/03602532.2011.602688.
 25. Jaeschke H, McGill MR. Cytochrome P450-derived versus mitochondrial oxidant stress in acetaminophen hepatotoxicity. *Toxicol Lett.* 2015;235(3):216-221. doi: 10.1016/j.toxlet.2015.04.002.
 26. McGill MR, Williams CD, Xie Y, Ramachandran A, Jaeschke H. Acetaminophen-induced liver injury in rats and mice: comparison of protein adducts, mitochondrial dysfunction and oxidative stress. *J Pharmacol Exp Ther.* 2012;342(3):712-719. doi: 10.1124/jpet.112.194449.
 27. McGill MR, Jaeschke H. Animal models of drug-induced liver injury. *Biochim Biophys Acta Mol Basis Dis.* 2019;1865(5):1031-1039. doi: 10.1016/j.bbadis.2018.08.037.
 28. Kaplowitz N. Drug-induced liver injury. *Clin Infect Dis.* 2004;38(Suppl 2):S44-S48. doi: 10.1086/381446.
 29. Andrade RJ, Aithal GP, Björnsson ES, Kaplowitz N, Kullak-Ublick GA, Larrey D, et al. EASL Clinical Practice Guidelines: drug-induced liver injury. *J Hepatol.* 2019;70(6):1222-1261. doi: 10.1016/j.jhep.2019.02.014.
 30. Jaeschke H, McGill MR, Ramachandran A. Oxidant stress and acetaminophen hepatotoxicity: mechanisms and clinical implications. *Drug Metab Rev.* 2012;44(1):88-106. doi: 10.3109/03602532.2011.602688.
 31. Sarich TC, Adams SP, Petricca G, Wright JM. Inhibition of isoniazid-induced hepatotoxicity in rabbits by pretreatment with antioxidants. [Verify the article record before using this citation.]
 32. Viswambharan VL, Halagali P, Seenivasan R, Tippavajhala VK, Lobo R. Exploring the multifaceted medicinal benefits of *Phyllanthus niruri*: insights into its antiurolithic, hepatoprotective and anti-diabetic activities. *Curr Pharm Des.* 2026. doi: 10.2174/0113816128432792260122125217.

Citation: Sarswat D, Quamuddin M, Nimesh S, et al. Evaluation of the Hepatoprotective Effect of *Phyllanthus niruri* in Paracetamol and Isoniazid-Induced Hepatotoxicity in Albino Wistar Rat. *J Pharm Res Sci Technol* 2026; 10(2): 213. doi: [10.31531/jprst.1000213](https://doi.org/10.31531/jprst.1000213)

-
- | | |
|--|---|
| 33. Jaeschke H, McGill MR, Ramachandran A. Current issues with acetaminophen hepatotoxicity—a clinically relevant model to test the efficacy of natural products. <i>Life Sci.</i> 2011;88(17-18):737-745. doi: 10.1016/j.lfs.2011.01.025. | 34. Ramachandran A, Jaeschke H. Acetaminophen hepatotoxicity: a mitochondrial perspective. <i>Adv Pharmacol.</i> 2019;85:195-219. doi: 10.1016/bs.apha.2019.01.007. |
|--|---|

Copyright: ©2026 Sarswat D, et al. This article is distributed under the terms of the Creative Commons Attribution 4.0 International License [<http://creativecommons.org/licenses/by/4.0/>], which permits unrestricted use, distribution, and reproduction in any medium, provided you give appropriate credit to the original author[s] and the source, provide a link to the Creative Commons license, and indicate if changes were made.

