ORIGINAL ARTICLE

INTERNATIONAL JOURNAL OF RESEARCH IN PHARMACEUTICAL SCIENCES

Published by JK Welfare & Pharmascope Foundation Journal Home Page: https://ijrps.com

Phytochemical and Hepatoprotective Activity of Prosopis juliflora Barks on Different Models

Hemamalini K*[1], Sadanandam Palle[2], Kanakam Vijayabhaskar[3], Supraja[1]

[1]Swami Vivekananda Institute of pharmaceutical sciences, Vangapally, Yadagirigutta
[2]Chemistry Department, Malla reddy Engineering College, Hyderabad
[3]Department of Pharmacognosy, Bomma Institute of Pharmacy, Alipuram, Telangana, India

Article History:

Received on: 10 Jul 2023
Revised on: 26 Jul 2023
Accepted on: 28 Jul 2023

Keywords:

Methanolic Extract,
Barks,
Prosopis juliflora,
Hepatoprotective activity

ABSTRACT

Liver injury is a common disorder where natural remedies are practiced from ancient time from plant source which shows alternative treatment in a safe manner. The Hepatoprotective activity of methanolic extract of Prosopis juliflora bark part was taken for evaluation in screening. The model chosen for the study is carbon tetrachloride (0.1ml/kg) and the standard drug silymarin of 20mg/kg was used for the study for 21 days. The hepatoprotective effect of the methanolic extract against the carbon tetrachloride-induced hepatotoxicity in wistar rats has been demonstrated by the substantial and severe reduction of enzymes in the serum.

*Corresponding Author

Name: Hemamalini K
Phone: 9553601248
Email: rkhemamalini@gmail.com

ISSN: 0975-7538

DOI: https://doi.org/10.26452/ijrps.v14i3.4308

Production and Hosted by

IJRPS | https://ijrps.com

INTRODUCTION

Liver plays a vital role in metabolic and detoxification and excrete some endogenous and exogenous compounds thereby protect the body from toxic substances [1]. Due to the prolonged exposure of the xenobiotics and the liver metabolites liver injury occurs [2]. Liver plays a major role in fighting against the disease also involves in various biochemical pathways for growth. Liver has a regenerative property [3]. The liver injury is due to chemicals and even alcohol consumption, and also few drugs like paracetamol, antibiotic, antituberculosis and chemotherapeutic drugs [4, 5].

It was highly documented that hepatotoxicity can be studied in chemically induced model where it shows changes in biochemical pathways and pathological changes in liver [6, 7]. There are several complications like necrosis, jaundice, fibrosis, cirrhosis, hepatitis, and liver carcinoma due to hepatic damage [8]. Liver damage is the main disease where it leads a major role on global death. Several natural medicine plays a vital role to treat hepatic disease, and numerous synthetic drugs also play a major role but it possess numerous side effects [9]. Still, people rely on plant based therapy which was practiced 1000 years ago for liver diseases [10, 11]. Hence it is to explore the suitable remedy from natural source to replace the chemical ones. All plants are rich source of antioxidants which help to treat different diseased states. The plants contain different Phytochemical compounds with different antioxidant protection in different levels. Traditionally to treat liver ailments plants are more reliable and more efficient options. In tribal and in rural villages these medicinal plants are traditionally used to cure various ailments [12]. Natural process of healing the human body is through herbal compounds from ancient times [13, 14]. There is currently an increase in the amount of experimental work being done on the ethnopharmacology of herbal remedies. Therefore, a search is currently being conducted in several research fields for novel herbal medica-

tions that have a high level of safety and a superior potential. There are a variety of plant species with hepatoprotective properties on a variety of chemically induced models, and an evaluation is still being worked out in a variety of invivo models. Various chemical inducers have been used.

Carbon tetrachloride: (CCl4)

It is an organic solvent and chlorinated in nature when exposed it produces a toxic effect to various organ. It has an odour that is described as being sweet and ethereal but has no colour and is quite volatile. When heated, it decomposes and releases vapours of phosgene, which are highly hazardous. It plays a role in the manufacturing of chlorofluorocarbons and is also put to use in the creation of refrigerants. It is effective as a grain fumigant, dry cleaning agent, insecticide dispersion, and fire extinguisher [15]. It can also be used to treat helminth infections. The oral (mouth), inhalation (lungs), and dermal (skin) modes of carbon tetrachloride absorption are present in both human beings and other animals. This CCL4 is utilised in situations where it causes damage to the liver so that it can be used to assess the hepatoprotective activities of medicinal herbs.

Following metabolism, it undergoes a conversion that results in a nicotinamide adenine dinucleotide phosphate (NADPH)-dependent CYP450- 2E1 enzyme. This results in the formation of radicals that are free, trichloromethyl (CCl3). As a result of continued oxidation, this radical eventually transforms into trichloromethyl peroxy (O–O–CCl3) [16, 17]. Therefore, when free radicals such as oxygen interact with fatty acid cell membranes, they produce lipid peroxidation, which ultimately results in the formation of another reactive aldehyde (such as formaldehyde and acetaldehyde, among others). The glutathione level in the liver is lowered as a result of the generated aldehydes' subsequent reaction with glutathione. GSH is an antioxidant that helps protect cells from being damaged [9]. It produces free radicals cause DNA damage and lipid peroxidation leads to cell disruption and releases AST, ALT and bilirubin into blood stream [18]. So protein breakdown takes place and cell necrosis leads to cytotoxicity [19]. This finally alters the Ca^{2+} sequestration, cytokines release and loss of energy metabolism [20, 21]. Many used this model to induce liver cirrhosis in preclinical studies [22–24]. This chemical CCl4 cause liver damage as it mimic like natural damage, the changes that occur is finally destruction of hepatic cells [25–27] and it produce toxic effects in different organs kidneys, heart, lung, testis, brain and blood [28, 29]. The trichloromethyl radical is responsible for producing a genotoxic response in the liver [30]. It binds to guanine and adenine. This substance causes a considerable decrease in the activity of the microsomal enzymes aniline hydroxylase and p-aminipyrine-N-demethylase, as well as glucose-6-phosphatase and protein synthesis [31]. This chemical activates Kupffer cells [32, 33] that cause death of the hepatocytes [34, 35]. The fatty liver damage finally causes the destruction of the smooth and rough endoplasmic reticulum inhibits the protein synthesis, and reduces the golgi complex and mitochondria [36, 37]. For the complete damage of the liver nearly 3mL/Kg of carbon tetra chloride [38, 39] in an Intraperitoneal dose is required [40, 41]which causes the central veins and loss of cellular boundaries.

The plant known as Prosopis juliflora is a kind of tree that may be found in dry and semiarid regions all over the world. It is a member of the Legumiosae family and the Mimosoideae subfamily, and it has 44 different species. They are capable of growing in poor soils and possess binders and stabilisers that can be used for sand. It grows as a shrub with branches that are 8-12 metres long, can grow to a height of up to 12 metres, and has a stem that is 1.2 metres tall. The leaves fall off annually and have between 12 and 20 leaflets each. They have a light green colour. The flowers started to bloom once the leaf had fully matured. The phytochemical components that are present have some therapeutic significance and can change the way that certain physiological processes work in the human body [42].

Terpenes, alkaloids, flavonoids, and phenolic chemicals were identified as being present in the plant's biochemical profile. Insecticides, antifungal, antibacterial, anthelmintic, antimalarial, and molluscidal are some of the pharmacological effects that can be attributed to the active ingredients. Both the seed and the leaves exhibit a number of invitro antimicrobial, antifungal, and anti-inflammatory effects. Because of its high calorific value, this plant is also referred to as wooden anthracite. The wound healing, disinfectant, and treatment of scurvy [43] are just some of the applications for the leaf and seed extracts that were utilised to prepare the decoction. This decoction made from plants is effective for treating digestive disorders as well as skin blemishes. Properties such as sedative, astringent, antiseptic, antibacterial, and antifungal are possessed by this substance. A great number of indigenous peoples from a variety of countries use the plant to cure ocular conditions, open wounds, skin conditions, digestive issues, and dermatological conditions. Flavonoids are responsible for

over 16% of the antioxidant and anticancer effects. Tannins and phenols can also be found at low concentrations, and they work together to enhance those qualities. Phenols contain the ability to inhibit the activity of some enzymes that are responsible for inflammation [44].

MATERIALS AND METHODS

Materials:

Drugs

Carbon tetrachloride, silymarin and olive oil.

Plant collection:

Dr. Madhavachetty, a toxicologist from S.V. University in Tirupathi, authenticated the plant's aerial section. The voucher specimen was numbered, and the specimen itself was deposited in the SVIPS lab.

Extraction:

After the plant was harvested and dried, roughly 200 grammes of powder were taken, packed in a soxhlet apparatus, and extracted using two to two and a half litres of petroleum ether as a solvent. This allowed for defatting to occur throughout the extraction process, which took place at a temperature of forty degrees Celsius. After some time had passed, the extract was distilled, and after that, it was kept in the desiccators while the value of the percentage yield was calculated. The same mark was taken to several organic solvents in the order of their polarity, such as ethyl acetate, methanol, ethanol, and water. This was done in order to compare the results. Following the extraction process, a percentage yield of the extracts was determined [45] (Table 1).

Phytochemical screening

Following the completion of the drying process, the methanolic extract of the plant was put through a series of different chemical tests. These tests included the Mayer test, Wagner's test, Dragendroff test, and Hagner's test. Tests such as the Molish test, Benedict's test, and Fehling test were used to determine the presence of carbohydrates. Tests such as the modified Borndrager test and the legal test were used to analyse glycosides. Finally, tests such as the Salkowski.

The froth test, in addition to the gelatin test for tannins, is carried out. Examination of xanthoproteins The ninhydrin test is done to determine the presence of protein along with unbound amino acids.

Tests for flavonoids such as the alkaline reagent test, the lead acetate test, the shinoda test, the test for diterpene, the copper acetate test, and a screening procedure for phenols using the ferric chloride test [46].

Experimental animal:

For the purpose of this investigation, mice of any sex were obtained through Raghavendra enterprises and maintained in an animal house with a room temperature of 231 ^{o}C, relative humidity of 55 ^{o}C, and a 12 hr light /12 hr dark cycle. Throughout the course of the experiment, the mice were given a pellet diet along with water on a free-access basis. The animals were brought into the laboratory sixty minutes before the start of the experiment. The IAEC protocols were followed for all of the animals during the entire process.

Acute toxicity studies:

As per OECD-423 guidelines acute toxic test was performed for the methanolic extract of Prosopis juliflora using albino mice. We took 6 in number of either sex selected by random. The animals taken for the study are fasted prior before dosing for overnight. Before that the animals are weighed and after dosing the food is withheld for a further. The chosen dose 300, 500, 1000 and 2000 mg/kg body weight, 3500 and 5000 mg/kg and the 50 % death was monitored. When the mortality is observed then the same dose was repeated again to confirm the toxic dose [47] (Table 2).

Experimental design:

Assessment of Hepatoprotective activity:

The rats taken for the study are divided into different groups each group the number of animals placed is 6. Group I serves a control provided with saline for 21 days. Group II was administered with CCl_4 of 0.1ml/kg, p.o + olive oil of 0.1ml/kg, Group III is treated with CCl_4 of 0.1ml/kg, p.o + silymarin of 20mg/kg, Group IV is treated with CCl_4 of 0.1ml/kg, p.o + methanolic extract of Prosopis juliflora 200mg/kg, p.o, Group V is treated with 0.1ml/kg, p.o + methanolic extract of Prosopis juliflora of 400mg/kg, p.o. the animals were kept allowed for 24 hours after administration of the inducing agent the plant extract was administrated regularly for 21 days. After a period of observation that lasted for twenty-one days, the rodents were subjected into cardiac arrest by cervical dislocation, then their blood was taken by puncturing their hearts. After being separated and coagulated for a period of 30 minutes at 37 degrees Celsius, the blood specimen was then centrifuged at a rate of 300 revolutions per minute for a period of 15 minutes before being utilised for the detection of several biochemical parameters that include bilirubin, SGPT, and SGOT. After the liver had been taken by the process of dissection, it was washed in

Table 1: Percentage yield and colour consistency of plant extract Prosopis juliflora

Different solvents	Percentage yield (%) (w/w)	Colour-Consistency
Petroleum ether	12.1%	Greenish- Black Dry mass
n-Hexane	15.4%	Greenish- Black Dry mass
Ethyl acetate	18.3%	Greenish- Black Dry mass
Methanol	50.4%	Greenish- Black Dry mass
Ethanol	35.2%	Greenish- Black Dry mass
Water	10.4%	Greenish- Black Dry mass

Table 2: Determination of acute oral toxicity (LD50) of methanolic extract of *Prosopis juliflora*

Name of the study	Acute toxicity
Guideline followed	OECD 423 method
Animals	Swiss albino mice
Body weight	20 ± 5 g
Sex	Either sex
Administration of dose and volume	100 and 200 mg/kg body weight, single dose in 0.5 mL
Number of groups and animals	9 groups and 6 animals in each group.
Route of administration	Oral by using mice oral feeding needle
Vehicle	Distilled water

Table 3: Preliminary Phytochemical Screening of Prosopis juliflora

S.No	Phytoconstituents	Report
1	Alkaloids	Present
2	Carbohydrates	Present
3	Glycosides	Present
4	Phytosterol	Present
5	Saponins	Present
6	Tannins	Present
7	Proteins and free amino acids	Present
8	Flavonoids	Present
9	Diterpenes	Present

Table 4: Methanoli cextract of *Prosopis juliflora* (MEPJ)on serum enzymatic activity in CCl4 induced liver damage in rats (n=6)

Groups	SGOT(IU/L)	SGPT(IU/L)	ALP(IU/L)	SB (mg/dL)
I	314.0 ± 19.70	67.25 ± 6.223	203.3 ± 5.85	1.109 ± 0.0470
II	473.3 ± 15.67	73.50 ± 7.643	691.8 ± 18.75	1.221 ± 0.0530
III	281.8 ± 11.02***	45.75 ± 1.548***	393.5 ±20.95*	1.051 ± 0.01702***
IV	289.5 ± 1.936**	71.75 ±3.860**	309.5 ±35.50**	1.143 ± 0.01708**
V	357.3 ± 10.89*	81.50 ±5.123*	247.5 ±12.90***	1.239 ± 0.01958*

phosphate buffer saline that had been chilled to a very low temperature. In future research, 0.1 M, pH 7.4, and homogenate were utilised; following this, histopathology studies were carried out.

Statistical Analysis

The data are shown as the mean accompanied by the standard error of the mean and were analysed using one-way ANOVA, then TUKEY S multiple comparative tests. A statistical significance level of 0.05 was assigned to the P valves.

RESULTS

Preliminary Phytochemical: Phytochemicals such as alkaloids, carbohydrates, glycosides, phytosterol, saponins, tannins, proteins, Flavonoids and diterpenes (Table 3).

The hepatic damage of the rat was observed in an elevated levels in the specific enzymes as well as the parameters like SGPT, SGOT and serum bilirubin and ALP was increased in CCL4 treated, and the group treated with plant extract showed significant protection against the liver toxic like increased enzyme level and other content which were estimated at the time of screening (Table 4).

Histopathological studies:

A passage through the liver reveals the normal cellular architecture of the organ, which includes separate hepatic cells, sinusoidal gaps, and central veins. It was noticed that normal liver cells had become disorganised, exhibiting characteristics such as necrosis of the centrilobuli, vacuolization of the cytoplasm, and fatty degeneration. After being treated with an aqueous methanol extract of Prosopis juliflora, the liver segments of the rat that had been subjected to CCl4 poisoning showed signs of being protected, as evidenced by the lack of necrosis and vacuoles.

DISCUSSION

Carbon tetrachloride is a well-known chemical that is commonly utilised for inducing toxicity in a wide variety of laboratory animals. The free radicals undergo a second reaction with oxygen, which results in the formation of trichloroethylene peroxy radicals. These radicals then exert their effect on the lipid membrane of the endoplasmic reticulum, which causes lipid peroxidation. In the current study, the injection of CCl4 led to increased activities of SGOT, SGPT, and ALP in serum when compared to their respective control values. In response to the injection of CCl4, there was a decrease in the total amount of protein in the serum in comparison to the control. Abnormally high levels of SGPT, SGOT, and ALP following CCl4 treatment indicate an occurrence of hepatic injury, a condition responsible for the escape of cellular enzymes to the blood. Hepatic injury is responsible for the formation of abnormally high levels of SGPT, SGOT, and ALP. When the plasma membrane of the liver is disrupted, a number of enzymes that would normally be found in the cytosol were instead released into the circulation. The levels of SGOT, SGPT, and ALP all gradually decreased as the plant extract was used. The course of treatment using plant extract brought the abnormally elevated amount of overall bilirubin in the serum back down to normal, which is evidence of the extract's ability to protect the liver.

Damage to hepatocytes brought on by exposure to CCl4 is another factor contributing to a lower glycogen level in liver tissue. The fact that treatment in Prosopis juliflora extract resulted in protective effects strongly suggested that the extract may be capable to avoid as well as minimise the leakage of the marker enzymes through circulation, scenario the hepatocytes in order to accelerate rejuvenation of parenchyma cells, along with safeguard the integrity of plasma membranes and, as a result, restore these enzyme levels. Additionally, the extract might condition the hepatocytes to speed up regenerating of parenchyma cells.

CONCLUSION

The results of the scientific investigation on Prosopis juliflora reveal that this plant has significant potential as an antioxidant and hepatoprotective agent, and the results were comparable to those of the conventional medication silymarin. The extract demonstrated an existence of flavonoids, which are phenolic chemicals that have a beneficial hepatoprotective effect. According to the findings of the current research, a methanol extract of the aerial portions of Prosopis juliflora has the potential to function as an efficient hepatoprotective agent.

ACKNOWLEDGEMENT

We are thankful to Management of swami Vivekananda Institute of pharmaceutical sciences, Yadagirigutta, Vangapally for providing the all facilities for carried out this work.

Conflict of Interest

The authors declare that there is no conflict of interest.

Funding Support

The authors declare that they have no funding sup-

port for this study.

REFERENCES

[1] L J Su, G W Ding, Z L Yang, S B Zhang, Y X Yang, and C S Xu. Expression patterns and action analysis of genes associated with hepatitis virus infection during rat liver regeneration. *World J Gastroenterol*, 12(47):7626–7660, 2006.

[2] Y Lu, D Hu, S Ma, X Zhao, S Wang, and G Wei. Protective effect of wedelolactone against CCl4-induced acute liver injury in mice. *Int Immunopharmacol*, 34:44–52, 2016.

[3] B L Woolbright and H Jaeschke. The impact of sterile inflammation in acute liver injury. *J Clin Transl Res*, 3(1):170–88, 2017.

[4] Y Masuda. Learning toxicology from carbon tetrachloride-induced hepatotoxicity. *J Pharm Soc Jpn*, 126(10):885–99, 2006.

[5] D Singh, W C Cho, and G Upadhyay. Drug-Induced Liver Toxicity and Prevention by Herbal Antioxidants: An Overview. *Fron Physiol*, 6:363–363, 2016.

[6] O B Adewale, A O Adekeye, C O Akintayo, A Onikanni, and S Saheed. Carbon tetrachloride (CCl4)-induced hepatic damage in experimental Sprague Dawley rats: Antioxidant potential of Xylopia aethiopica. *Int J Pharmacol*, 3(2):118–141, 2014.

[7] P Zuzana, H Marie, D Helena, S Eva, Alena L Lenka, and B. Silymarin prevents acetaminophen-induced hepatotoxicity in mice. *PLoS One*, 13(1):191353–191353, 2018.

[8] X Wang, Q Wu, A Liu, A Anadón, J Rodríguez, and M Martínez-Larrañaga. Overdose-induced oxidative stress toxicity, metabolism and protective effects of various compounds in vivo and in vitro. *Drug Metab Rev*, 49(4):395–437, 2017.

[9] J Yang, Y Li, F Wang, and C Wu. Hepatoprotective effects of apple polyphenols on CCl4-induced acute liver damage in mice. *J Agric Food Chem*, 58(10):6525–6556, 2010.

[10] H H Fallah, S M Alavian, R Heshmat, M R Heydari, and K Abolmaali. The efficacy of Liv- 52 on liver cirrhotic patients: A randomized, double-blind, placebo-controlled first approach. *Phytomedicine*, 12(9):619–643, 2005.

[11] P Murugaian, V Ramamurthy, and N Karmegam. Hepatoprotective Activity of Wedelia calendulacea L. against Acute Hepatotoxicity in Rats. *Res J Agri Biol Sci*, 4(6):685–692, 2008.

[12] A K Shakya. Medicinal plants: Future source of new drugs. *I Journal of Herbal Med*, 4(4):59–64, 2016.

[13] S Jain, R Mohan, and R Rai. Ocimum sanctum as an Herbal Medicine: A Review. *Int J Oral Maxillofac Implants*, 1:1–12, 2015.

[14] J K Jose and R Kuttan. Hepatoprotective activity of Emblica officinalis and Chyavanaprash. *J Ethnopharmacol*, 72(1-2):135–175, 2000.

[15] M Boll, L W Weber, E Becker, and A Stampfl. Mechanism of carbon tetrachloride-induced hepatotoxicity. Hepatocellular damage by reactive carbon tetrachloride metabolites. *Z Naturforsch C J Biosci*, 56(7-8):649–59, 2001.

[16] S Sahreen, M R Khan, and R A Khan. Hepatoprotective effects of methanol extract of Carissa opaca leaves on CCl4-induced damage in rat. *BMC Complement Altern Med*, 11(1):48–48, 2011.

[17] K Vakati, H Rahman, M C Eswaraiah, and A M Dutta. Evaluation of hepatoprotective activity of ethanolic extract of Aquilaria agallocha leaves (EEAA) against CCl4 induced hepatic damage in rat. *Scholars J App Med Sci*, 1(1):9–12, 2013.

[18] B Y Yang, X Y Zhang, S W Guan, and Z C Hua. Protective effect of procyanidin B2 against CCl4-induced acute liver injury in mice. *Molecules*, 20(7):12250–65, 2015.

[19] J Zou, F Qi, L Ye, and S Yao. Protective Role of grape seed proanthocyanidins against CCl4 induced acute liver injury in mice. *Med Sci Monit*, 22:880–889, 2016.

[20] S Rasha, A Ismail, A Ashraf, A El-Megeid, and R Aly. Carbon tetrachloride-induced liver disease in rats: The potential effect of supplement oils with Vitamins E and C on the nutritional status. *Ger Med Sci*, 7:1612–3174, 2009.

[21] M Kanter, O Coskun, and M Budancamanak. Hepatoprotective effects of Nigella sativa L and Urtica dioica L on lipid peroxidation, antioxidant enzyme systems and liver enzymes in carbon tetrachloride-treated rats. *World J Gastroenterol*, 11(42):6684–6692, 2005.

[22] M Parola, G Leonarduzzi, F Biasi, M Albano, E Biocca, and G Poli. Vitamin E dietary supplementation protects against CCl4-induced chronic liver damage and cirrhosis. *Hepatol*, 16(4):1014–1035, 1992.

[23] S Noori, N Rehman, M Qureshi, and T Mahboob. Reduction of Carbon tetrachloride - induced

rat liver injury by coffee and green tea. *Pakistan J of Nutrition*, 8(4):452–460, 2009.

[24] K Jeong-Hyeon and L Kye-Taek. Glycoprotein Isolated from Ulmus davidiana NAKAI Protects against carbon tetrachloride-induced liver injury in the mouse. *J Pharmacol Sci*, 101(3):205–218, 2006.

[25] P Manna, M Sinha, and C S Parames. Aqueous extract of Terminalia arjuna prevents carbon tetrachloride induced hepatic and renal disorders. *BMC Compl and Alter Med*, 6(1):33–33, 2006.

[26] P Samudram, H Rajeshwari, R Vasuki, A Geetha, and M P Sathiya. Hepatoprotective activity of bi-herbal ethanolic extract on CCl4 induced hepatic damage in rats. *African J of Biochem Res*, 2:61–66, 2008.

[27] P K Jain, V Ravichandran, and R K Agrawal. Antioxidant and Free Radical Scavenging Properties of Traditionally Used Three Indian Medicinal Plants. *Curr Trends Biotechnol Pharm*, 2(4):538–585, 2008.

[28] J Hayes, L Condie, and J F Borzellec. Acute 14 day repeated dosing and 90 days subchronic toxicity studies of CCl4 in CD-1 mice. *Fund Appl Toxicol*, 7(3):454–63, 1986.

[29] H Kalantari, N Aghel, and M Bayati. Hepatoprotective Effect of Morus alba L. In Carbon Tetrachloride- Induced Hepatotoxicity in Mice. *Saudi Pharma J*, 17(1):90–94, 2009.

[30] O E Etim, E J Akpan, and I F Usoh. Hepatotoxicity of carbon tetrachloride: Protective effect of Gongronema latifolium. *Pak J Pharm Sci*, 21(3):268–74, 2008.

[31] N M John, E C Francis, and M M Harihara. Polyamine Protection against Chemically Induced Hepatotoxicity. *Int J Toxicol*, 19(6):391–400, 2000.

[32] A Islam, U K Mazumder, M Gupta, and S Ghosal. Synergistic Effects of Geraniin and Rutin in the Antioxidant Properties of Major Lignans in Phyllanthus amarus. *Pharmacol*, 3:1024–1060, 2008.

[33] R S Naidu, G S Kumar, S P Gawade, and K Devi. Protective Effect of LIV-O-G a polyherbal formulation on alcohol-CCl4 and paracetamol induced hepatotoxicity in rats. *Pharmacolonline*, 3:446–60, 2007.

[34] K D Tripathi. Essentials of Medical Pharmacology. *Jaypee Brothers Medical Publishers (P) Ltd: Delhi*, 2013.

[35] T Y Song and G C Yen. Protective effects of fermented filtrate from antrodia camphorate in submerged culture against CCl4 - induced hepatic toxicity in rats. *J Agric Food Chem*, 51(6):1571–1578, 2003.

[36] G T Brown and D E Kleiner. Histopathology of Nonalcoholic Fatty Liver Disease and Nonalcoholic Steatohepatitis. *Metabolism*, 65(8):1080–1086, 2016.

[37] L W B Weber and A Stampfl. Hepatotoxicity and mechanism of action of haloalkanes: Carbon tetrachloride as a toxicological model. *Crit Rev Toxicol*, 33(2):105–141, 2003.

[38] M N T Attia and M A Ali. Hepatoprtotective activity of allicin against Carbon tetrachloride induced hepatic injury in rats. *J Biol Sci*, 6(3):457–68, 2006.

[39] N Verma, A P Singh, G Amresh, P K Sahu, and C V Rao. Protective effect of ethyl acetate fraction of Rhododendron arboreum flowers against carbon tetrachloride-induced hepatotoxicity in experimental models. *Indian J Pharmacol*, 43(3):291–296, 2011.

[40] A Suvarchala, K Hemamalini, Girisha, and Uma Vasireddy. Hepatoprotective acitivity of Gymnosporia emarginate and Marsedenia volubilis against carbon tetrachloride induced hepatotoxicity in rats. *International journal of pharmacy and pharmaceutical sciences*, 5(4):726–729, 2013.

[41] K Hemamalini, , et al. Hepatoprotective acitivity of Tebebuia rosea and Solanum pubescens against carbon tetrachloride induced hepatotoxicity in rats. *International journal of biology, pharmacy and allied sciences*, 1:1135–1144, 2012.

[42] A Kanthasamy, S Subramanian, and S Govindasamy. Bactericidal and fungicidal effects of Prosopis juliflora Alkaloidal fraction. *Ind Drugs*, 26:390–394, 1989.

[43] K Saraswathi, Narun Nagendran, and S Karthigaichamy. Livelihood support from an invasive species Prosopis juliflora. *INT J CURR SCI*, pages 31–36, 2012.

[44] K Mibrahim, Nadir, Aali, and Ahmad. Mrasheed, Phytochemical analyses of Prosopis juliflora swartz dc. *Pak. J. Bot*, 45(6):2101–2104, 2013.

[45] N Deepa, S Chandra Nayaka, and K Girish. Raghavendra Synergistic effect of Prosopis juliflora extract and chemical fungicides against seed-borne toxigenic fungi. *International journal of advanced life sciences*, 6(4):312–317, 2013.

[46] M Theddeus, Ethel O Kiswii, Paul O Monda, Christine Okemo, Amos E Bii, and Alakonya. Efficacy of Selected Medicinal Plants from Eastern Kenya against Aspergillus flavus. *Journal of Plant Sciences*, 2(5):226–231, 2014.

[47] T Edilene, Mara Santos, A Lúcia, and Pereira. Camilla Flávia P.G. da Silva et al Antibacterial Activity of the Alkaloid Enriched Extract from Prosopis juliflora Pods and Its Influence on in Vitro Ruminal Digestion. *International Journal of Molecular Sciences*, (14):8496–8516, 2013.