RESEARCH ARTICLE

EFFECT OF AQUEOUS CRUDE EXTRACT OF ALOE VACILLANS LEAVES ON INDUCED HEPATIC DAMAGE IN MALE RABBITS

Mohammed Ali Qassem

Faculty of Dentistry, University of Aden, Aden, Yemen

Corresponding author: Mohammed Ali Qassem; E-mail: aliunidi3moh1Ali3@gmail.com

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Abstract

This study was designed to evaluate the effects of aqueous extract of Aloe vacillans leaves juice on carbon tetrachloride (CCl₄)- induced hepatotoxicity in rabbits. Hepatotoxicity was induced in rabbits by intraperitoneal injection of (CCl₄) at dose 1 ml/kg on day 7 and 8. The aqueous crude extract of Aloe vacillans leaves was administrated at dose 100 mg, 300 mg and 500 mg/kg of body weight pass orally (p.o) daily for 8 days. The hepatotoxicity and its prevention were assessed by serum parameters like alanine aminotransferase (ALT), aspartate aminotransferase (AST), bilirubin (Bil) and total protein (T.P).

In CCl₄ treated rabbits, a significant, increasing the ALT, AST, bilirubin and decreasing the Total protein levels were shown (p<0.05), due to liver damage, when compared with the normal group. Treatment with the aqueous extract of Aloe vacillans could significantly decrease the (ALT), (ALT) and bilirubin, increased T.P in serum at p< 0.05 when compared with CCl₄ –treated group.

The data concluded that oral administration of aqueous extract of the leaves of Aloe vacillans significantly decreases the intensity of hepatic damage induced by CCl₄ in rabbits.

Keywords: Carbon tetrachloride, Alanine aminotransferase (ALT), Aspartate aminotransferase (AST), Total Protein (T.P), Bilirubin (Bil), Hepatic damage, Rabbits.

Introduction:

The largest organ in the human body, the liver, play a very important role in the foreign compounds entering the body. The exposure to the foreign may be through consumption of alien contaminated food, from exposure to a chemical substance in the occupational environment or through synthetic drugs consumed for various from exposure to a chemical substance in the occupational environment or through synthetic drugs consumed for the pathological condition. These compounds have many toxic manifestations on the human liver.[1]. In humans, hepatitis or liver injury is also caused by viruses, chemicals, alcohol and autoimmune diseases. [2, 3, 4]

Carbon tetrachloride (CCl₄) is often used to induce oxidation stress-related. The reactive metabolites such as trichloromethyl (CCl₃) and trichloromethyl peroxy (CCl₃OO) radical emanated from CCl₄ initiate per oxidation of membrane unsaturated fatty acid. This lipid per oxidation of membrane seriously impairs its function and produces liver injury. And the rise in serum level of aspartate transaminase (AST), alanine transaminase (ALT) has been attributed to the damaged structural integrity of the liver because they are cytoplasmic in location and released into circulation after cellular damage. [5-6]

Liver diseases remain one of the serious health problems and medicinal plants and herbs have been in use for treating these in the world. Aloe plants grow in warm tropical areas and cannot survive freezing temperature. The plant is native to southeastern Africa and subsequently introduced into northern African, The Arabian Peninsula, China, the Mediterranean countries and west India. It is commercially cultivated in Aruba, Bonaire, Haiti, South Africa, the United State of America and Venezuela [7]. The various species of aloe have the same effective phenolic compounds (anthraquinones) such as aloe-modin, aloesin, barbaloin, aloenin, and isobarbaloin [8-9].

A number of investigators have previously demonstrated that antioxidants prevent CCl₄ toxicity particularly hepatotoxicity, by inhibiting lipid peroxidation [10], the leaves juice of Aloe plant is used in eyes diseases and
enlargement of the spleen and liver [11]. Anthraquinones may act as antioxidants and radical scavenger, reactive oxygen species and free-radical mediated reactions are involved in the inflammatory response and can contribute to liver necrosis [12]. Thus, the current study has been aimed to evaluate the effects of aqueous extract of Aloe vacillans leaves juice on CCl₄-induced hepatotoxicity in rabbits.

2. Material and methods:

2.1- Plant material and Extraction:
Aloe vacillans leaves were collected from the lauder region, Abyen Governorate, Yemen. The leaves of the plant were washed with water, dried under shade and powdered to fine grade by using a laboratory scale mill, the powdered extracted with distilled water (250 g/4 litres) for 18 h with concomitant shaking. The filtrate was evaporated rotary to yield a brown powder, which was administered orally according to a bodyweight of animals. [13]

2.2- Chemicals:
CCl₄ were purchased from sigma chemical Co.
All other chemicals and reagent were purchased from SPINREACT, S.A.U

2.3- Animals:
Male rabbits weighing 1000-1200 gm were purchased from and used in these experiments. The animals were housed at room temperature (28±2C) in standard cages with standard pellet food and kept under a controlled environment following relative humidity (60±5%) with a 12h light/dark cycle.

2.4- Behavioral and Toxic Effects:
The acute toxicity study was evaluated in rabbits according to the method of [18]. Five groups of 6 animals were administrated with 125,250, 500, 1000, 2000 mg/kg of the Aloe vacillans extracts orally, while one group with the same number of rabbits kept as a control group.
The animals were observed continuously for 72 h, and then after every 24 h for 15 days for any gross behavioral change, symptoms of toxicity or mortality. [14]

2.5- CCl₄- induced hepatic damage in rabbits:
About 30 male rabbits were divided into five groups of ten animals each
Group I. Serve as normal control and received 1 ml distilled water
Group II. Rabbits received an equal mixture of carbon tetrachloride and paraffin (1:1) 1 ml/kg b.wt.i.p on days 7 and 8 and served as CCl₄ control
Group III, IV and V were administered with aloe extract at 100,300, and 500 mg/kg b.wt.o.p respectively for 8 days and on the day 7 and 8 received mixture of carbon tetrachloride and paraffin 1ml/kg b.wt, i.p.

2.6- Preparation of serum from blood:
After 24 hours of the second dose of CCl₄ the rabbits were sacrificed, and the blood samples were collected by tubes from each animal. The blood allowed clotting for 30 min at room temperature. Serum was separated by centrifugation at 3000 rpm for 10 minutes [15], and analyzed for various biochemical parameters including serum aspartate aminotransferase (AST) serum alanine aminotransferase (ALT), total protein (T.P) and Total Bilirubin (T.B) by using (UV) spectrophotometer (model screen master plus RM4040

2.7- Statistics:
All the values are expressed as a mean± SEM. The data are evaluated using one way (ANOVA) test to determine the significance of the difference between the normal group and the CCl₄ treated group only. Differences between the CCl₄-treated group alone and the CCl₄ groups treated with extract at three different doses. (100 mg/kg, 300 mg/kg and 500mg/kg) were compared for significance differences below (p<0.05) are considered as significant.

3. Results:
The results of CCl₄- induced hepatotoxicity are shown in figures. 1, 2 and 3
The hepatic injury induced by CCl₄ in rabbits caused significantly elevated serum levels of ALT, AST and bilirubin, a total protein that whereas a significant decrease in the level of the total protein in the CCl₄ alone group compared with the normal group.
The rabbits treated with aqueous extract of aloe at three dose levels (100,300 and 500 mg/kg b.w, o.p) showed a significant reduction in marker enzymes ALT, AST and serum bilirubin. Whereas increase in the level of total protein was showed in these groups in comparison to that of the CCl₄ alone group. Figures 1, 2 and 3
**Fig. 1.** Values are represented as mean ±SEM (n=10) Anova test (p<0.05) is used. CCl₄ alone significantly different from normal control.

*, **, significantly different from CCl₄ treatment only.

**Fig. 2.** Values are represented as mean ±SEM (n=10) Anova test (p<0.05) is used. CCl₄ alone significantly different from normal control.

*, **, significantly different from CCl₄ treatment only.
4. Discussion:

In the toxicity study, the rabbits when fed with *Aloe vacillans* up to 2000 mg/kg, p.o exhibited no mortality or any sign of gross behavioral changes when observed for 72 h and then after every 24 h for 15 days. The LD50 was greater than 2000 mg/kg p.o., it may be considered relatively safe.

In this study, rabbit's treatment with a dose of CCl₄ developed significant hepatic damage, which was observed from a substantial increase in activities of serum ALT, AST and bilirubin, decrease total protein significantly p< 0.05 compared with the normal group.

CCl₄ is biotransformed by the cytochrome p450 system in the endoplasmic reticulum to produced trichloromethyl free radical. trichloromethyl free radical then combined with cellular lipids and proteins in the presence of oxygen to form trichloromethyl peroxy radical, which may attack lipids on the membrane of endoplasmic reticulum faster than trichloromethyl radical. Thus, trichloromethyl peroxy radical lead to elicit lipids peroxidation, and finally results in cells death. [16]

Increase of levels enzymes ALT, AST metabolic activation, reduction of protein synthesis and loss of glucose-6-phosphatase activation indicative of cellular leakage and loss of functional integrity of the cell membranes in the liver. [17-18]

Reduction in the levels of the ALT, AST, and bilirubin, increase in the level of total proteins by plant aqueous extract groups at three different doses (100, 300, 500 mg/kg b.w.o.p) significantly p<0.05 compared to alone CCl₄ group.

The *Aloe vacillans* aqueous extract at the highest dose (500 mg/kg) significantly lowered the levels of ALT, AST, and bilirubin, increased the T.P when compared with a lower dose (100,300 mg/kg) at p<0.05 and this finding in current results were agreement with according to results of [14-19].

The results [20-23] indicate that stabilization of plasma membranes as well as repair of hepatic tissue damage caused by CCl₄. This effect view that serum levels of transaminase return to normal with the healing of hepatic parenchyma and the regeneration of hepatocytes. [24]. The *Aloe vacillans* extract might contain phenolic compounds such as aloe-emodin, emodin, barbaloin, antioxidant activity as indicated by protection against increased lipid peroxidation.

5. Conclusion:

We conclude that treatment with aqueous extract of *Aloe vacillans* reduced the degree of hepato-cellular injury as evidenced by improved biochemical parameters. The reason for this improvement may be that *Aloe vacillans* extract that contains phenolic compounds, which might have scavenged the free radicals offering hepatoprotection, and the extract at a dose (500 mg kg-1) is more hepatoprotective.

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References:


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Effect of aqueous crude extract of Aloe vacillans leaves on induced hepatic damage in male rabbits

Mohamed Ali Qassem

كلية الأسنان، جامعة عدن، اليمن

الباحث الممثل: محمد علي قاسم

البريد الإلكتروني: aliunidi3moh1Ali3@gmail.com

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المُلخّص

تهدف هذه الدراسة إلى تقييم مدى تأثير المستخلص المائي لخام أوراق نبات الصبر Aloe vacillans التهاب الكبد المحدث برابع كلوريد الكربون في الأرانب. أحدثت الأذية الكبدية في الأرانب عن طريق الحقن في البريتوان برابع كلوريد الكربون (1 مل/كلم) من وزن الجسم في اليوم السابع واليوم الثامن أثناء حقن المستخلص المائي للنبات في الأرانب عن طريق ألم ثلاث جرعات مختلفة (100 ملجم، 300 ملجم، و 500 ملجم/كلم) من وزن الجسم مرة في اليوم لمدة 7 أيام. بعد 24 ساعة من آخر جرعة ضحى بالحيوانات وجمع الدم لمعايرة أثر الأذية الكبدية والحماية منها بقياس مستويات أنزيمات الكبد في الدم. أظهرت النتائج في المجموعة المعالجة برابع كلوريد الكربون فقط ارتفاعاً معنوي في مستويات أنزيمات الكبد (ALT,AST) والبيليروبين، ونقصاً معنوي في البروتين الكلية. أما في المجموعات المعالجة بالمستخلص المائي فكان هناك تحسن في مستويات أنزيمات الكبد (ALT,AST) والتقليل في البيليروبين وزيادة في البروتين الكلية ناتجة عن تقليل شدة الأذية الكبدية المحدثة في الأرانب بواطبة CCl4.

الكلمات الرئيسية: رابع كلوريد الكربون، اسبارتات أمينو ترانسفيراز، الانين أمينو ترانسفيراز، البروتين الكلي، البيليروبين، أذية كبدية، الأرانب.

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