Effects of Grape Seed Oil on Liver of Rats

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ABSTRACT

OBJECTIVES: *To study the effect of oral administration of grape seed oil (GSO) against carbontetrachloride (CCl4)-induced hepatotoxicity in rat Liver damage was induced in male Wistar rats (150-200 g) by administering CCl4 (0.5 ml/kg, i.p.) once per day for 7 days and the extent of damage was studied by assessing biochemical parameters such as alanine aminotransferase (ALT), aspartate aminotransferase (AST), and alkaline phosphatase (ALP) in serum and concentrations of glutathione (GSH), superoxide dismutase (SOD), and total protein (TP) in liver. The effect of co-administration of GSO (3.7 g/kg, orally) on the above parameters was further investigated and compared with a vitamin E (100 mg/kg, orally) treated group.*
RESULTS: *Oral administration of GSO (3.7 g/kg, body weight orally) for 7 days resulted in a significant reduction in serum AST, ALT, and ALP levels and significant improvement in glutathione, SOD, and TP, when compared with CCl4 damaged rats. The antioxidant effect of GSO at 3.7 g/kg for 7 days was found to be comparable with vitamin E (100 mg/kg, orally) in CCl4-treated rats.*
CONCLUSION: *The GSO has protected the liver from CCl4 damage. Probable mechanism of action may be due to the protection against oxidative damage produced by CCl4.*
Key Words: *Grape seed oil and liver function*

INTRODUCTION

Grape seed oil (GSO) is obtained from grape seeds after the wine pressing in Italy and France. The GSO contains 75% linoleic acid, 15% oleic acid, 6% palmitic acid, 3% stearic acid, and 1% linolenic acid.[1]

Studies revealed the beneficial HDL effect of GSO and research shows that subjects were instructed to use up to 45 ml of GSO in their daily diet as a substitute for their usual oil and within 2 weeks there was 13-14% increase in HDL level.[2] The GSO has a very high level of antioxidant vitamin E (60-120 mg/100 g), which makes the oil very stable. The antioxidant property is claimed to be the mechanism of hepatoprotective activity.[3] The GSO exhibits a variety of interesting properties such as reducing platelet aggregation, prevents hypertension caused by sodium excess, normalizes lesions occurring from obesity and diabetes.[4]

Among the various mechanisms involved in the hepatotoxic effect of carbontetrachloride (CCl4), one is oxidative damage through free-radical generation[5] and antioxidant property is claimed to be one of the mechanisms of hepatoprotective effect of indigenous drugs.[6] The GSO has antioxidant properties.[2],[3] Hence, the objective of the study was to evaluate

the effect of GSO on CCl4-induced hepatotoxicity.

MATERIALS & METHODS

Drugs and chemicals

The GSO is a kind gift from LoDuca Bros Inc., Milwaukee, USA.

Carbontetrachloride (CCl4) was obtained from Sigma.

Experimental animals

Male Wistar albino rats (150-200g) were used. The animals were acclimatized to laboratory conditions for 5 days prior to the experiments and had access to food and water ad libitum.

Selection of dose of GSO

The dose for the hepatoprotective studies was adjusted based on the observation during the toxicity studies. The GSO at a dose of 3.7 g/kg (4 ml/kg) was administered orally to study the hepatoprotective activity.

Hepatoprotective studies

Animals were divided into five groups, consisting of six animals each. ***Group I:*** served as control which recieved orally for 7 days. Group II received GSO (3.7 g/kg, orally) for 7 days. Group III received CCl4 0.5 ml/kg, i.p. for 7 days.[9],[10],[11] Group IV received CCl4 0.5 ml/kg, i.p. and GSO (3.7 g/kg, orally) simultaneously for 7 days. Group V received CC14 0.5 ml/kg, i.p. and vitamin E (100 mg/kg, orally)[12] simultaneously for 7 days. After 7 days of treatment, the rats were kept overnight fasting and killed. At the end of the treatment, blood samples were collected by direct cardiac puncture under ether anaesthesia and the serum was used for the assay of marker enzymes mainly, aspartate aminotransferase (AST), alanine aminotransferase (ALT), and alkaline phosphatase (ALP).[13],[14] The enzyme levels were assayed using the standard kits from laboratories. The results were expressed as units/liter (U/l). Liver samples were dissected out and washed immediately with ice-cold saline to remove as much blood as possible. Liver homogenates (5% w/v) were prepared in cold 50 mM potassium phosphate buffer (pH 7.4) using homogenizer. The unbroken cells and cell debris were removed by centrifugation at 1000 rpm for 10 min. The supernatant was used for the estimation of GSH,[15] superoxide dismutase (SOD),[16] and total protein (TP)[17] levels.

Statistical analysis

Values were represented as mean + SE. Data were analyzed using one-way analysis of variance (ANOVA) and group means were compared using Duncan′s multiple range test. P values <0.05 were considered significant.[18]

RESULTS

In acute toxicity study, no signs, and symptoms of toxicity and mortality were observed. There was a significant ($P<0.05$) increase in the serum hepatic enzyme levels after CCl4 treatment, which was prevented with GSO. The GSO when administered alone did not alter the enzyme levels when compared to the control values. There was a significant ($P<0.05$) rise in GSH, SOD contents of liver after treatment with GSO. There was a significant decrease in TP level after CCl4 treatment, which was prevented with GSO [Table - 1] and [Table - 2].

Table 1: Effect of GSO on serum ALT, AST, and ALP in rats after 7 days treatment

Groups	ALT (U/l)	AST (U/l)	ALP (U/l)
Control	51.3±4.3*	266.3±66.7*	1006.3±126.5*
GSO (3.7g/kg)	59.9±5.7*	255.6±53.0*	1097.0±4.5*
CCl4(0.5ml/kg,I,p,)	231.6±12.4**	695.3±116.6**	1479.4±311.6**
CCl4+GSO (3.7g/Kg)	73.3±7.3***	292.8±93.0***	1124.3±21.6***
CCl4+VE(100mg/kg)	74.3±7.3*,***	291.3±13.4*,***	1146.8±114.0*,***
F	58.3	117.9	245.9
d.f.	4,25	4,25	4,25
P	<0.05	<0.05	<0.05

Values are mean ± SE; n=6 in each group. Values with different superscripts (*,**and***) differ significantly from each other at p<0.05 (Duncan ‘s multiple range test)

Table 2: Effect of GSO on liver GSH, SOD, and TP in rats after 7 days treatment

Groups	GSH Mg/100g Tissue	SOD Units/mg Protein	Total Mg/mi Protein
Control	39.8±11.8*	7.70±0.25*	17.15±1.34*
GSO (3.7g/kg)	37.0±2.2*	7.81±0.22*	16.42±3.5*
CCl2 (0.5m/kg i.p)	18.8±1.8**	4.51±0.09**	7.70±0.70**
CCl4 + GSO (3.7 G/KG)	35.6±4.5***	5.04±0.34***	15.30±0.16***
CCl4+ VE (100 mg/kg)	37.3±4.1*	7.48±0.24*	17.16±5.60*
F	22.4	4.67	3.47
d.f	4.25	4.25	4.25
P	<0.05	<0.05	<0.05

Values are mean±SE; n=6 in each group. Values with different superscripts (*,**and***) differ significantly from each other at p<0.05 (Duncan s multiple range test)

DISCUSSION

The CCl4 is one of the most commonly used hepatotoxins in the experimental study of liver diseases.[19] The lipid peroxidative degradation of biomembranes is one of the principal causes of hepatotoxicity of CCl4.[20] This is evidenced by an elevation in the serum maker enzymes, namely AST, ALT, and ALP. The GSO has significantly reduced these liver enzyme levels. Further, GSO has increased the level of TPs, which indicates hepatoprotective activity.

Stimulation of protein synthesis accelerates the regeneration process and the production of liver cells. In our study, elevation in the levels of lipid peroxidation in CCl4-treated animals was observed. Maheswory,[21] found that the increase in MDA and hydroperoxide levels in liver with CCl4 suggests that enhanced lipid peroxidation leads to tissue damage and failure of

antioxidant defence mechanisms. Treatment with GSO significantly prevented these changes. Hence, the mechanism of hepatoprotection of GSO may be due to its antioxidant effect. Since GSO has significantly increased the glutathione, and SOD contents of the liver, it may also be useful in hepatotoxicity induced by other agents. The antioxidant enzyme levels of the CCl4-treated group were decreased whereas that of GSO-treated group is almost similar to that of the control and vitamin-E-treated groups.

Pretreatment with GSO exhibited protection, which confirmed the results of biochemical studies. These results of our study indicate that simultaneous treatment with GSO protects the liver against CCl4-induced hepatotoxicity.

The GSO offers vast possibilities in the treatment of various liver disorders. This may be due to the high level of antioxidant vitamin E, which was claimed to be the mechanism of hepatoprotection. Further studies on any other models and extensive clinical trials are needed to confirm these results.

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تاثير زيت بذره العنب على كبد الجرذان البيضاء

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كليه البنات للاداب والعلوم والتربيه– جامعه عين شمس

تم دراسه تاثير زيت بذره العنب على رباعى كلوريد الكربون المسبب لتدمير خلايا الكبد فى الجرذان البيضاء. تم اعطاء المجموعه الاولى زيت بذره العنب للجرذان لمده ٧ ايام وللمجموعه الثانيه تم اعطاء رباعى كلوريد الكربون لمده ٧ ايام اما المجموعه الثالثه فاعطيت كل من زيت بذره العنب ورباعى كلوريد الكربون اما المجموعه الرابعه فاعطيت رباعى كلوريد الكربون بالاضافه الى فيتامين هـ.

ووجد ان هناك زياده معنويه فى كل من الفوسفاتيز القلوى والترانس امينيز بالاضافه للسوبر اوكسيد ديمستيولاز بعد اعطاء رباعى كلوريد الكربون اما بعد اعطاء زيت بذره العنب وجد تحسن معنوى فى الفوسفاتيز القلوى والترانس امينيز و السوبراوكسيد ديمستيولاز.

ووجد ان هناك نقصا فى كل من الجلوتاثيون والبروتين الكلى بعد اعطاء رابع كلوريد الكربون منفردا اما اذا اتحد مع زيت بذره العنب وجد ان هناك تحسن معنوى فى كل من الجلوتاثيون والبروتين الكلى.