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PHARMACOGNOSIE

Protective Effect of Curcuma Against Chromium Hepatotoxicity in Rats

L'effet protecteur de curcuma contre l'hépatotoxicité du chrome chez les rats

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Abstract This study was carried out to investigate the antioxidant effects of curcuma against chromium-induced alterations in hepatic indices and dysfunctions in the antioxidant system. Forty male *Wistar* rats were randomly divided into four groups and were treated for 30 consecutive days. The control group (0-0) received per os mineral water and normal diet. The second group (0-Cur) received mineral water and an experimental diet containing 2% of curcuma powder, whereas the third group (Cr-0) was orally fed (per os) with 15 mg/kg body weight/day of potassium dichromate and normal diet. The last group (Cr-Cur) received per os 15 mg/kg of potassium dichromate and a diet with 2% of curcuma. The treatment by chromium was found to elicit a perturbation in biochemical parameters producing a significant increase in glycemia, triglycerides, cholesterol, ALP, ALT, AST, and LDH levels. On the contrary, a significant reduction was observed in the oxidative stress-related parameters (GSH, GPx, CAT, and GST). Moreover, we noticed that liver sections of rats intoxicated with chromium showed a disrupted architecture. However, the administration of curcuma revealed an intense reduction in the oxidative stress induced by chromium, ameliorating the levels of the majority of the previous parameters. The data of this study revealed the potent antioxidant effects of curcuma in reducing oxidative stress damage induced by the hexavalent chromium.

Keywords Chromium · Oxidative stress · Hepatotoxicity · Curcuma · Rats

Résumé L'objectif de ce travail est l'étude des effets antioxydants et hépatoprotecteurs du curcuma contre les altérations induites par le chrome. Quarante rats mâles de la souche Wistar ont été divisés en quatre groupes de dix rats chacun, puis traités quotidiennement pendant 30 jours consécutifs. Le groupe témoin (0-0) recevant de l'eau minérale per os et un régime alimentaire normal. Le deuxième groupe (0-Cur) recevant de l'eau minérale et un régime alimentaire contenant 2 % de curcuma, tandis que le troisième groupe (Cr-0) a reçu per os 15 mg/kg de poids corporel par jour de dichromate du potassium et un régime normal. Le dernier groupe (Cr-Cur) recevant per os 15 mg/kg de dichromate du potassium et un régime alimentaire à 2 % de curcuma. Le traitement au chrome a provoqué une perturbation des paramètres biochimiques se traduisant par une augmentation significative de la glycémie, des triglycérides, du cholestérol, des TGP, des TGO, de la PAL et de la LDH. Toutefois, une réduction significative des paramètres liés au stress oxydatif (GSH, GPx, CAT et GST) a été observée. Par ailleurs, les différentes coupes histologiques hépatiques des rats traités par le chrome ont montré une architecture tissulaire perturbée. Cependant, l'administration du curcuma a démontré une réduction intense du stress oxydatif induit par le chrome, améliorant ainsi la majorité des paramètres biochimiques et histologiques précédents. Les données de cette étude ont révélé les puissants effets antioxydants et hépatoprotecteurs du curcuma contre les dommages oxydatifs et les altérations induits par l'administration du chrome.

Mots clés Chrome · Stress oxydatif · Hépatotoxicité · Curcuma · Rats

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Introduction

Heavy metals are highly toxic compounds widely spread in the environment, due to their high degree of toxicity, arsenic, cadmium, chromium, lead, and mercury rank among the priority metals with high public health significance [1]. Chromium is released in the environment from industrial processes, wood preservation, pigment, plating, welding, leather tanning, manufacture of stainless steel, and metal finishing [2]; therefore, its level in the industrial waste consists

of an important health concern related to the environmental contamination [3]. Furthermore, chromium has been reported for its potential genotoxicity, cytotoxicity [4], and carcinogenicity effects [5]. In the same way, its exposure has been linked to cell damages and DNA disruption [6].

The organs that are the most affected by chromium bioaccumulation are liver, kidney, and spleen. In fact, liver is one of the main toxicity targets as it is the biotransformation organ of the majority of xenobiotics. The latter lead to an oxidative stress resulting in the alteration of the antioxidant defense as well as the stimulation of the production of reactive oxygen species (ROS), thus an impairment in hepatic function markers, leading to histopathological changes [7].

Curcuma is a perennial herb of the ginger family that is cultivated in India, China, Indonesia, and other tropical countries. The powder of its dried rhizomes (turmeric) is commonly used as a food additive for organoleptic purposes [8]. The most bioactive element in turmeric is curcumin [9], which has been reported for its powerful medicinal properties over many centuries due to its well-considered phenolic content [10]. In addition, curcuma has exhibited excellent therapeutic activities among which we can list anticancer and anti-inflammatory activities [11], as well as antifungal, antibacterial, antiviral, insecticidal, and nematicidal activities [12]. Likewise, many researchers have reported an antitumor, anti-invasion, antimetastatic, cell growth regulation, radioprotection, radiosensitization, immune modulation, protection in skin diseases, cardioprotective effects, protection from acute and chronic lung diseases, nephroprotective and hepatoprotective effects, and antidiabetic activities [13]. Moreover, it was found that curcuma has antioxidant effects by being a potent scavenger of many reactive oxygen species including superoxide radical (O_2°), hydroxyl radical ($^{\circ}OH$), and hydrogen peroxide (H_2O_2) [14,15].

The present study aims to evaluate the modulation of the toxic effects and the oxidative hepatic injury following the chromium sub-chronic exposure using curcuma as a natural antioxidant.

Materials and methods

Animals

Forty male *Wistar* rats were used in this study (160 ± 10 g body weight; 10 weeks old) and were obtained from Pasteur Institute of Algiers, Algeria. Animals were housed in polypropylene cages and were maintained under standard environmental conditions (12 h light/dark cycle, 21 ± 2 °C temperature, and 50 ± 10% relative humidity). Rats were fed on specific diet prepared according to Upreti et al. [16] regime and water *ad libitum*. The animals were acclimatized for 15 days before the commencement of the experiments. The research procedures were carried out according to the National Institute of Health Guidelines for Animal Care and approved by the Animal Ethics Committee.

Preparation of chromium solution (induction of oxidative stress)

Potassium dichromate ($K_2Cr_2O_7$) powder (Biochem Chemopharma Company, USA) was dissolved in mineral water and was administrated orally by gavage (per os); the volume of each dose was adjusted to deliver 15 mg/kg of body weight/day.

Preparation of Curcuma Powder

Curcuma longa rhizomes were purchased locally from the market. To obtain turmeric fine powder, rhizomes were milled in the laboratory using mortar and pestle, and then were pulverized with a knife grinder and sieved to get uniform size range.

Animal treatment

Rats were randomly divided into four groups and were treated for 30 consecutive days. The first group (0-0) served as control and received mineral water only through oral gavage (per os) with a normal diet. The second group (0-Cur) received mineral water and an experimental diet containing 2% of curcuma powder, while the third group (Cr-0) was treated per os with a dose of potassium dichromate (15 mg/kg body weight) and received a normal diet. As for the fourth group (Cr-Cur), it received both an oral dose of potassium dichromate (15 mg/kg) and an experimental diet containing 2% curcuma powder.

Experimental methods

During 30 days of treatment, animals were weighed daily. At the end of the experimental period, all the animals were sacrificed by cervical decapitation and blood samples were collected in serum plain tubes. These samples were centrifuged at 5,000 rpm for 10 minutes to obtain serum, which afterward served for the measurement of various biochemical analyses (glycemia, triglycerides, cholesterol, alkaline phosphatase, lactate dehydrogenase, alanine aminotransferases, and aspartate aminotransferases) using automatic biochemistry analyzer (ARCHITECT ci4100) and kits that were provided by Spinreact, Spain.

Liver was immediately extracted from dissected rats, washed with a phosphate buffer (0.1 M, pH = 7.4) to remove the excess blood and adhering tissues, weighed, and then divided into two parts. The first part of the liver was immediately fixed in 10% formol solution for histological study, whereas the second part was preserved at −20 °C to stop

metabolic activities and served for oxidative stress parameters. The hepatosomatic index shows the weight of liver as a percentage of body weight [17]. The hepatic reduced glutathione content (GSH) and protein concentration were determined by spectrophotometry using the methods of Weckbercker and Cory [18], and Bradford [19], respectively. Catalase activity (CAT) was estimated by the Aebi [20] method, while glutathione peroxidase activity (GPx) was measured according to Flohé and Günzler [21] method. Finally, the glutathione S-transferase (GST) activity was estimated by Habig et al. [22] method.

Statistical analysis

All values are expressed as means ± SEM. Data were performed by one-way analysis of variance (Anova) by multiple comparisons. The Graph Pad Prism 5.0 software was used to draw the graphs (Graph Pad software, Inc., San Diego, USA). Results were considered as statistically significant when $P < 0.05$.

Results

Physiological Parameters' Study

Exposure of rats to chromium did not produce any overt sign of toxicity/mortality. However, Table 1 shows a significant decrease in body weight accompanied by an increase in liver wet weights and hepatosomatic index in chromium-treated rats (Cr-0) in comparison with control group. On the contrary, the curcuma addition in (Cr-Cur) group ameliorated these settings compared to (Cr-0) group.

Biochemical Parameters' Study

Results about the determination of biochemical parameters, shown in table 2, affirm the existence of a large metabolic disturbance. The oral chromium administration to rats caused a significant increase in glycemia, triglycerides, cholesterol, ALP, LDH, AST, and ALT compared to those of control group, while the supplementation of curcuma in the diet of the chromium–curcuma-treated group (Cr-Cur) stopped the hepatic profile disturbance and generally restored these biochemical parameters to near normal level.

Oxidative Stress Parameters' Study

Chromium intake was found to cause an impressive oxidative stress which occurs mainly through a significant reduction in all biomarkers of antioxidant defense systems in liver, especially GSH level (Fig. 1) and activities of GPx (Fig. 2), CAT (Fig. 3), and GST (Fig. 4) compared to the control grope. Conversely, the co-administration of curcuma with chromium in (Cr-Cur) group pronounced a significant amelioration of the abovementioned oxidative markers by significant increase compared to those of (Cr-0) group.

Histopathological examination

The representative photomicrographs in figure 5 show the transverse sections of liver tissue from the studied groups. Control (5a) shows normal hepatocytes tightly attached to each other containing centrilobular vein zone (CV). Unlike curcuma group (5b), which shows normal hepatic cells, chromium administered rats (5c) show congested hepatocytes (asterisks in 5c3). Moreover, chromium intoxication caused hepatocytes vacuolization (arrowheads in 5c1, 5c2), several parenchyma dilatations were observed throughout the hepatic tissue (closed arrows in 5c1, 5c2, 5c3, 5c5), as well as endotheliitis involving the centrilobular vein (open arrows in 5c4, 5c5), and leucocyte inflammatory cells (round head arrow in 5c1). In contrast, liver of chromium–curcuma-treated group (5d) shows less liver damage by presenting normal hepatic cells and parenchyma when compared to chromium-administered rats.

Table 1 Physiological parameters in control (0-0) and treated rats (0-Cur, Cr-0, Cr-Cur) after 30 days of treatment (values represent the mean ± SEM of seven rats)

Parameters		Groups			
		0-0	0-Cur	Cr-0	Cr-Cur
Body weight	Initial (g)	226.3 ± 3.44	223.7 ± 1.89	220.7 ± 3.70	226.7 ± 3.40
	Final (g)	268.0 ± 8.29	248.9 ± 3.31	231.0 ± 9.76*	251.7 ± 6.33
	Gain (%)	18.42	11.26	4.66	11.02
Live weight	Wet weight (g)	5.86 ± 0.30	6.24 ± 0.19	7.80 ± 0.22***	6.62 ± 0.14*$^{\#\#\#}$
	Hepatosomatic index (%)	15.39 ± 0.90	15.32 ± 0.61	18.23 ± 0.47*	16.42 ± 0.32$^{\#\#}$

*$P < 0.05$, ***$P < 0.001$; Significantly difference from control (0-0) group
$^{\#\#}P < 0.01$, $^{\#\#\#}P < 0.001$; Significantly difference from (Cr-0) group

Table 2 Serum biochemical markers in control (0-0) and treated rats (0-Cur, Cr-0, Cr-Cur) after 30 days of treatment (values represent the mean ± SEM of seven rats)

Parameters	Groups			
	0-0	**0-Cur**	**Cr-0**	**Cr-Cur**
Glycemia (g/L)	0.76 ± 0.03	0.73 ± 0.02	1.02 ± 0.09*	0.91 ± 0.04*
Triglycerides (g/L)	0.62 ± 0.07	0.58 ± 0.14	1.39 ± 0.18**	0.78 ± 0.12$^{\#}$
Cholesterol (g/L)	0.35 ± 0.04	0.37 ± 0.05	0.62 ± 0.04**	0.51 ± 0.05*
ALP (UI/l)	140.3 ± 8.03	134.3 ± 13.81	281.6 ± 26.9***	165.5 ± 9.07$^{\#}$
LDH (UI/l)	2559 ± 314.5	2396 ± 199.5	3586 ± 242.8*	3263 ± 256.1
ALT (UI/l)	21.12 ± 2.03	19.65 ± 2.28	34.83 ± 4.16*	29.49 ± 2.09*
AST (UI/l)	213.7 ± 14.20	181.5 ± 17.25	311.7 ± 10.38***	275.1 ± 12.27**$^{\#}$

*$P < 0.05$, **$P < 0.01$, ***$P < 0.001$; Significantly difference from control (0-0) group
#$P < 0.05$, ##$P < 0.01$, ###$P < 0.001$; Significantly difference from (Cr-0) group

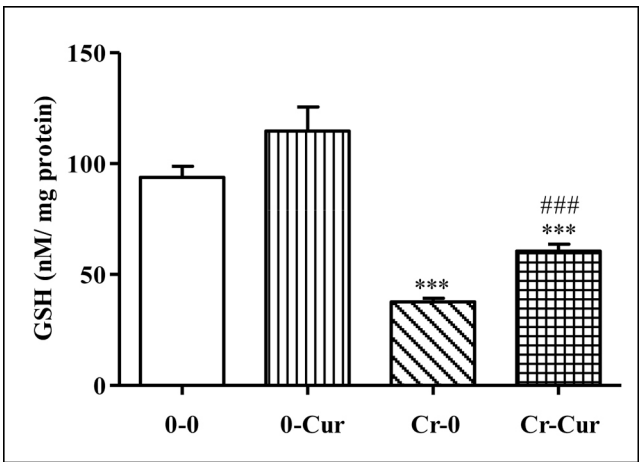

Fig. 1 Glutathione content (nM/mg protein) in control (0-0) and treated rats (0-Cur, Cr-0, Cr-Cur) after 30 days of treatment (values represent the mean ± SEM of seven rats). ***$P < 0.001$; Significantly difference from control (0-0) group, ###$P < 0.001$; Significantly difference from (Cr-0) group

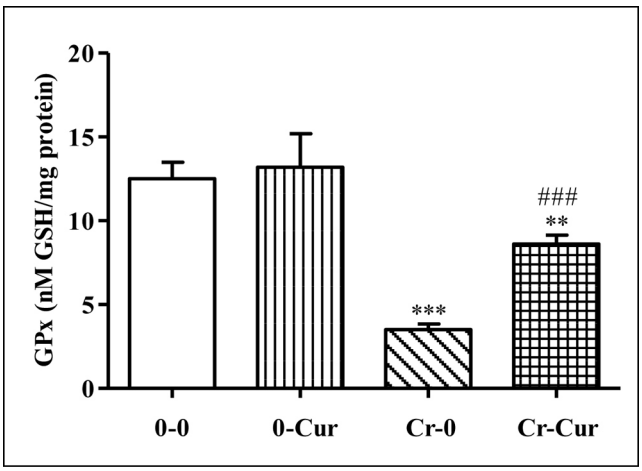

Fig. 2 Glutathione peroxidase activity (nM GSH/mg protein) in control (0-0) and treated rats (0-Cur, Cr-0, Cr-Cur) after 30 days of treatment (values represent the mean ± SEM of seven rats). **$P < 0.01$, ***$P < 0.001$; Significantly difference from control (0-0) group, ###$P < 0.001$; Significantly difference from (Cr-0) group

Discussion

The protection against heavy metals is a problem that is not been solved so far. Transition metals can act as catalysts for the oxidative deterioration of macromolecules, through the formation of reactive oxygen species (ROS), increased lipid peroxidation, decreased sulfhydryl groups, and oxidative tissue damage [23]. This study has a major aim to evaluate the impact of chromium, a toxic pollutant, on adult *Wistar* rats and the possible protective action of curcuma, a spice rich in natural antioxidants.

Regarding the studied physiological parameters, the 15 mg/kg of potassium dichromate treatment for 30 days induced a critical decrease in the body weight of rats with an increase in liver wet weight leading eventually to a high hepatosomatic index. Likewise, the study of De Lucca et al. [24] reported the toxic effect of chromium attributed to changes in bone formation and metabolic disorders. This can be explained by the hypertrophy of the liver tissue and the intense accumulation of the metal within the organ. As for the loss of weight, it is also due to the loss of appetite following the alteration of the food organoleptic proprieties caused by the chromium [25]. Moreover, we have found through our investigation that supplementation with curcuma in (Cr-Cur) group had a protective impact against the adverse action of chromium by keeping the previous parameters in the range of normal values. Similarly, García-Niño et al. [26] reported the potential protective effect of curcumin toward liver damages.

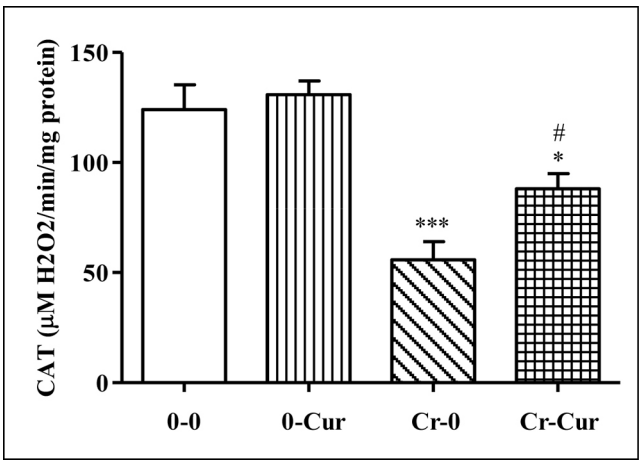

Fig. 3 Catalase activity (µM H_2O_2/min/mg protein) in control (0-0) and treated rats (0-Cur, Cr-0, Cr-Cur) after 30 days of treatment (values represent the mean ± SEM of seven rats). *$P < 0.05$, ***$P < 0.001$; Significantly difference from control (0-0) group, #$P < 0.05$; Significantly difference from (Cr-0) group

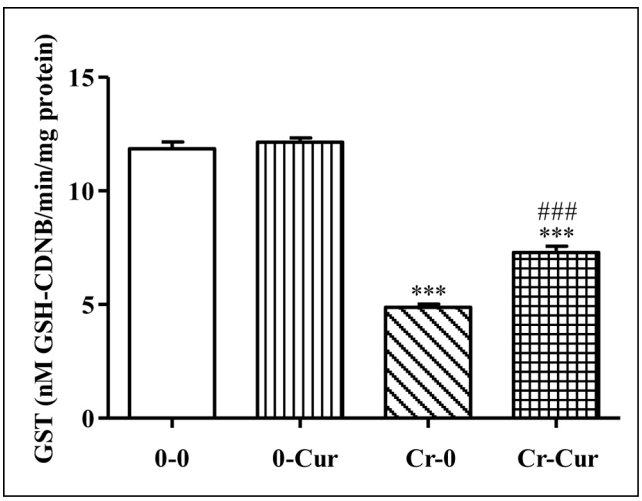

Fig. 4 Glutathione S-transferase activity (nM GSH-CDNB/min/mg protein) in control (0-0) and treated rats (0-Cur, Cr-0, Cr-Cur) after 30 days of treatment (values represent the mean ± SEM of seven rats). ***$P < 0.001$; Significantly difference from control (0-0) group, ###$P < 0.001$; Significantly difference from (Cr-0) group

In our study, exposure to chromium induced a significant increase in the serum glucose levels; this result is consistent with the study of Ghafghazi et al. [27], suggesting that the chromium, as a hyperglycemic factor, inhibits insulin release correlated with insulin resistance. Our results also revealed that treatment with curcuma ameliorates the glycemia, which can be attributed to the anti-hyperglycemic effect of curcuma. However, previous studies on adult rats mentioned the curcumin as potential treatment for diabetes that enhances

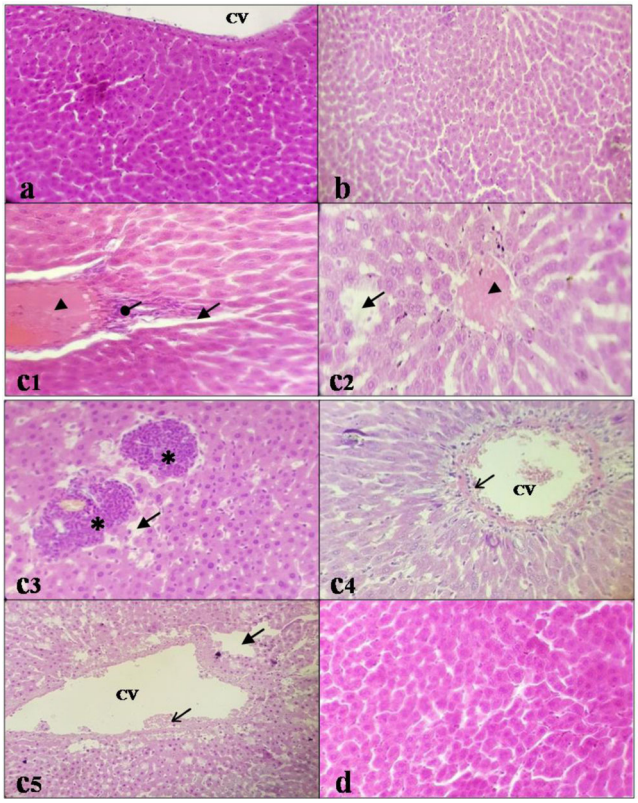

Fig. 5 Histological sections of the liver in control (0-0) and treated rats (0-Cur, Cr-0, Cr-Cur) after 30 days of treatment. Optic microscopy (×400, H&E)

the insulin resistance, by interfering within the pancreatic β-cells, and contributing to the hypoglycemia [28], which improves the absorption and utilization of glucose.

As for the lipid profile, we observed a significant increase in the serum concentration of cholesterol and triglycerides in rats treated with chromium, and this hyperlipidemia can be explained by intense disruption of lipid and cholesterol metabolism and can be a sign for membrane lipid peroxidation. Experimental studies on rats with chromium administration for 30 days induced hypercholesterolemia due to the hypermetabolic state of the animals, or to the impaired liver function [29]. Conversely, the administration of curcuma in the (Cr-Cur) group demonstrated a significant improvement in the serum cholesterol and triglycerides. In fact, curcuma participates in the decrease of lipid status [30], and it was reported by Feng et al. [31] that curcumin possesses ability to inhibit the absorption of cholesterol by suppressing the expression of NPC1L1, the principal transporter of cholesterol in the enterocytes.

The present study showed a significant elevation in the alkaline phosphatase (ALP) activity following the chromium administration, which probably due to the alteration of the hepatocyte membrane permeability or necrosis. These findings agree with those reported by Al-Heidery et al. [32]. On

the contrary, the pretreatment with curcuma maintained the ALP activity at the normal state. In fact, many studies noted that curcuma is an inhibitor of the ALP activity in the plasma [33]. Apparently, curcuma has a potential protective impact on reducing the enzymes released by damaged hepatocytes (AST, ALT, and ALP) and can also inhibit the secretion of liver necrosis indices such as TNF-a and IL-1 [34].

Lactate dehydrogenase (LDH) is a key enzyme in the anaerobic pathway, playing an important role in the production of energy, by catalyzing the interconversion of pyruvate into lactate in the process of glycolysis. The variations in LDH reflect the metabolic alterations. In cell culture medium, the different concentrations of potassium dichromate induce cell death and consequently increase the LDH release [35]. Similarly, our results revealed a significant increase in LDH activity following the chromium exposure, but it also revealed that the treatment with curcuma in the administrated-chromium rats improved LDH activity. Bao et al. [36] mentioned that the pretreatment with curcumin induces powerful antioxidant effects and inhibits the release of LDH from damaged hepatocytes by decreasing lipid peroxidation, through his scavenging properties for free radicals [37].

Transaminases (AST, ALT) are biomarkers of tissue damage reflecting the functional activity of liver and heart; their increase is generally related to alteration of the hepatocyte membrane permeability, leading to an important leakage of enzymes in the plasma [38]. Our findings showed a significant elevation of serum transaminases activities for the chromium-exposed group, compared to the control group, indicating both liver and heart damages. These results are in line with the data published by Elshazly et al. [7], reporting that the exposure to the chromium induces the increase in serum ALT levels, which indicates the presence of hepatocellular and hepatobiliary lesion, linked to cell destruction. We also found that curcuma reduced the transaminase enzyme activities in chromium-curcuma treated rats, which recovers the liver damage. Moreover, it stabilizes the hepatocytes membranes and repairs the alterations of the liver tissue by scavenging free radicals, which also improves the histological architecture, by suppressing the leakage of extracellular enzymes [39].

The glutathione (GSH) is a tripeptide responsible for the cytoprotection against ROS and the detoxification of endogenous and exogenous toxins of electrophilic compounds [25]. In addition, depletion of GSH can impair the cell defense against the toxic actions of ROS and can result in cell damages [40]. In this study, chromium induced a decrease in hepatic GSH concentration. Kart et al. [41] reported that after intraperitoneal injection of chromium and the GSH in mice, the GSH allows reducing extracellular chromium, preventing then the oxidative stress status. Moreover, the treatment with curcuma significantly improved GSH levels. In fact, curcuma increases the availability of GSH. Also, it protects the cell against oxidative damages induced during GSH depletion, as a supplement it enhances both cellular GSH levels and ROS scavenging ability [42].

The cell possesses for its protection a first antioxidant defense system, the GSH that has a sulfhydryl function (-SH). The latter permits the GSH to bind to toxic metabolites. Besides, GSH, in synergy with a complex antioxidant system including superoxide dismutase (SOD), glutathione peroxidase (GPx), glutathione S-transferase (GST), and catalase (CAT), plays an antioxidant role that neutralizes ROS such as hydroxyl radicals (°OH), superoxide anions (O_2°), and hydrogen peroxide (H_2O_2). Furthermore, Yang and Lee [43] reported that these antioxidant enzymes can serve as redox biomarkers in various human diseases, because they are the first to indicate the cell oxidoreduction state. Our results showed a significant reduction in the activity of these enzymes following chromium exposure. According to Dolai et al. [44], the decline of GPx activity in male rats induced by chromium is due to the intracellular accumulation of ROS with development of liver injury. While the CAT decrease indicates the overproduction of H_2O_2 during the chromium exposure, thus treatment with chromium leads to an alteration of the hepatic antioxidant system, confirmed by the decline of antioxidant enzymatic activities such as CAT and GPx. In fact, this is due to the inhibition of antioxidant enzymes and leads to the accumulation of H_2O_2 [45]. Conversely, as shown by our results, the pretreatment with curcuma reveals a significant protection against the reduction of the antioxidant enzymatic activities. Similar study performed by García-Niño et al. [46] reported that curcumin induced a significant increase in the GSH content, GPx, and GST activities.

Conclusion

The present study reveals a clear hepatoprotective effect of curcuma through its antioxidant effects resulting in biochemical and physiological improvement against ROS generation. Hence, curcuma can be qualified as a therapeutic agent against chromium toxicity.

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Conflicts of Interests: we wish to confirm that there are no known conflicts of interest associated with this publication, and there has been no significant financial support for this work that could have influenced its outcome.

References

1. Tchounwou PB, Yedjou CG, Patlolla AK, et al (2012) Heavy metals toxicity and the environment. Clin Environ Toxicol 101:133–64
2. Blade LM, Yencken MS, Wallace ME, et al (2007) Hexavalent chromium exposures and exposure-control technologies in American enterprise: results of a NIOSH field research study. J Occup Environ Hy 4:596–618
3. Mishra S, Bharagava RN (2016) Toxic and genotoxic effects of hexavalent chromium in environment and its bioremediation strategies. J Environ Sci Health C Environ Carcinog Ecotoxicol Rev 34:1–32
4. Patlolla A, Barnes C, Hackett D, et al (2009) Potassium dichromate induced cytotoxicity, genotoxicity and oxidative stress in human liver carcinoma (HepG2) cells. Int J Environ Res Public Health 6:643–53
5. Kim J, Seo S, Kim Y, et al (2018) Review of carcinogenicity of hexavalent chrome and proposal of revising approval standards for an occupational cancer in Korea. Ann Occup Environ Med 30:7
6. Khalil S, Awad A, Elewa Y (2013) Antidotal impact of extra virgin olive oil against genotoxicity, cytotoxicity and immunotoxicity induced by hexavalent chromium in rat. Int J Veterinary Sci Med 1:65–73
7. Elshazly MO, Morgan AM, Ali ME, et al (2016) The mitigative effect of *Raphanus sativus* oil on chromium-induced geno- and hepatotoxicity in male rats. J Adv Res 7:413–21
8. Lim TK (2016) Edible medicinal and non-medicinal plants. Modified Stems & Roots Edition, Bulbs, London, 12 p
9. Paulucci VP, Couto RO, Teixeira CC, et al (2013) Optimization of the extraction of curcumin from *Curcuma longa* rhizomes. Braz J Pharmacogn 23:94–100
10. Caiqin L, Weiqing C, Nan W, et al (2018) Optimization of extraction of antioxidants from turmeric (*Curcuma longa* L) using response surface methodology. J Natural Sci 23:63–9
11. Aggarwal BB, Yuan W, Li S, et al (2013) Curcumin-free turmeric exhibits anti-inflammatory and anticancer activities: Identification of novel components of turmeric. Mol Nutr Food Res 57:1529–42
12. Luthra PM, Singh R, Chandra R (2001) Therapeutic uses of *Curcuma longa* (turmeric). Indian J Clin Biochem 16:153–60
13. Aggarwal BB, Surh YJ, Shishodia S (2007) The molecular targets and therapeutic uses of curcumin in health and disease. Advances in experimental medicine and biology, New York (USA), 595 p
14. García-Niño WR, Pedraza-Chaverrí J (2014) Protective effect of curcumin against heavy metals-induced liver damage. Food Chem Toxicol 69:182–201
15. Tanvir EM, Sakib Hossen MD, Fuad Hossain MD, et al (2017) Antioxidant properties of popular turmeric (*Curcuma longa*) varieties from Bangladesh. J Food Quality 2017:1–8
16. Upreti KK, Das M, Kumar A, et al (1989) Biochemical toxicology of argemone oil. IV. Short-term oral feeding response in rats. Toxicology 58:285–98
17. Berté S, Kouamélan EP, Ouattara NI, et al (2008) Cycle de reproduction et fécondité de *Distichodus rostratus* (Characiformes, Distichodontidae) dans un bassin ouest africain (fleuve Bandama, Côte-d'Ivoire) [Reproductive cycle and fecundity of *Distichodus rostratus* (Characiformes, Distichodontidae) in a West African basin (Bandama River, Ivory Coast)]. Tropicultura 26:104–7 [French]
18. Weckbercker G, Cory JG (1988) Ribonucleotide reductase activity and growth of glutathione-depended mouse leukaemia L1210 cells in vitro. Cancer Lett 40:257–64
19. Bradford MM (1976) A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. Anal Biochem 72:248–54
20. Aebi H (1984) Catalase in vitro. Methods Enzymol 105:121–6
21. Flohé L, Günzler WA (1984) Assays of glutathione-peroxidase. Methods Enzymol 105:114–21
22. Habig WH, Pabst MJ, Jakoby WB (1974) Glutathione-S-transferases. The first enzymatic step in mercapturic acid formation. J Biol Chem 249:7130–9
23. Kehili N, Saka S, Aouacheri O (2017) L'effet phytoprotecteur de la nigelle (*Nigella sativa*) contre la toxicité induite par le cadmium chez les rats [The phytoprotective effect of nigella (*Nigella sativa*) against the toxicity induced by cadmium in rats]. Phytothérapie 1:1–10 [French]
24. De Lucca RC, Dutrey PL, Villarino ME, et al (2009) Effect of different doses of hexavalent chromium on mandibular growth and tooth eruption in juvenile Wistar rats. Exp Toxicol Pathol 61:347–52
25. Saka S, Aouacheri O (2017) The investigation of the oxidative stress-related parameters in high doses methotrexate-induced albino Wistar rats. J Bioequiv Availab 9:372–6
26. García-Niño WR, Tapia E, Zazueta C, et al (2013) Curcumin pretreatment prevents potassium dichromate-induced hepatotoxicity, oxidative stress, decreased respiratory complex I activity, and membrane permeability transition pore opening. Evid Based Complement Alternat Med 2013:1–19
27. Ghafghazi T, Maghbareh A, Barnett R (1979) Chromium-induced hyperglycemia in the rat. Toxicology 12:47–52
28. Zhang DW, Fu M, Gao SH, et al (2013) Curcumin and diabetes: a systematic review. Evid Based Complement Alternat Med 2013:1–16
29. Kumar A, Barthwal R (1991) Hexavalent chromium effects on hematological indices in rats. Bull Environ Contam Toxicol 46:761–8
30. Lee CH, Kim AY, Pyun CW, et al (2014) Turmeric (*Curcuma longa*) whole powder reduces accumulation of visceral fat mass and increases hepatic oxidative stress in rats fed a high-fat diet. Food Sci Biotechnol 23:261–7
31. Feng D, Ohlsson L, Duan RD (2010) Curcumin inhibits cholesterol uptake in Caco-2 cells by down-regulation of NPC1L1 expression. Lipids Health Dis 9:1–5
32. Al-Heidery HHA, Mashkur SH, Al-Heidery ZHA (2008) Protective effects of vitamin C on albino rats Exposed to hexavalent chromium (Cr VI). Kufa Med J 11:1
33. Peschel D, Koerting R, Nass N (2007) Curcumin induces changes in expression of genes involved in cholesterol homeostasis. J Nutr Biochem 18:113–9
34. Salahshoor M, Mohamadian S, Kakabaraei S, et al (2016) Curcumin improves liver damage in male mice exposed to nicotine. J Tradit Complement Med 6:176–83
35. Das J, Kang MH, Kim E, et al (2015) Hexavalent chromium induces apoptosis in male somatic and spermatogonial stem cells via redox imbalance. Sci Rep 13921:1–15
36. Bao W, Li K, Rong S, et al (2010) Curcumin alleviates ethanol-induced hepatocytes oxidative damage involving heme oxygenase-1 induction. J Ethnopharmacol 128:549–53
37. Naik RS, Mujumdar AM, Ghaskadbi S (2004) Protection of liver cells from ethanol cytotoxicity by curcumin in liver slice culture in vitro. J Ethnopharmacol 95:31–7
38. Kumari K, Ranjan N, Sinha RC (2011) Multiple biomarker response in the fish *Labeo rohita* due to hexavalent chromium. 2nd International Conference on Biotechnology and Food Science IPCBEE vol.7. IACSIT Press, Singapore
39. Joshia D, Mittal DK, Shukla S, et al (2016) *Curcuma longa* Linn extract and curcumin protect CYP 2E1 enzymatic activity against

mercuric chloride-induced hepatotoxicity and oxidative stress: a protective approach. Exp Toxicol Pathol 69:373–82
40. Aouacheri O, Saka S, Krim M, et al (2015) The investigation of the oxidative stress related parameters in type 2 diabetes mellitus. Can J Diabetes 39:44–9
41. Kart A, Koc E, Dalginli KY, et al (2016) The therapeutic role of glutathione in oxidative stress and oxidative DNA damage caused by hexavalent chromium. Biol Trace Elem Res 174:387–91
42. Harish G, Venkateshappa C, Mythri RB, et al (2010) Bioconjugates of curcumin display improved protection against glutathione depletion mediated oxidative stress in a dopaminergic neuronal cell line: implications for Parkinson's disease. Bioorg Med Chem 18:2631–8
43. Yang HY, Lee TH (2015) Antioxidant enzymes as redox-based biomarkers: a brief review. BMB Rep 48:200–8
44. Dolai DP, Dey SK, Dash SK, et al (2016) Study the chromium-induced oxidative stress on mitochondria from liver and lungs origin. Am J Appl Sci Res 2:59–64
45. Soudani N, Bouaziz H, Sefi M, et al (2013) Toxic effects of chromium (VI) by maternal ingestion on liver function of female rats and their suckling pups. Environ Toxicol 28:11–20
46. García-Niño WR, Zatarain-Barrón ZL, Hernández-Pando R, et al (2015) Oxidative stress markers and histological analysis in diverse organs from rats treated with a hepatotoxic dose of chromium (VI): effect of curcumin. Biol Trace Elem Res 167:130–45