DOI: https://doi.org/10.53350/pjmhs22161438

ORIGINAL ARTICLE

Evidence of Hepatoprotective Effect of Fluvastatin in a Rabbit Model

ATTYA ZAHEER[1], ASHIQ HUSSAIN[2], SHAZIA TUFAIL[3], FAIZA IRSHAD[4], FATIMA QAISER[5], SHAKEELA NAZIR[6], MUFASSAR NISHAT[7]

[1]*Assistant Professor of Anatomy. Rashid Latif Medical College Lahore.*
[2]*Associate Professor of Anatomy. Sahiwal Medical College Sahiwal*
[3]*Associate professor Anatomy Allama iqbal medical college Lahore*
[4]*Associate professor Anatomy M.Islam Medical & Dental College Gujranwala*
[5,6]*Professor of Anatomy Rashid Latif medical college Lahore*
[7]*Associate professor plastic surgery University medical & dental college. Faisalabad*

ABSTRACT

Fluvastatin's hepatoprotective impact is being investigated in the current study. It is possible that the oxidative stress pathway is implicated in PCM hepatotoxicity. The acute hepatotoxicity of PCM was investigated in this study.

Place of study: Rashid Latif Medical College Lahore

Materials and Methods: The rabbits were divided into five groups of ten each. The saline group received (the vehicle). Fluvastatin (20 mg/kg), PCM (600 mg/kg), and PCM + 10 mg Fluvastatin (600 mg PCM + 20 mg Fluvastatin) were administered to the animals. The livers from all the animals were taken out and cleansed. 70% alcohol was used for washing extensively, after which the liver tissue was embedded in paraffin and then 5 mm thick paraffin slices were stained by aid of hematoxylin and eosin.

Results: Male rabbits were shown to be harmful to paracetamol ($P<0.05$). However, as compared to group 1, the levels of bilirubin, ALT, AST, and ALP were considerably higher ($P<0.0001$) (In the control group.) Fluvastatin (10mg/kg and 20mg/kg) combined with paracetamol resulted in significant reductions in ALP, ALT, and bilirubin levels.Slices of liver were analyzed histologically.

Conclusion: Fluvastatin reduced the severity of all of these side effects, although it did not completely eliminate them**.**

Keywords: Fluvastatin, Hepatotoxicity, Paracetamol, Hepato protection, histopathology.

INTRODUCTION

A vital foreign material processing organ, the liver is placed in the transitional zone between resorption and circulation. The liver is the most critical organ for foreign chemical detoxification, but it is also the most dangerous because of the amount of toxins it produces. In other words, liver damage causes alterations in a number of critical metabolic processes. Paracetamol is a commonly used analgesic and antipyretic. Paracetamol, in contrast to phenacetin, does not appear to be carcinogenic. Paracetamol, in contrast to aspirin, is well tolerated and readily available without a doctor's prescription. It is used to treat symptoms such as fever, headaches, and pains. In addition to paracetamol, NSAIDs or opioid analgesics are used to treat severe pain. In the Western world, the most prevalent cause of liver failure is paracetamol intoxication, which increases the likelihood of consuming alcoholic beverages. The Toxic Dose of this substance varies greatly. Poisoning in adults occurs when a single dosage of 10 grams (200 mg/kg) or more is administered incorrectly while in case of children it occurs when poisoning levels raise greater than 200 mg/kg. An overdose of paracetamol in a kid can be lethal. However, it takes around 6-8g of paracetamol per adult to for liver necrosis to develop and he rarely inquires as to how much paracetamol a youngster need. However, persistent paracetamol usage is responsible for the majority of kid poisonings . One gram of paracetamol taken four times daily produces an increase in liver function test results in one-third of those who take it; however, it is unclear whether this results in liver failure. Acute liver damage is widespread due to the high tolerance of the drug and the fact that it is available over-the-counter. Because of the saturation of the sulphate and glucuronide pathways, more paracetamol is transferred to the cytochrome P450 system, where it is converted to NAPQI. Glutathione hepatocellular stores are reduced as a result of increased demand outstripping regeneration. The buildup of NAPQI in the liver results in severe hepatocyte damage and death, ultimately resulting in acute hepatic necrosis. It is possible to get hepatotoxicity when glutathione levels fall below 65% of normal. Angiotensin-converting enzyme inhibitors (ACEIs), resveratrol, melatonin, quercetin, and other flavonoids are among the drugs used to treat hepatic paracetol toxicity, among other things. In a recent study, it was shown that certain plants may decrease or detoxify poisons, as well as protect the liver from medications and chemicals. It works by inhibiting 3-hydroxy-3methylglutaryl coenzyme A reductase, which is the rate-limiting enzyme in cholesterol production. In spite of the fact that statins can decrease triglycerides in the liver and alleviate steatosis, their therapeutic value in individuals with NASH is still up for discussion. To determine fluvastatin's efficacy in preventing paracetamol-induced hepatotoxicity in rabbits, histological changes and variations in liver enzymes were observed in the animals.

MATERIALS AND METHODS

Male rabbits weighing 200-250 g were used in this investigation, which was conducted on 10-week-old rabbits. The animals were purchased at a local market in the area. They were kept in a well-ventilated animal housing at a temperature of 24°C with 12 hours of light and darkness every day, and they were fed twice a day.

Drugs: Fluvastatin and Paracetamol drug were dissolved in normal saline before use.

Experimental design: As a control group, the first group received normal saline (the vehicle) 0.5ml/animal/day i.p. for four weeks at a dose of 0.5ml/animal/day i.p. During the second phase (induction), the participants were given PCM 600mg/kg orally three times per week for eight weeks. During the eighth week of the study, the third group received fluvastatin 20mg/kg daily via gavage, whereas the fourth group received fluvastatin 10mg/kg daily with 600mg/kg PCM. A total of 20 mg/kg daily fluvastatin and 600 mg/kg PCM were given to the animals in Group 5. At the conclusion of the trial, the subjects were anaesthetized with diethyl ether.

Biochemical Examination: The animals were killed by dislocating their necks after a brief period of moderate anesthetic had been administered. Blood samples were obtained using a cardiac puncture. This is the point at which the blood samples acquired coagulate. We centrifuged the sera at 3500 rpm for 10 minutes, which yielded a clear serum. Then, using a spectrophotometric autoanalyzer, blood samples were examined for AST, ALT, ALP, total bilirubin, and albumin activity, as well as other markers of liver function.

Histological Examination: The livers were removed and cleansed. After extensive washing in 70% alcohol, the liver tissue was processed for paraffin embedding and 5 mm thick paraffin slices were stained with hematoxylin and eosin.

RESULTS

As demonstrated in Table 1, the effects of fluvastatin on albumin, bilirubin, and liver enzymes (ALT, AST, and ALP) in APAP-treated rabbits are proven to be beneficial. When exposed to paracetamol, serum albumin (milligrammes per deciliter) decreases ($P<0.05$). Administering fluvastatin alone to healthy rabbits (group 3) showed little or no impact on total bilirubin, albumin, ALT, AST, and ALP levels, however giving fluvastatin with other drugs had a significant effect. ALT, AST, and ALP all dropped dramatically in serum total bilirubin, ALT, AST, and ALP ($P<0.001$). Groups 4 and 5 received oral fluvastatin in combination with paracetamol, which resulted in increased total albumin levels in the blood (P0.05) when compared to rabbits given paracetamol. However, it did not correct the situation. Control group participants' liver tissue was found to be normal (Figure.1). Death and vacuolization of liver cells are normal processes.

Table 1: Fluvastatin low and high dosage effects on Paracetol induced hepatotoxicity in rabbits

Parameters	Group 1	Group 2	Group 3	Group 4	Group 5
Bilirubin mg/dl	0.61±	1.78±	0.54±	1.68±	1.14±
Albumin g/dl	4.61±	3.00±	4.51±	2.99±	3.47±
ALT U/L	57.49±	161.67 ±	56.51±	160.82 ±	109.98 ±
AST U/L	74.08±	228.57 ±	74.97±	233.01 ±	140.01 ±
ALP U/L	140.06 ±	251.27 ±	138.91 ±	247.06 ±	10.14±

The liver cells of rabbits given paracetamol showed signs of degeneration, desquamation, and necrosis (Figure.2). The hepatic peripheral zone is strewn with waste. It also contains dilated central veins and occluded hepatic blood arteries, among other things (Figure 3). Two dosages of fluvastatin (10mg/kg and 20mg/kg B.w.) delivered at the same time significantly decreased paracetamol-induced liver damage in rabbits. The hepatic sinusoids had a more pleasing appearance than the sinusoids in the paracetamol group.

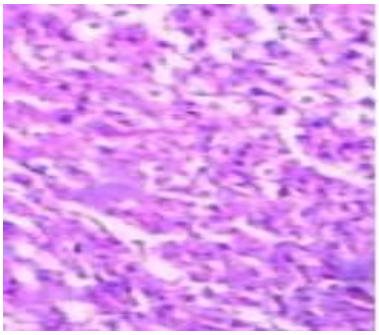

Figure 1: Slices of the control group exhibited typical cellular architecture with distinct liver cells, sinusoidal gaps, and central vein.

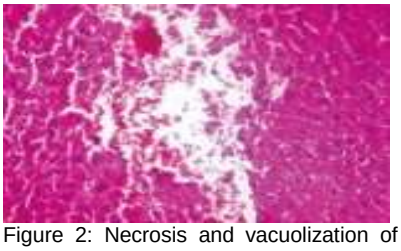

Figure 2: Necrosis and vacuolization of normal hepatic cells in paracetamol-treated rabbit livers. Hepatic cell degeneration and desquamation noted.

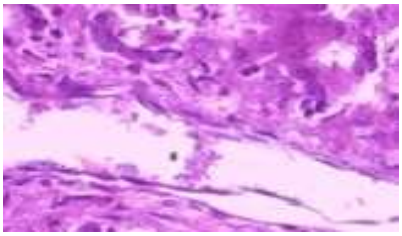

Figure 3: PCM-treated rabbit liver section The hepatic central vein has cellular debris. Also, hepatic central vein dilatation , hepatic cell degeneration

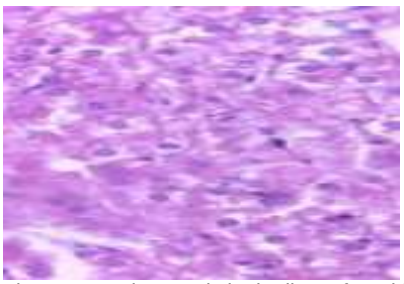

Figure 4: Angiogenesis in the liver of a rabbit treated with PCM and Fluvastatin Cellular desquamation was limited, and the hepatic sinusoids had better morphology than the paracetamol group.

DISCUSSION

The acute liver necrosis model caused by paracetamol is a reliable model . Acute hepatocellular damage, on the other hand, causes an increase in plasma transaminases, particularly ALT. Intrahepatic cholestasis, on the other hand, is characterized by high levels of enzyme and plasma ALP activity. It is a widely used analgesic and antipyretic medication. The medication doses were found to be safe. Conjugates of sulphate and glucuronide are formed. The induction of liver injury by paracetamol is widely used to evaluate the efficiency of hepato protective drugs . The use of melatonin, vitamin E, and N-acetylcysteine to reduce paracetamol-induced liver and kidney damage has been shown to be effective. It is responsible for organizing the chemical environment of the organism. The dangers of toxic agent-induced liver damage are well known. D-galactosamine and acetaminophen, both of which are toxic to the liver, induce ammonia to accumulate in the bloodstream. Paracetamol is an analgesic and antipyretic that is hepatotoxic. It has been used to examine the effects of liver-protective agents. Paracetamol causes liver damage by covalently joining its poisonous metabolite (n-acetylP-benzoquineune amine) with the sulfhydryl group of protein generated in cell necrosis and lipid peroxidation, which causes cell necrosis and lipid peroxidation to occur. During supper, turbulent plasma leakage increases the levels of serum enzymes. We wanted to see if fluvastatin may help to decrease or perhaps eliminate the hepatototoxic effects of paracetamol. Hepatic serum enzymes and bilirubin levels were significantly elevated in group II, which received just a paracetamol treatment (AST, ALT, and ALP). The liver function in group II (the positive control) was found to be diminished (Table1). They corroborate prior findings from rat studies on paracetamol . When administered orally to group II, fluvstatin at doses of 10 and 20 mg/kg p.o. reduced the rise in blood enzyme markers caused by paracetamol. It serves to protect the plasma membrane of the hepatocyte . Paxil caused liver damage, which resulted in a decrease in albumin. Transaminases are considered to be the gold standard for assessing liver dysfunction. Statins, on the other hand, appear to inhibit the generation of proinflammatory cytokines by microglia, astrocytes, and mononuclear cells. Statins have been shown to protect the brain and lungs. They were minimizing neutrophil inflow, which may have helped to reduce macrophage influx, lymphocyte activation, and cytokine release, all of which were detrimental. Statins inhibit the expression of interleukin-6, interleukin-8, and GM-CSF in human cells. Statins are anti-oxidants that prevent the cell death process known as apoptosis. Statins have the potential to modify these pathways by activating intracellular prenylation and GTP-binding proteins. Statins also help to protect the brain from ischemia by increasing the production of ENOS (eNOS). In mice, these medicines decreased the extent of ischemia lesions as well as neurologic deficits. Statins raised eNOS and tPA, but not PAI-1, as previously reported. It is possible that fluvastatin's hepatoprotective effects are due to its anti-inflammatory qualities.

CONCLUSION

The findings of this study suggest that statins (fluvastatin) can diminish PCM-induced hepatotoxicity in rabbits, and that they may have a comparable impact in humans. Fluvatstin's antioxidant properties are likely to have contributed. However,. Fluvastatin is an anti-inflammatory medication with antioxidant effects. Given its antioxidant characteristics and lack of liver damage, fluvastatin may be able to treat PCM-induced hepatotoxicity in some cases.

REFERENCES

1 Tobert JA. Lovastatin and beyond: the history of the HMG-CoA reductase inhibitors. Nat Rev Drug Discov 2003;2:517–26.
2 Kim SN, Seo JY, Jung da W, Lee MY, Jung YS, Kim YC. Induction of hepatic CYP2E1 by a subtoxic dose of acetaminophen in rats: increase in dichloromethane metabolism and carboxy hemoglobin elevation. Drug Metab Dispos 2007;35:1754–8.
3 Rowden AK, Norvell J, Eldridge DL, Kirk MA. Updates on acetaminophen toxicity. Med Clin North Am 2005;89:1145–59.
4 Gumbrevicius G, Sveikata A, Sveikatienė R, Stankevičius E. Paracetamol and simvastatin: a potential interaction resulting in hepatotoxicity. Medicina (Kaunas) 2012;48:379–81.
5 Kasmi I, Sallabanda S, Kasmi G. Fulminate hepatic failure in a 5 year old female after inappropriate acetaminophen treatment. Open Access Maced J Med Sci 2015;15;3:443–6.
6 Karthivashan G, Kura AU, Arulselvan P, Md Isa N, Fakurazi S. The modulatory effect of Moringa oleifera leaf extract on endogenous antioxidant systems and inflammatory markers in an acetaminophen-induced nephrotoxic mice model. Peer J 2016;7:e2127.
7 Eguia L, Materson BJ. Acetaminophen related acute renal failure without fulminant liver failure. Pharmacotheraphy 1997;17:363–70.
8 Bhattacharya P, Chaturvedi S. Dyslipidemia management. Continium (MinneapMinn) 2011;17:1242–54.
9 Vasudevan AR, Hamirani YS, Jones PH. Safety of statins: effects on muscle and the liver. Cleve Clin J Med 2005;72:990–3, 996–1001.
10 Bhardwaj SS, Chalasani N. Lipid-lowering agents that cause drug-induced hepatotoxicity. Clin Liv Dis 2007;11: 597–613, vii.
11 Rangnekar AS, Fontana RJ. An update on drug induced liver injury. Minerva Gastroenterol Dietol 2011;57:213–29.
12 Dormuth CR, Hemmelgarn BR, Paterson JM, James MT, Teare GF, Raymond CB, et al. Use of high potency statins and rates of admission for acute kidney injury: multicenter, retrospective observational analysis of administrative databases. Br Med J 2013;346:f880.
13 Raje RR, Bhattacharya S. Lovastatin-acetaminophen subchronic toxicity in mice. Res Commun Chem Pharmacol 1990;69:373–6.
14 Aguirre L, Hijona E, Macarulla MT, Gracia A, Larrechi I, Bujanda L, et al. Several statins increase body and liver fat accumulation in a model of metabolic syndrome. J Physiol Pharmacol 2013;64:281–8.
15 Demirci B, McKeown PP, Bayraktutan U. Blockade of angiotensin II provides additional benefits in hypertension-and ageing-related cardiac and vascular dysfunctions beyond its blood pressure-lowering effects. J Hypertens 2005;23:2219–27.
16 Jukema JW, Cannon CP, de Craen AJ, Westendorp RG, Trompet S. The controversies of statin therapy: weighing the evidence. J Am Coll Cardiol 2012;60:875–81.
17 Nasri H, Hasanpour Z, Nematbakhsh M, Ahmadi A, Kopaci MR. The Effect of the various doses of atorvastatin on renal tubular cells, an experimental study. J Nephropathol 2016;5:111–5.
18 Caliskan D, Koca T, Kumbul Doguc D, Ozgocmen M, Akcam M. The protective effect of pomegranate juice in paracetamoll nduced acute hepatotoxicity in rats. Türk Pediyatri Arşivi 2016;51:72–8.
19 Uysal HB, Dagli B, Yılmaz M, Kahyaoglu F, Gokcimen A, Omurlu IK, et al. Biochemical and histological effects of thiamine pyrophosphate against acetaminophen-induced hepatotoxicity. Basic Clin Pharmacol Toxicol 2016;118:70–6.
20 Uysal HB, Dagli B, Yılmaz M, Kahyaoglu F, Gökcimen A, Omurlu IK, et al. Protective effects of dexpanthenol against acetaminophen-induced hepatorenal damage. Biomed Res 2017;28:740–9.