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Research Article
Camel Urotherapy and Hepatoprotective Effects Against Carbon Tetrachloride-induced Liver Toxicity

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Abstract

Background and Objective: Camel urine contains many active components and essential inorganic elements which plays a protective role as antibacterial, antifungal, antiviral and anticancer agents. Thus, this study aimed to assess the protective role of camel urine (CU) against CCl_4-induced liver damage in rats. In addition, *in vitro* antioxidant activities of the camel urine components that contribute to the hepatoprotective activity against CCl_4-toxicity were identified. **Materials and Methods:** Thirty Sprague Dawley rats divided into 3 groups, 10 rats each. The first group served as controls, the second group received CCl_4 and the third group received CCl_4 plus CU. Histopathological and biochemical measurements evaluated in bold and liver tissues samples following 2 months of treatment. In addition, active components and essential inorganic elements present in CU as well as its antioxidant activity estimated via *in vitro* assessments analyses using GC and ICP-MS analysis and scavenging assays, respectively. **Results:** Significant improvement in a histopathological pattern of liver cells and liver enzymes were reported in the CCl_4/CU treated group compared to CCl_4 treated rats. In the CCl_4/CU treated rats, a significant decrease ($p<0.05$) in SGPT, SGOT and increased ($p<0.05$) in serum albumin concentrations was reported in comparison with CCl_4 treated rats. In addition, 20 active metabolites and 28 inorganic essential elements such as; K, Mg and Mn were significantly identified in CU using GC-MS analysis. Antioxidant activity of these active metabolites and inorganic essential elements were shown to be higher against *in vitro* free radicals (H_2O_2, FRAP and SOR) suggesting the potential antioxidant property of CU against CCl_4 toxicity. **Conclusion:** Camel urine showed to play a promising anti-oxidative and anti-free radical scavenging mechanism against hepatic dysfunction.

Key words: Urotherapy, camel urine, carbon tetrachloride, complementary medicine, alternative medicine, active metabolites

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Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Carbon tetrachloride (CCl_4) is a xenobiotic agent that causes tissue injury in various organs. It induces cytochrome-P450 to produce free radicals, such as; the trichloromethyl radicals (CCl_3) and peroxy trichloromethyl radicals ($OOCCl_3$)[1]. These free radicals bind to proteins, lipids and nucleic acids of the cell causing extensive damage in the liver, kidney, brain and other tissues. The CCl_4 has been used in studies to investigate the effects of different chemicals on the liver and kidney[2,3].

For centuries, Bedouins in the Arabian Peninsula have used camel urine (CU) for treating various diseases and up till today, this practice continues. A recent 2018 cross-sectional study found that 15.7% of cancer patients who sought in complementary or alternative medicine in Saudi Arabia used CU[4]. They drank it alone or mixed with camel milk. There is a belief that CU can be prophylactic and therapeutic for different diseases including cancer. The available research on CU use is limited and incoherent. One study found an *in vitro* pharmacological role for CU in inhibiting platelet aggregation with neither human nor bovine urine showing the same effect[5]. Another study by a researcher investigated the effects of CU cytotoxicity on cancer cells and found that 216 mg mL^{-1} of lyophilized CU can induce apoptosis in >80% in cancer cells. Cancer enhancing proteins, such as; cyclin-D1 and β-catenin were down-regulated[6]. Moreover, CU showed an anti-metastatic effect on breast cancer cells in mice[7].

Camel urine showed a role in preventing the development of liver cancer, it ameliorates tetrachlorodibenzo-p-dioxin (TCDD)-induced CYP1A1 gene expression an oncogene for hepatocellular carcinoma in mice[8]. Drinking CU, however, increases the risk of various infections in immune-compromised patients. Fatal Brucellosis and Middle East Respiratory Syndrome (MERS) infections showed a link with CU consuming[4-8].

A lot of chemical constituents present in camel urine showed several biological activities, particularly as antibacterial, antifungal, antiviral and anticancer agents[6-9]. In Asian countries, the therapeutic efficacies of camel urine were clinically investigated such as; treating of diabetic neuropathy[6-9].

Now-a-days, profiling of urinary acids and metabolites by NMR and GC–MS meets with more importance in clinical studies[9,10]. Although there are not enough clinical studies, camel milk and urine are regarded traditionally as a high-quality alternative remedy for curing a number of diseases[11,12], particularly chronic imbalance of the liver[13,14]. Series of *in vivo* experiments in albino mice showed no side effects of using camel urine and milk in treating liver and stomach[15], this indicated that the biological activity of either milk or urine of the camel or its combination mixtures is a safe alternative medicine. Thus exploring, identifying and characterizing the components present in camel urine were of significant interests[15]. Little is known about the potential activity of camel urine against liver cell toxicity especially in CCl_4-induced liver damage[16].

Previous studies only proposed antioxidant activities for CU against CCl_4-induced liver damage without studying the free radical activities of volatile metabolites and essential inorganic elements present in camel urine (CU)[17]. Thus, this study was aimed to assess the protective role CU against CCl_4-induced liver damage in rats. In addition, *in vitro* antioxidant activities of the camel urine components that contribute to the hepatoprotective activity against CCl_4-toxicity were identified.

MATERIALS AND METHODS

Chemicals: Camel urine (CU) was collected from 4 female lactating camels in Marsa Matrouh desert, Egypt, in 2018. Camels were fed on mountainous weeds and foliage. At first, approximately 250-300 mL of the urine sample from each of the animals was collected directly into stainless steel containers and then transferred to suitable glass vials. Urine samples were then carried out to the laboratory and stored at -80°C until further use. All chemicals used in this study are of analytical grade. Carbon tetrachloride was purchased from Sigma, Aldrich (USA).

Experimental animals: Thirty adult male Sprague Dawley rats, with a mean weight of 220 g were obtained from Mansoura University Animal house. The animals were acclimatized for 30 days at the experiment site animal breeding room under optimal environmental conditions [12 h light-dark cycles, temperature (20±2°C) with moderate humidity (60±5%)]. All the animal handling procedures used in this study were approved based on the guidelines of the experimental animal care, College of Medicine, Institutional Research Board (IRB) of Mansoura University, Mansoura, Egypt. The Ethics Committee of the Experimental Animal Care Society and Institutional Research Board (IRB) at Mansoura University approved the experimental procedures (Permit Number: PT1019).

Experimental design: In this study, rats divided into three groups (10 rats each). The first group served as controls, the second group rats received CCl_4 (3 mL kg^{-1} b.wt., of 50% dissolved in groundnut oil) for 1 day and third group rats received CCl_4 (3 mL kg^{-1} b.wt., of 50% dissolved in ground nut oil) for one day[17,18] and CU (2 mL/100 g by an oral intubation through a special oral tube for 60 days)[16]. Blood samples were taken after 2 months via direct cardiac puncture to measure liver enzyme's level. Animals were sacrificed after two months for histopathological examination of the liver after fixing with 10% formalin.

Assessments of camel urine (CU) metabolites

GC-MS analysis: In this experiment, the camel urinary components estimated by the GC–MS analysis in a Perkin Elmer Clarus 600 gas chromatograph linked to a mass spectrometer (Turbomass)[10]. For extraction of the camel urinary components, dichloromethane (DCM) with a low boiling point was selected whereas most of the urinary components were easily dissolved in DCM and the derivatization reaction was completed within 20 min. Freeze dried samples were dissolved in a suitable amount of DCM[10]. The samples were transferred to GC vials in an appropriate aprotic solvent such as; DCM. Traces of methanol were removed because it could react with the reagent. About 80 lL of BSTFA and 50 lL of pyridine were added to the sample. This amount is enough for a sample containing 100 Lg of total derivatizable material and dissolved in 100 LL of solvent. The vials were capped tightly which were heated at 65°C for 20 min. The heating step was performed to ensure the completion of the reaction. After heating, the samples were allowed to cool down at room temperature and injected on the GC/MS. An aliquot of 2 mL of extract was injected into the Elite-5MS column of 30 m, 0.25 Lm film thickness and 0.25 mm internal diameter[10].

ICP-MS analysis: For the elemental analysis of camel urine the instrument ELAN-DRC-II, Perkin Elmer, USA was utilized[10]. One gram of freeze-dried sample and 50 mL of 20% nitric acid was added to an Erlenmeyer flask. The mixture was heated to 70-85°C for 48 h. During the heating period, the volume of the flask was maintained at the same level intermittently adding 20% nitric acid. After the completion of digestion, the contents of the Erlenmeyer flask filtered using a Nalgene filter (Thermo scientific) unit. The filtrate collected in a 100 mL volumetric flask and allowed to cool. After cooling the volume was made up to 100 mL using Milli Q water and analyzed with ICP-MS[10].

In vitro assessments of camel urine (CU) antioxidant activity

Superoxide radical (SOR) scavenging assay: Scavenging activity of CU against superoxide free radicals (SORs) estimated as previously reported[18]. In this experiment, 50 µL of different concentrations of CU, 50 µL of NBT (200 µM), 50 µL of NADH (624 µM) and 50 µL of phenazine methosulfate (80 µM) added sequentially and the reaction mixture incubated at room temperature for 5 min. Then, tunable automated microplate reader (AVersa Max TM, Molecular Devices, Sunnyvale, CA., USA) used to estimate the absorbance at 560 nm and phosphate buffer used as a negative control. Due to its instability, fresh phenazine methosulfate solution was prepared for each set of experiments[19]. The scavenging percentage of CU calculated according to the following equation[19]:

$$\text{Scavenging (\%)} = \frac{\text{1-Abs at 560 nm}}{\text{ABsC at 560 nm}} \times 100$$

Where:
Abs = Absobance of sample
AbsC = Absorbance of control

***In vitro* H_2O_2 scavenging assay:** Scavenging activity of CU against H_2O_2 free radicals evaluated as mentioned previously[18]. In H_2O_2 scavenging assay, 50 µL of freshly prepared 2 mM H_2O_2 solution, mixed with 50 µL of different concentrations of CU. The reaction mixture incubated at room temperature (20±2°C) for 20 min. The mixture (100 µL) made from freshly prepared HRP (0.3 mg mL^{-1}) and phenol red (4.5 mM) in 0.1 M phosphate buffer added to the reaction mixture containing CU and H_2O_2. Following incubation at room temperature for 10 min, the absorbance measured at 610 nm using the Versa Max TM microplate reader. All CU samples prepared in 50% methanol, while the other used reagents prepared in 0.1 M phosphate buffer (pH 7.4). The scavenging percentage was calculated as[19]:

$$\text{Scavenging (\%)} = \frac{\text{1- Abs at 560 nm}}{\text{ABsC at 560 nm}} \times 100$$

Where:
Abs = Absobance of sample
ABsC = Absorbance of control

Ferric (Fe^{3+}) reducing antioxidant power assay (FRAP): Antioxidant activity of CU significantly evaluated by using BioVision's FRAP assay kit (Catalog # K515-200, S. Milpitas Blvd., Milpitas, CA 95035, USA)[19], which provides a quick, sensitive and easy way for measuring the antioxidant capacity of various biological samples. In this method, after kinetic mode reaction for 60 min at 37°C, antioxidant capacities of active metabolites present in CU identified using ferric reducing antioxidant power (FRAP) whereas, Fe^{3+} reduced to Fe^{2+} at low pH and causes to the formation of a colored ferrous-probe complex from a colorless ferric-probe complex. The absorbance of the resultant blue-colored complex (Fe^{2+} tripyridyltriazine) immediately measured calorimetrically at 594 nm (19-22). The antioxidant capacity of CU samples calculated from the following equation[19-22]:

$$\text{Sample FRAP equivalents} = B\ XD/V = \text{nmol}\ \mu L^{-1} = \text{mM}\ Fe^{2+}\ \text{equivalents}$$

Where:

B = Ferrous ammonium sulphate amount from standard curve (nmol)
D = Dilution factor
V = Sample volume added into the reaction well (µL)

Serum biochemical analysis: Blood samples allowed to clot for 30 min and then centrifuged at 3000 rpm for 15 min. Clear sera obtained and stored at -20°C. Reitman and Frankel's method used to measure serum levels of Aspartate transferase (AST/SGOT) and Alanine transaminase (ALT/SGPT)[23] and the methods by previous report to measure serum albumin concentrations[24-26].

Histopathological examination: A rotatory microtome used to slice 5 µm thick sections from the liver. The sections stained with hematoxylin-eosin (H and E) dye (Merck) and examined at 200x magnification using a power light microscope (Zeiss, Germany). In this experiment, the modified hepatitis activity index (HAI) used efficiently to evaluate liver tissue injury.

Statistical analysis: The Statistical Package for Social Sciences (IBM, Chicago, Version 22 for Windows) employed to conduct the statistical analysis. Data was expressed as means±standard deviations and then, the one way ANOVA test was employed for comparing means. This followed by Tukey's *post hoc* analysis to determine the significance of between-group differences. The statistical significance was assigned at $p<0.05$.

RESULTS

Urinary metabolites and essential inorganic elements in camel urine (CU): In this experiment, 20 different components identified in the camel urine using GC-MS analysis. The most prominent volatile components present in CU were 2-Deoxygalactopyranose (24.1 µg mL^{-1}), Melibiose (23.4 µg mL^{-1}), D-Galactose (19.7 µg mL^{-1}), Pseudouridine (19.3 µg mL^{-1}), Hexadecanoic acid (18.6 µg mL^{-1}), Trans-9-octadecenoic acid (18.3 µg mL^{-1}), D-Glucuronic acid (17.6 µg mL^{-1}), Benzene propanoic acid (16.9 µg mL^{-1}), Azelaic acid (16.7 µg mL^{-1}) and Prostaglandin F1A (16.7 µg mL^{-1}), followed by Hippuric acid, Ribitol, Canavanine and Galactose. This in addition to other volatile compounds estimated in lowest amounts as shown in the Table 1.

Table 1: Urinary metabolites estimated in camel urine (CU) by GC-MS analysis

Metabolites	100 µg of CU (250-300 mL)	Metabolite pathways
2-deoxygalactopyranose	24.1	Glycolysis
Melibiose	23.4	Glycolysis
D-galactose	19.7	Glycolysis
Pseudouridine	19.3	Nucleoside
Hexadecanoic acid	18.6	Fatty acid
Trans-9-octadecenoic acid	18.3	Fatty acid
D-glucuronic acid	17.6	Androgens, mineralocorticoids and fatty acid metabolism
Benzene propanoic acid	16.9	Fatty acid
Azelaic acid	16.7	Tyrosine inhibitor
Prostaglandin F1A	16.7	Arachidonic acid
Hippuric acid	15.9	Carboxylic acid found in herbivores
Ribitol	15.6	Riboflavin and Flavin mononucleotide
Canavanine	14.9	Amino acid
Galactose	13.8	De Ley Doudoroff pathway
N-phenylacetylglycine	13.6	Amino acid
Creatinine	10.2	Creatinine phosphate
Erythritol	9.1	Insecticide
Aminomalonic acid	8.6	Calcium binding properties to proteins
Propanedioic acid	7.9	Amino acid lysine
Pyrotartaric acid	6.8	Amino acid lysine and tryptophan

Table 2: Urinary essential inorganic elements estimated in freeze dried camel urine by ICP–MS

Element name (Symbol)	Concentration of element of camel urine (ppm)
Sodium (Na)	548.293866
Potassium (K)	296.2938
Magnesium (Mg)	18.114516
Manganese (Mn)	14.09652
Chromium (Cr)	2.254291
Iron (Fe)	1.82154
Phosphorus (P)	1.592003
Aluminum (Al)	0.18692
Strontium (Sr)	0.122024
Nickel (Ni)	0.091562
Zinc (Zn)	0.06356
Copper (Cu)	0.054221
Boron (B)	0.036857
Calcium (Ca)	0.029845
Barium (Ba)	0.015605
Lithium (Li)	0.006928
Selenium (Se)	0.002813
Cobalt (Co)	0.002326
Antimony (Sb)	0.002003
Tin (Sn)	0.000563
Arsenic (As)	0.000468
Chlorine (Cl)	0.0004267
Iodine (I)	0.000361
Silver (Ag)	0.000316
Mercury (Hg)	0.000293
Platinum (Pt)	0.000256
Gold (Au)	0.000196
Cadmium (Cd)	0.000096

In addition, most of the volatile components identified in CU chemically related to glycolysis, fatty acid synthesis, nucleoside, arachidonic acid androgens, mineralocorticoids, tyrosine inhibitor and the arachidonic acid pathway (Table 1).

In addition, a total of 28 essential inorganic elements significantly identified in CU using ICP-MS analysis, Na, K, Mg, Mn and Cr levels were present in higher quantities in camel urine followed by Fe and P with minority ratios of other inorganic elements as shown in the Table 2.

***In vitro* camel urine (CU) antioxidant activity:** Higher antioxidant activity of CU was reported against SOR and H_2O_2, FRAP free radical reagents at higher concentrations (100 µg of CU) compared to ($p = 0.001$) those of lower doses (Fig. 1a-c). The CU at a dose of 100 µg showed strong free radical scavenging activity against SOR and H_2O_2 and FRAP free radicals, respectively (Fig. 1a-c). The results indicated that both volatile metabolites along with inorganic elements play an essential role as free radical scavengers against oxidative stress radicals in the biological system which initiates cellular damage (Fig. 1a-c).

Liver function: A significant increase ($p<0.05$) in serum levels of liver enzymes (SGPT and SGOT) and lower concentrations ($p<0.05$) of serum albumin reported in CCl_4 treated rats in comparison with normal control rats, indicating marked liver toxicity of CCl_4. However, treatment of CCl_4 intoxicated rats with CU significantly ($p<0.001$) ameliorated the observed increases in serum levels of both enzymes (Table 3). In addition, treatment of CCl_4 intoxicated rats with CU significantly ($p<0.001$) increased the levels of serum albumin in comparison with rats, treated with CCl_4 alone (Table 3).

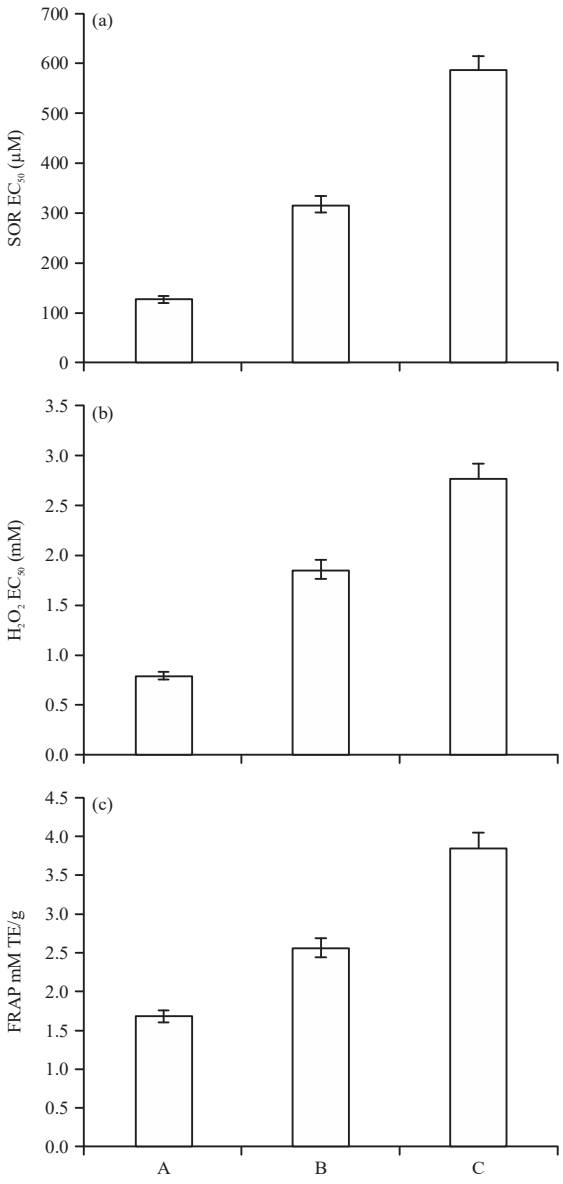

Fig. 1(a-c): *In vitro* SOR scavenging, H_2O_2 scavenging and FRAP value of Camel urine metabolites. Sequential increase in free radical scavenging activity of camel urine (CU) was significantly ($p = 0.001$) reported with higher concentrations of CU values at concentration of (a) 25 µg mL^{-1}, (b) 50 µg mL^{-1} and (c) 100 µg mL^{-1} of CU

TE: Trolox® equivalent

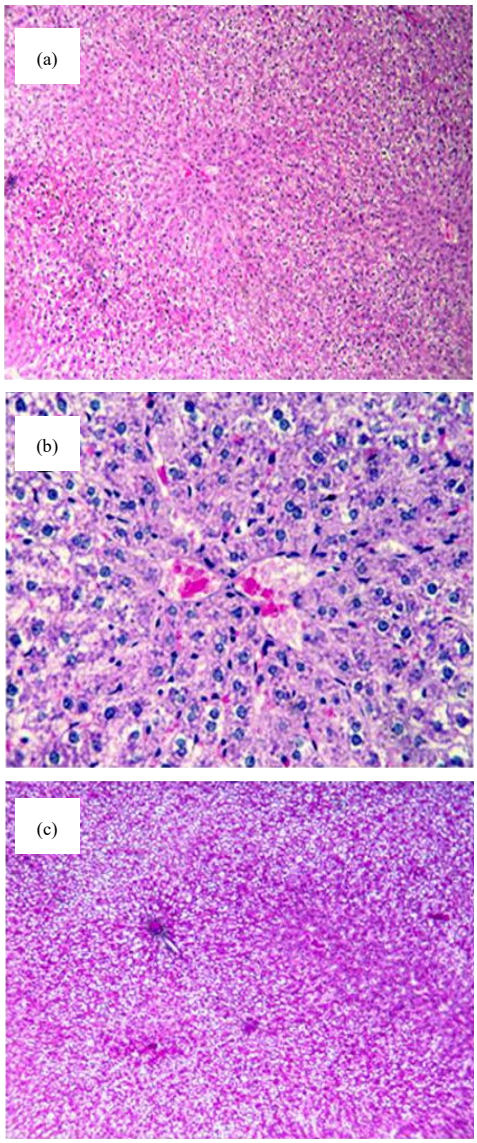

Fig. 2(a-c): Microscopic tissue sections from normal control rats, (a) Preserved architecture with uniform hepatocytes (H and E, 10x), (b) Uniform hepatocytes (H and E, 40x) and (c) No fibrous expansion (Masson, 10x)

Table 3: Effects of camel urine on serum albumin and liver enzymes concentration in CCl_4-intoxicated rats

Parameters	Control	CCl_4	CCl_4+camel urine
SGPT (U L^{-1})	26.6±8.3	54.5±5.7[a]	41.0±8.9[b]
SGOT (U L^{-1})	81.6±8.5	167.6±20.6[a]	121.7±10.6[b]
Albumin (mg dL^{-1})	4.5±0.09	3.7±0.16[a]	3.9±0.08[b]

Data are mean±standard deviations. Values with different superscript letters within the same row are significantly different at [a]p<0.05 (Control vs. CCl_4), [a]p<0.001 (CU treated vs. CCl_4). CCl_4: Carbon tetrachloride, GOT: Glutamic oxaloacetic transaminase, GPT: Glutamic pyruvic transaminase

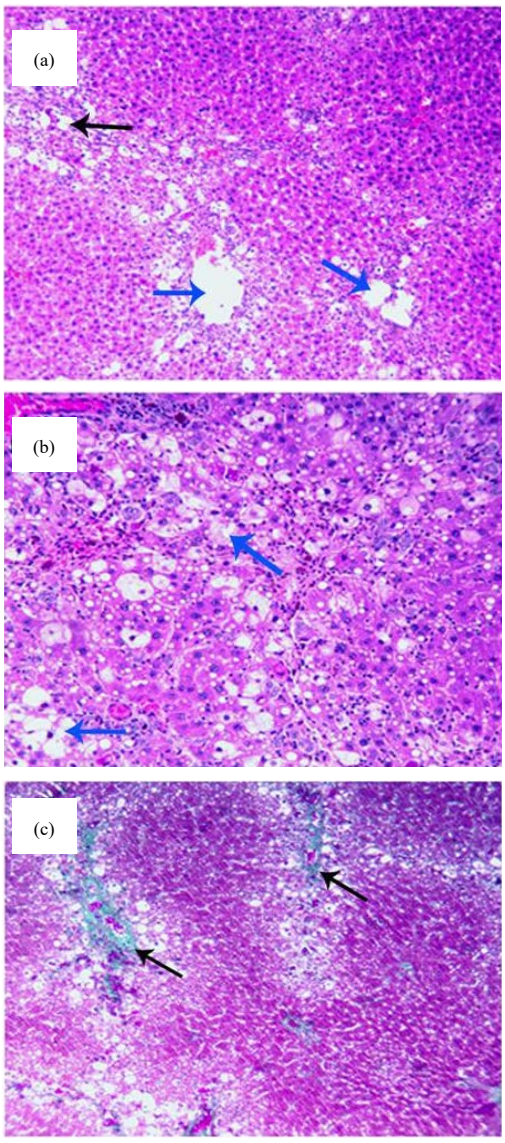

Fig. 3(a-c): Microscopic tissue sections from CCL_4 intoxicated rats, (a) Still preserved architecture, focal confluent necrosis (Black arrow), scattered foci of lytic necrosis (Blue arrows) (H and E, 10x), (b) Scattered foci of lytic necrosis (Blue arrows) (H and E, 20x) and (c) Fibrous expansion of some portal areas with short fibrous septa (Black arrows) (Masson, 10x)

Camel urine and liver cell protection: The control group showed normal liver architecture with uniform hepatocytes appearance in H and E stained sections (Fig. 2a, b) with normal fibrous tissue expansions in Masson trichrome stained sections (Fig. 2c). The CCl_4 treated rats showed a focal confluent necrosis, scattered foci of lytic necrosis in H and E sections (Fig. 3a, b) as well as fibrous expansion of some

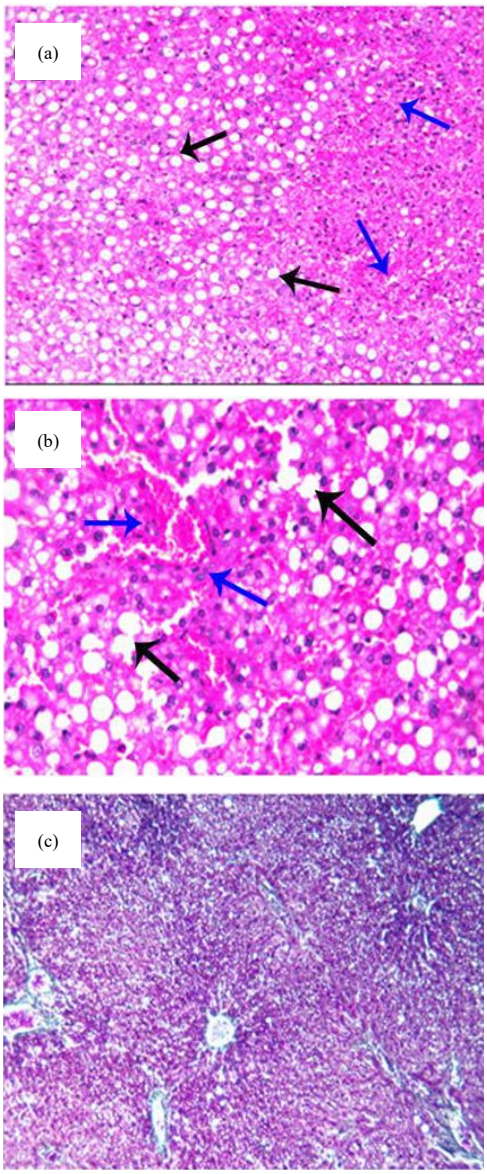

Fig. 4(a-c): Microscopic tissue sections from CCL_4 intoxicated rats treated with camel urine, (a) Steatosis is seen in hepatocytes (Black arrows), focal confluent necrosis is evident (Blue arrow), no evidence of lytic necrosis or portal inflammation (H and E, 20x), (b) Steatosis is seen in hepatocytes (Black arrows), congested sinusoids (Blue arrows) (H and E, 40x) and (c) No fibrous expansion (Masson, 10x)

portal areas with expansion of the fibrous septa (Fig. 3c). Interestingly, in rats treated with CCl_4 and CU, hepatic tissue sections showed significant improvement with no lytic necrosis or portal inflammation in H and E sections (Fig. 4a, b) and less marked fibrous tissue expansion in Masson trichrome stained sections (Fig. 4c), however, there was still some evidence of steatosis with few areas of focal confluent necrosis.

DISCUSSION

In this study, camel urotherapy treatments for 60 days significantly alleviate hepatotoxicity induced by CCl_4 in rats. This urotherapeutic effect of CU was established via *in vitro* antioxidant property of 20 different active volatile metabolites along with 28 essential inorganic elements which were significantly identified in CU using ICP-MS analysis. In this experiment, 20 different components were entirely identified by GC-MS analysis in camel urine, which showed to be varied depending on the climate, food and categories. In addition, most of the volatile components identified in CU chemically related to glycolysis, fatty acid synthesis, nucleoside, arachidonic acid androgens, mineralocorticoids, tyrosine inhibitor and the arachidonic acid pathway. Different metabolites like canavanine, erythritol, benzenepropanoic acid and melibiose were present in camel urine. Canavanine is excreted in camel urine in about 2% of the total urine content and shown to be as a toxic antimetabolite of L-arginine with potent antineoplastic activity[27-30]. The data of this study supported other studies which showed that camel urine components depict significant biological activities especially as anticancer agents[31,32]. Previous research studies showed that the presence of melibiose, D-galactose, 2-deoxy galactopyranose and benzenepropanoic acid as active metabolites of fatty acid derivatives and as the byproducts of glycolysis in CU significantly supported the biological activity of CU as antiplatelet activity, antibacterial, antifungal and anti-parasitic[11-14].

Similarly, a total of 28 essential inorganic elements significantly identified in CU using ICP–MS analysis, Na, K, Mg, Mn and Cr levels were present in higher quantities in camel urine followed by Fe and P with minority ratios of other inorganic elements. The presences of active volatile metabolites along with inorganic elements played an essential role in the biological system which may enhance the cellular biological activities against toxicological and abnormal disorders. Various metals including Na, K, Mg, Ca, Fe, Zn, Cu, Mn, Cr, Mo and Se known to be essentially required for normal biological functions such as; biosynthesis of ATP, the synthesis of DNA and RNA and as a co-factor in more than 300 enzymatic reactions necessary for the structural function of proteins, nucleic acids and mitochondria[6-10,31,32].

Based on the evidence from previous studies, camel urine components are said to inhibit the growth of cancer cells, shrink tumors and secondary metastases, *in vitro* as well as *in vivo*, in humans and animals as well as intoxicated liver cirrhosis, skin and hair ailments[33-36].

In this study, to explore the biological activity of CU metabolites, *in vitro* CU antioxidant activity estimated with SOR and H_2O_2, FRAP free radical reagents. Higher antioxidant activity of CU reported at doses of 100 µg. The CU at a dose of 100 µg showed strong free radical scavenging activity against SOR and H_2O_2 and FRAP free radicals, respectively. The results indicated that both volatile metabolites along with inorganic elements play an essential role as free radical scavengers against oxidative stress radicals in the biological system which initiated cellular damage. The results of this experiment confirmed the previously biological activity of CU against more diseases including cancer, microbial and toxicological effects which lead to cellular damage[6,31-36]. The species of evidence regarding the biological activity of CU containing volatile metabolites along with inorganic elements gave the researchers to study the probable hepatoprotective activity of CU against CCl_4-intoxication. This study also investigated the role of CU in alleviating CCl_4-induced hepatic injury in rats. The CU found to protect against CCl_4-induced liver injury. The levels of SGPT, SGOT and albumin significantly improved with no evidence of fibrous expansion, lytic necrosis or portal inflammation in CU treated rats compared to those intoxicated with CCl_4 only. The CCl_4 shown to produce a severe toxicity via cellular free radical initiation mechanisms which lead to hepatotoxic effects such as; fatty degeneration, fibrosis, inflammation, hepatocellular death, DNA damage, lipid peroxidation and carcinogenicity[37-44].

Matched to the current results, recent interests studied CU biological activity and proved that CU contains high amounts of vitamin C with other active metabolites which have antioxidant activity and was suggested to be of significant importance in treating many diseases[10,46,47]. Similarly, it was reported that CU alone or in combination with Silymarin (hepatoprotective agent) significantly improved liver function and decrease liver cell necrosis on histopathology levels[10,16,47]. This may be explained by the potent antioxidant activity of CU since uric acid and creatinine, present in camel urine are known to be potent antioxidants[47].

The data obtained recommend for future studying the effects of CU metabolites activities against liver cell toxicity on molecular and cellular mechanisms such as; liver cell apoptosis or necrosis. Also, the results of this study implicate for a future full elucidation of CU active metabolites and essential inorganic compounds as promising targets for non-drug therapy against liver cell toxicity.

CONCLUSION

Camel urine showed to play a protective and curative role against hepatic dysfunction via anti-oxidative, anti-free radical scavenging activities of its present volatile metabolites and inorganic essential elements. It could be used efficiently among alternative medicines as a prototype of urotherapy. However; further investigations were recommended at cell molecular levels in future studies.

SIGNIFICANCE STATEMENT

Active volatile metabolites and inorganic essential inorganic elements present in camel urine (CU) significantly alleviate liver cell injury induced by CCl_4 intoxication in rats. This hepato- urotherapeutic effect of CU was established via an antioxidant property of these metabolites which protect liver tissues from cellular oxidative free radicals initiated by CCl_4 toxicity. The proposed hepatoprotective effect of CU was through cellular radical scavenging mechanism which needs further investigations in future studies. The present study proposed that camel urine showed to play a protective and curative role against hepatic dysfunction via anti-oxidative, anti-free radical scavenging activities of its present volatile metabolites and inorganic essential elements. It could be used efficiently among alternative medicines as a prototype of urotherapy.

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