Journal of Engineering Science and Technology
Vol. 18, No. 1 (2023) 508 - 520

HEPATOPROTECTIVE PROPERTY OF PERUVIAN GROUND APPLE (*Smallanthus Sonchifolius*) TUBER AGAINST PARACETAMOL-INDUCED LIVER DAMAGE IN MICE

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Abstract

Peruvian ground apple (*Smallanthus sonchifolius* (Poepp. et Endel.) H. Robinson) tuber was investigated to evaluate its antioxidant and hepatoprotective activity against liver damage induced by paracetamol. Liver damage was induced in experimental animals by administering paracetamol orally. For seven consecutive days, varied treatments Per (water, 100 mg/kg of Silymarin, and 40.32 ml/kg of Peruvian ground apple tuber extract) were administered with Silymarin as the positive control drug. Serum marker enzymes like aspartate transaminase and serum glutamic-oxaloacetic transaminase in blood were used to determine the hepatoprotective effect. There was a significant elevation of the activities of the marker enzymes in paracetamol-treated mice, which have been considerably recovered closer to a nearly average level in animals administered with extract of tubers of Peruvian ground apple. Percent change in the initial and final SGPT levels of Peruvian ground apple and Sylimarin are not significantly different, suggesting that they have comparable effects in protecting the liver from damage. Histopathological analysis confirmed the biochemical investigations where Silymarin and Peruvian ground apple treated Swiss mice exhibited reduced mean necrotic hepatic cells. The outcomes suggest that the tuber of Peruvian ground apple exhibits hepatoprotective property, which can be traced to the obtained total phenols content of 27.628 ug/mL of 1 mL of sample and total flavonoids of 4.762 ug/mL of the 500 uL of the sample (100 ppm).

Keywords: Antioxidant, Flavonoids, Hepatotoxicity, Liver damage, Phenolics.

1. Introduction

Peruvian ground apple (*Smallanthus sonchifolius* (Poepp. et Endl.) H. Robinson) has been reported to prevent diabetes, regulate blood pressure, control cholesterol levels, helps in weight loss, improve digestion and prevent cancer. Phytochemical screening revealed that Peruvian ground apple tuber contains flavonoids and phenolics. The result of the antioxidant property analysis using the mean absorbance and the percent % of free radical inhibition using the DPPH Assay method revealed an antioxidant property of Peruvian ground apple. ANOVA results showed a significant difference in the antioxidant property in the mean absorbance of the plant's tuber extract and the standard drug ascorbic acid. Further, the results for both the tuber extract and the standard drug ascorbic acid show that their antioxidant property is dose or concentration-dependent; that is, increasing the dose or concentration results in a more significant antioxidant activity for both [1]. Although several studies have been conducted about Peruvian ground apple, like in the study of Salas et al. [2] on the hepatoprotective of *S. sonchifolius* leaves. Result revealed that the leaves of *S. sonchifolius* have hepatoprotective effect in a model of acetaminophen poisoning in rats. Studies on the hepatoprotective effects of Peruvian ground apple tubers have not been conducted. To be specific, there is a research gap in terms of the protective effects of Peruvian ground apple tuber on paracetamol-induced mice.

Plants are endowed with free radical scavenging molecules, such as vitamins, terpenoids, phenolic acids, lignins, stilbenes, tannins, flavonoids, quinones, coumarins, alkaloids, amines, betalains, and other metabolites, which are rich in antioxidant activity. The intake of antioxidant compounds present in food is an important health- protecting factor. Natural antioxidants present in food and other biological materials have attracted considerable interest because of their presumed safety and potential nutritional and therapeutic effects. An imbalance between too much oxidative stress and too little antioxidative defence has been suggested to cause a variety of liver diseases. Therefore, antioxidant could have a potential role in patients with liver disease.

Statistics from the Centers for Disease Control and Prevention (CDC) rank mortality related to chronic liver disease and cirrhosis as the 12th most common cause of death in adults in the US in November 2010. Using a modified definition that includes diseases such as viral hepatitis, liver cancer and obesity-related fatty liver disease (liver diseases), Mayo Clinic-led researchers have found that liver-related mortality is as high as fourth for some age groups, and eighth overall [3]. According to WHO, about 46% of global diseases and 59% of the mortality is because of chronic diseases and almost 35 million people in the world die of chronic diseases. Liver disease rates are steadily increasing over the years. According to National statistics in the UK, liver diseases have been ranked as the fifth most common cause of death. Liver diseases are recognized as the second leading cause of mortality amongst all digestive diseases in the US [4]. Likewise, on the latest WHO data published in in 2020 Liver Disease Deaths in Philippines reached 7,076 or 1.05% of total deaths. The age adjusted Death Rate is 8.65 per 100,000 of population ranks Philippines #135 in the world. [5].

Due to the availability of hepatotoxic drugs over the counter and alcohol consumption, liver toxicity is one of the leading causes of mortality in both developed and developing countries. A well-known example of drug-induced

toxicity is paracetamol. Paracetamol is an over-the-counter analgesic and antipyretic drug that is extensively used. When administered in multiple doses or taken of a large single dose, this is when it results to a significant hepatotoxicity. Toxic liver damage from drugs and chemicals can mimic virtually any form of naturally occurring liver disease. Hepatoprotective effects have been investigated against chemicals and drugs that have caused hepatotoxicity in rats, such as alcohol, CCl4, galactosamine, paracetamol, isoniazid, and rifampicin, etc. [6]. Liver damage can be diagnosed through several tests to determine the liver's health by measuring the levels of proteins, liver enzymes, or bilirubin in the blood. However, most of this test does not determine the overall function of the liver.

Therefore, the aim of the research was to evaluate the protective effect of Peruvian ground apple tubers against paracetamol-induced liver injury *in vitro* and *in vivo*. Total Phenol Content (TPC) and Total Flavonoid Content (TFC) are determined. For investigating the hepatoprotective effects of Peruvian ground apple tuber in vivo, the mice experiments were carried out, and the biochemical parameters in serum, including the level of SGPT, SGOT were also measured. In addition, histopathological examinations also have been conducted.

In order to prove the efficacy of Peruvian ground apple tuber for the treatment of illnesses, scientific studies were carried out through the quantification and isolation of its active constituents, its pharmacological action, and its proper way of administration, hence this study. Results of the study can serve as benchmark information as a basis for conducting further an extensive pharmacologic property testing of the plant not only via *in-vitro* experiments but also via *in-vivo* experiments.

2. Experimental Procedure

Smallanthus sonchifolius tuber was obtained from Baguio City, Benguet. The tubers were washed thoroughly with distilled water. The obtained samples were brought to the Natural Science Research Unit, St. Louis University, Baguio City for the Quantitative Phytochemical Analysis and the University of Northern Philippines, College of Health Sciences for the Toxicity testing and hepatoprotective and histopathological examination.

2.1. Measurement of total phenols and flavonoids

Total flavonoid content (TFC) was determined by the colorimetric method of aluminum chloride. Five hundred microliters of alcoholic extracts (100ppm) were separately mixed with 1.5 mL of 95% ethanol, 0.1 mL of 10% aluminum chloride, 0.1 mL of 1M potassium acetate, and 2.8 mL of distilled water. After incubating for 30 minutes at room temperature, the absorbance of the reaction mixture was reduced to 415 nm against a blank. In the blank, 10% aluminum chloride was substituted by the same amount of distilled water. Similarly, 500 μL of diluted standard solutions of quercetin in 80% ethanol (5, 10, 40, and 80 μg/mL) were reacted with aluminum chloride as described above to prepare the calibration curve for the concentration of flavonoids in samples were derived. Total polyphenol content (TPC) was measured spectrophotometrically with gallic acid as the standard according to the method described. in ISO 14502-1:2005 with slight modification. Briefly, 1.0 mL of the diluted sample extract was transferred in triplicate to separate tubes containing 5 mL of a 1/10 dilution of Folin- Ciocalteu phenol reagent in water. Within 3 to 8 minutes after adding the dilute Folin-

Ciocalteu phenol reagent, 4.0 mL of a sodium carbonate solution (7.5% w/v) was added. The tubes were then left at room temperature for 60 minutes before measuring its absorbance at 765 nm for the reagent blank. The concentration of polyphenols in the sample was determined from the standard curve of gallic acid in the following range 10 to 50 μg/mL.

2.2. Laboratory animals

Nine male Swiss mice aged eight weeks old, weighing 30- 32g from the MMSU- Laboratory Animal Care Facility (LACF), were used in the study. The animals were kept in 12 x 8.5 x 7 inches plastic cages with metal screen cover at room temperature (20-26°C), with 55- 75% humidity, ventilation of at least 10-15 fresh air changes per hour, and under natural light/darkness cycles (12 hours light and 12 hours dark cycle). Each cage contained three mice according to their replicated treatment. The animals were allowed access to feed and water *ad libitum* and allowed seven days to acclimatize before the commencement of the experiment. The food supply was withdrawn 8 hours before the experiment. Animals have maintained following the National Research Council of the National Academies recommendation on the guide for the laboratory animals care and use [7].

2.3. Dosing method

Administration of treatment is via gastric intubation using an 18-20 gauge feeding tube (gavage) about 1.5 inches in length with a rounded tip. The maximum dosing volume is 40 mg/kg for mice. Dosing was only done once within 24 hours for seven days [8].

2.4. Experimental design

Nine male Swiss albino mice were randomly divided into three groups of three test organism each and treated. Group I was given water. Group II was given a single daily dose of Silymarin (100mg/kg) orally. Group III was given orally a single daily dose of Peruvian ground apple tuber extract (40.32ml/kg). Each group of mice received the respective dose of treatment solution orally via oral gavage for seven consecutive days. The oral administration of paracetamol hepatotoxicity was performed on the last day (7th day). Two hours after the last treatment, each mouse received one dosage of paracetamol, and the animals fasted for 12-14 hours. After 12-14 hours of fasting, blood was extracted from the tail end of the mice to assess liver damage for the final analysis of biochemical parameter SGPT. On the 3rd day of the study, blood was extracted on the tail of mice via the tail clipping method for the analysis of initial marker enzymes parameters aspartate aminotransferase (AST or SGPT). The serum was prepared using the standard method. Blood was allowed to clot for 30 minutes and then centrifuged at 2500 rpm for 15 minutes, and serum was harvested using a pipette. Liver damage was assessed by estimating serum activities of Alanine transaminase (ALT or SGPT) before and after the treatments were given [9, 10].

2.5. Histological study

The animals were sacrificed by cervical dislocation, and the livers were excised immediately, washed with chilled normal saline. One part of the liver tissues

isolated from the test animals were fixed in 10% formalin for 24h. The fixed tissues were processed manually through graded ethanol, cleared in xylene, impregnated, and embedded in paraffin wax. Thin sections were cut with a rotary microtome, stained by hematoxylin and eosin technique, and were examined microscopically for pathological changes.

2.6. Statistical analysis

The data were analysed using SPSS 26 for windows. All variables were compared using a one-way analysis of variance (ANOVA) followed by an LSD post hoc test. $P \leq 0.01$ was considered statistically significant [11].

3. Results and Discussion

Phenolic compounds ubiquitous in plants are an essential part of the human diet and are of considerable interest due to their antioxidant properties. These compounds possess an aromatic ring bearing one or more hydroxyl groups. Their structures may range from that of a simple phenolic molecule to that of a complex high-molecular-weight polymer. Flavonoids, which bear the C_6-C_3-C_6 structure, account for more than half of the over eight thousand different phenolic compounds. The antioxidant activity of phenolic compounds depends on the structure, particularly the number and positions of the hydroxyl groups and the nature of substitutions on the aromatic rings [12].

Total phenolic content (TPC) and total flavonoid content (TFC)

The data presented in Table 1 illustrate the values of total flavonoids as mg quercetin equivalent (QE)/g of the extract and total phenols as mg gallic acid equivalents (GAEs)/g of the extract. The result of the test revealed that 500µL of sample extract (100 ppm) contained Total Flavonoid Content of 4.762µg/mL and 1mL of sample extract contained Total Phenol Content of 27.628µg/mL.

Flavonoids are just one of the types of phenolic compounds. From the quantitative analysis, 27.628µg are phenolics, and out of this, there are 4.762 µg flavonoids.

Table 1. Total flavonoids and phenols of Peruvian ground apple tuber extract.

Sample	Quantitative Value (µg/mL)*
Total Phenolics	27.628
Total Flavonoids	4.762

3.1. Hepatoprotective Property of Peruvian ground apple (*Smallanthus sonchifolius*) Crude Extract, Silymarin and Water in Swiss Mice through Enzyme Markers

To evaluate the hepatoprotective property of Peruvian ground apple *(Smallanthus sonchifolius)* tuber extract, the SGPT analysis was performed. Prior to the SGPT analysis, the initial and final body weights of Swiss mice were determined as summarized in Table 2. The body weight is an essential factor in monitoring an

individual's health to analyse the impact of treatments. Furthermore, the loss in body weight is frequently the first indicator of the onset of adverse effects [13].

Table 2. Initial and Final mean body weight (grams) of mice in different treatments.

Treatments	Initial	Final
Untreated	35.3	35.3
Silymarin	34.0	34.7
Peruvian ground apple Extract	34.0	34.0
F-Test	ns	ns

Legend: ns-not significant

Table 2 shows mice's initial and final mean body weight in different treatments after acclimatization and after the final application. The mean initial body weights of the mice range from 34 to 35 grams, and the mean final body weight ranges from 34 to 35 grams. Based on the analysis of variance, there was no significant difference in the initial and final body weight of mice between the control and extract.

3.2. Effects of the different treatments on hepatotoxicity marker enzymes

One of the capacities of the liver is to cleanse and eliminate the harmful constituents in the bloodstream. The liver is accountable for managing alcohol not to cause impairment to the rest of the system. It also processes other drugs that get in the system, including medicines and leisure drugs. Paracetamol or Acetaminophen as hepatotoxin can damage the liver. Acute acetaminophen (APAP) overdose in mice and humans causes centrilobular necrosis in the liver within hours [14]. To further determine the effects of paracetamol as hepatotoxin in the test animals, the hepatoprotective test was performed.

A liver function test was conducted through a serum assay that gives information about the state of the liver. The concentration of Serum Glutamic Pyruvic Transaminase (SGPT) is commonly used as a biochemical marker of hepatocellular damage. Li et al. found out in 2017 that the highest concentration of ALT is found in liver cells; moderate amounts are found in kidney cells; and smaller amounts are found in the heart, pancreas, spleen, skeletal muscle, and red blood cells. When liver damage occurs, serum levels of ALT may increase as much as 50 times on average, making this a sensitive test for evaluating liver function. ALT is part of a group of tests known as LFTs or liver function tests used to evaluate liver function. ALT, AST, and Alp are liver enzyme markers, and elevated levels of these markers in serum indicate the loss of hepatocyte structural integrity and liver injury. ALT is a more liver-specific enzyme among the enzyme markers. In addition, increased ALT activity lasts longer than increased AST activity [15].

To further evaluate the hepatoprotective property of the *S. sonchifolius* tuber extract, the initial and final SGPT enzyme levels were compared. Escalation in SGPT enzyme level signifies liver damage, and a decline in SGPT level indicates protection to the liver. The total average of initial and final SGPT of mice in different treatments are presented in Table 3.

Table 3. Total Average of initial and final SGPT (μg/dl) of mice in different treatments.

Treatments	Initial	Final	Percent % Change
Untreated	64.7[a]	69.1[a]	-6.80[a]
Silymarin	68.3[a]	65.0[a]	4.83[b]
Peruvian ground apple Extract	86.3[b]	84.3[b]	2.32[b]
F-Test	**	**	**

Legend: **-significant at 0.01 level

The table reveals that the untreated mice exhibited a final average SGPT level of 69.1 and had a negative percentage change of 6.80 which shows a percentage increase: when paracetamol was induced, it indicates inflammation or damage to cells in the liver due to the harmful effects of paracetamol in this organ. High levels of ALT may be a sign of liver damage from hepatitis, infection, cirrhosis, liver cancer, or other liver diseases. [16]. Silymarin's hepatoprotective and antioxidant activity is due to its ability to inhibit free radicals resulting from the metabolism of toxic substances such as ethanol, acetaminophen and carbon tetrachloride [17]. The production of free radicals is known to damage cell membranes and cause lipid peroxidation. Silymarin can increase liver glutathione and contribute to the liver's antioxidant defences. Moreover, the high prevalence of liver diseases such as cirrhosis and chronic hepatitis requires efficient and cost-effective treatments. In this context, Silymarin, an extract from the seeds of *Silybum marianum* or milk thistle, is widely used as promising over-the-counter drugs for the treatment of liver disease [18]. Moreover, the Peruvian ground apple-treated mice exhibited a very high level of initial and final SGPT. According to the Clinical Biochemistry Values [18] of different animal species, mice have a normal Aspartate Aminotransferase (AST/SGOT) level of 55-251 μ/L and Alanine Aminotransferase (ALT, SGPT) level of 28-184 μ/L. Since the Peruvian ground apple-treated mice exhibited the highest initial and final SGPT between the two treatments, factors that contributed to the high levels were considered.

According to Kim et al. [19], ALT activity varies day to day, by 10% to 30%. Within a given day, there is a significant diurnal variation, with ALT activities being up to 45% higher in the afternoon than in the early morning. In acute hepatocellular injury, serum AST levels usually rise immediately, reaching a higher level than ALT initially, due to the higher activity of AST in hepatocytes and its release with liver injury. Within 24 to 48 hours, particularly if ongoing damage occurs, ALT will become higher than AST, because of its longer plasma half-life. In chronic hepatocellular injury, ALT is more commonly elevated than AST; however, as fibrosis progresses, ALT activities typically decline, and the ratio of AST to ALT gradually increases, so that by the time cirrhosis is present, AST is often higher than ALT. One notable exception to the predominance of serum ALT activity in chronic liver disease is alcoholic liver disease where AST activity is generally higher than ALT levels.

Additional factors that affect serum ALT levels include body mass index (BMI) and triglyceride levels, regardless of gender. Total cholesterol levels and alcohol consumption among men have a positive correlation, whereas smoking, physical activity and age have a negative correlation with ALT levels. Glucose levels, have a positive correlation with ALT activities, whereas use of oral contraceptives tends

to lower ALT values. Some of these correlations (such as BMI) may be explained by ALT being higher with fatty liver disease. Similarly, patients with hyperlipidemia or hyperglycemia may also have fatty liver disease, as a part of the metabolic syndrome. Since serum ALT levels rise in disease states that cause hepatocellular injury, serum ALT levels can effectively identify an ongoing liver disease process. The probability of clinically significant liver disease increases, particularly if the elevated ALT is associated with symptoms such as fatigue, anorexia or pruritus.

The SGPT level of Silymarin and Peruvian ground apple extract reduced from the initial result of 68.3 and 86.3 μg/dl to the final result of 65 and 84.3 μg/dl after hepatotoxin was induced. This indicates that Silymarin and Peruvian ground apple crude extract have the capability to protect the liver when a toxin is induced. Analysis of Variance revealed that treatment and controls are significantly different. It also revealed significant difference in the percent (%) change between the initial and final SGPT level.

3.3. Comparison of Hepatoprotective Property of Peruvian ground apple (Smallanthus sonchifolius) Crude Extract, Silymarin and Water in Swiss Mice through Histological Examination

The liver is the heaviest organ in the body and is responsible for functions vital to life. The liver is composed of hepatic lobules, the building blocks of the liver parenchyma tissue consisting of a portal triad, hepatocytes arranged in linear cords between a capillary network, and a central vein. The parenchyma cells of the liver are otherwise known as hepatocytes, making up to 70 to 80 % of the liver's cytoplasmic mass. The hepatocytes are arranged closely packed in normal liver tissue, no visible signs of necrosis (cell death), and the central vein is well-oriented and not damaged.

The mechanisms and the sequence of events by which free radicals interfere with cellular functions are not fully understood. Still, one of the most important events seems to be lipid peroxidation, which results in cellular membrane damage. This cellular damage causes a shift in the net charge of the cell, changing the osmotic pressure, leading to swelling and eventually cell death. Free radicals are molecules, usually of oxygen, that have lost an electron. That loss makes them unstable and reactive. They begin to covet their neighboring molecules' electrons. In stealing an electron, they operate as terrorists in the body. The most dangerous free radicals are the small, mobile, and highly reactive oxy radicals. Other dangerous atomic and molecular varieties of oxygen are known as reactive oxygen species (ROS). [20]. In this study, histopathological observation of the liver was performed to further support the hepatoprotective property of Peruvian ground apple tuber extract together with the positive and harmful treatments.

To thoroughly determine the effects of each treatment in the liver of the test animals, three liver lobes were chosen, the left, middle, and right lobe, from one of the replicates in each treatment. The liver slides were observed in a microscope with a 40x (HPO) magnification using a compound microscope; the image was captured using a built-in camera with a resolution of 1920x1080 pixels.

Figures 1(a), 1(b), and 1(c) have normal hepatic cells. However, the rest of the figures, i.e., 1(d) to 1(l) showed necrotic hepatic cells. Necrosis is a form of cell

injury caused by external factors to cells or tissue, such as infection and toxins, which result in the unregulated digestion of cell components. The extreme increase of serum behaviors of liver enzyme markers (SGPT or ALT) in untreated mice was reflected from the liver cell death after the introduction of paracetamol.

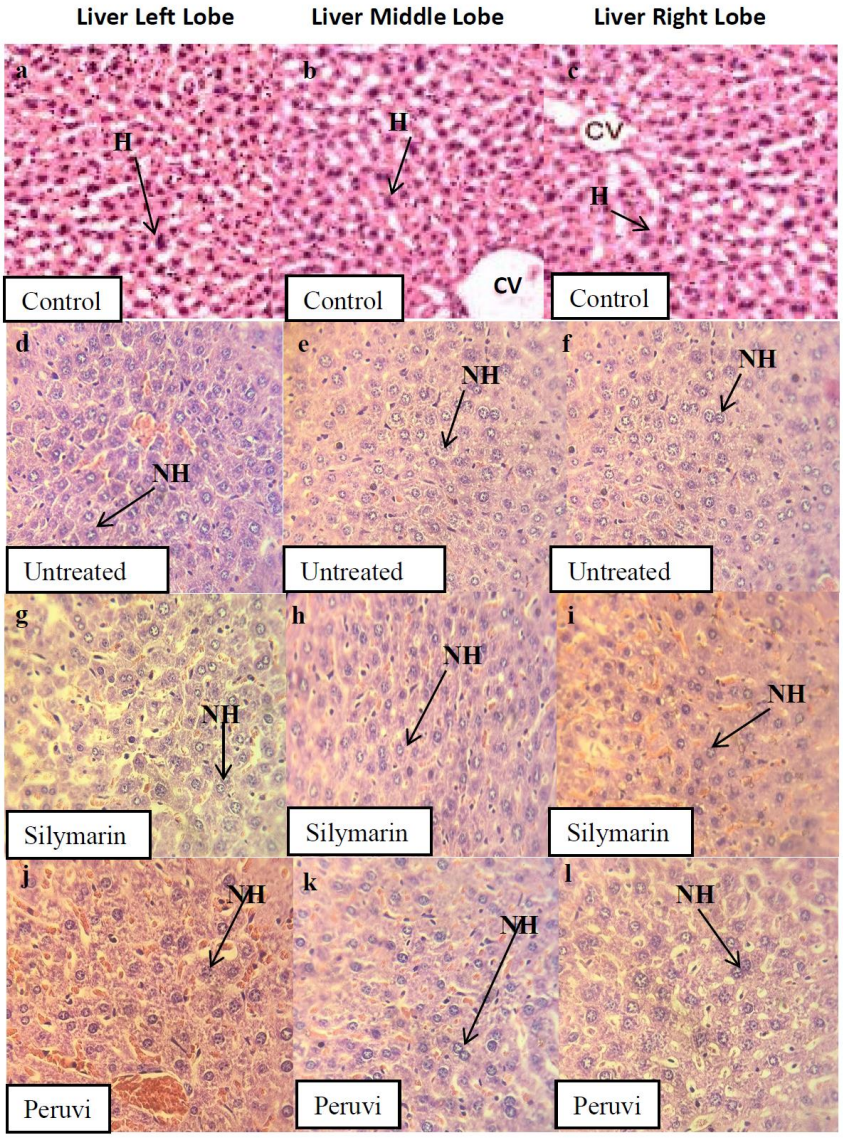

Fig. 1. Photomicrograph of left, middle, and right liver lobes stained with H&E showing the following: Normal architecture with the central vein (CV) surrounded by normal hepatocytes (H). Destruction of hepatic architecture or necrotic hepatocytes found in the untreated, Silymarin and Peruvian ground apple groups. (H&EX40).

Hepatocytes can be chronically infected by hepatotropic viruses, which can cause chronic liver injury. Alcohol and drugs such as Acetaminophen are metabolized and detoxified in the liver and, in excess, can damage the liver [21].

Three random areas of each lobe were observed using a microscope under a total magnification of 40x, and only the necrotic hepatic cells were counted. Necrosis, also known as death of a cell, is the process by which living cells become incapable of survival and the contents of the cells are destroyed. Under physiological conditions, it is a healthy and regulated process that leads to the exchange of individual old cells. However, necrosis is the result of various tissue damage.

Table 4. Mean necrotic hepatic cells in mice given varied treatments.

Treatments l	Liver Left Lobe	Liver Middle Lobe	Liver Right Lobe
Normal	0	0	0
Untreated	126^{a}	108^{a}	107^{a}
Silymarin	59^{c}	59^{b}	59^{b}
Peruvian ground apple Extract	85^{b}	69^{b}	88^{a}
F-Test	**	**	**

Legend: **-significant at 0.01 level

The untreated mice's left, middle, and right liver lobe revealed 126, 108, 107 necrotic hepatic cells, respectively, and revealed the highest number of hepatocellular injuries among the treatments. Peruvian ground apple extract-treated mice revealed 85, 69, 88 necrotic hepatic cells in their left, middle, and right lobe. Mice treated with Silymarin revealed 59 necrotic hepatic cells in each lobe and revealed the lowest number of hepatocellular necrosis or injury.

Moreover, Analysis of Variance shows a significant difference between and among the treatments used in terms of mean necrotic hepatic cells in all liver lobes of mice; this implies that the treatments are not comparable. To determine where the difference or differences lie, Scheffe Test was employed.

Scheffe test revealed a significant difference between Peruvian ground apple and untreated mice and Peruvian ground apple and Silymarin in terms of mean necrotic hepatic cells in the left liver lobe. However, Peruvian ground apple and Silymarin are not significantly different in the middle liver lobe in terms of mean necrotic hepatic cells. Moreover, there is a significant difference between Peruvian ground apple and Silymarin and Peruvian ground apple and untreated mice in terms of mean necrotic hepatic cells in the right liver lobe. Since Silymarin has the least number of necrotic hepatic cells and the least mean the difference among the three treatments, Silymarin as a commercially available medicine for liver protection is more effective than Peruvian ground apple.

The abovementioned result is congruent with the earlier result of the percent (%) change between the initial and final SGPT levels. It revealed a significant difference between and among the treatments.

The flavonoid of Silymarin and one of its components, called silybinin, is a substance with documented hepatoprotective activities. In the Peruvian ground apple tuber extract, the presence of secondary metabolites such as flavonoid and other phenolic compounds support the extracts' potential to protect the Swiss mice

liver from damage as shown in the photomicrographs and result of the mean necrotic hepatic cells [22].

4. Conclusions

An investigation has been made to evaluate its antioxidant and hepatoprotective activity against liver damage induced by paracetamol. Some concluding observations from the investigation are given below.

- Peruvian ground apple tuber extract showed a hepatoprotective effect in the liver of Swiss mice. This is due to the presence of phytochemicals, flavonoids, and phenols that have been earlier proven using DPPH Assay to contain an antioxidant activity.
- The flavonoids component is recognized to possess the hepatoprotective effect.
- The Peruvian ground apple has a moderate amount of phenols and flavonoids.

Nomenclatures

QE/g	Quercetin equivalent per gram
GAEs/g	Gallic acid equivalents per gram

Abbreviations

ALT	Alanine transaminase
ANOVA	Analysis of Variance
APAP	Acute acetaminophen
AST	Aspartate aminotransferase
CV	Central vein
DPPH	2,2-diphenyl-1-picrylhydrazyl
H	Hepatocyte
H&E	Hematoxylin and eosin
HPO	High Power Objective
LACF	Laboratory Animal Care Facility
LSD	Least Significant Difference
MMSU	Mariano Marcos State University
SGOT	Serum glutamic-oxaloacetic transaminase
SGPT	Serum glutamic pyruvic transaminase
SPSS	Statistical Package for the Social Sciences
TFC	Total Flavonoid Content
TPC	Total Phenolic Content

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