Introduction and Objectives: Leflunomide is a drug used to treat autoimmune diseases. However, despite its therapeutic benefits, adverse effects have been reported after six months of treatment, including liver damage.

Materials and Patients: A 19-year-old female from Acayucan, Veracruz, without significant hereditary or family history, drug addiction denied, with a pathological history of hypothyroidism for 2 years and rheumatoid arthritis since the age of 15, treated with leflunomide 20 mg, chloroquine 150 mg, levothyroxine 100 mcg. She presented in October 2022 with nausea, vomiting, postprandial abdominal pain, jaundice. Abdominal ultrasound revealed chronic calculous cholecystitis. Laparoscopic cholecystectomy was performed in December 2022 without complications. After surgical treatment, he persisted with jaundice, alteration of liver function tests of mixed pattern that evolves to cholestatic pattern (R factor 0.2, see Table 1). Non-reactive to viral hepatitis A, B and C, HIV, negative antibodies ANA, AML, AMA, anti-DNAds, anti-Smith, anti-Rho, complement C3 145 and C4 33. Cholangiography, with hepatomegaly, diffuse hepatic steatosis without evidence of biliary lesions. Liver biopsy showed features of portal and lobular hepatitis, intracytoplasmic and intracanalicular cholestasis, macrovesicular steatosis, and F2 fibrosis according to METAVIR (Fig. 1), which were not consistent with autoimmune or viral disease. RUCAM score with possible relation to hepatoxicity (4 points), so it is considered drug-induced liver injury (leflunomide).

Results: Leflunomide was discontinued and treatment was adjusted to chloroquine 150 mg and prednisone 50 mg. Clinical and biochemical improvement was observed.

Conclusions: This case highlights the importance of suspecting DILI in patients treated with potentially hepatotoxic drugs and with alterations in liver biochemistry. It is a rare disease for which there are no specific markers. Therefore, liver biopsy is a useful tool for early diagnosis.

Ethical statement

The identity of the patients is protected. Consentment was obtained.

Declaration of interests

None

Funding

This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

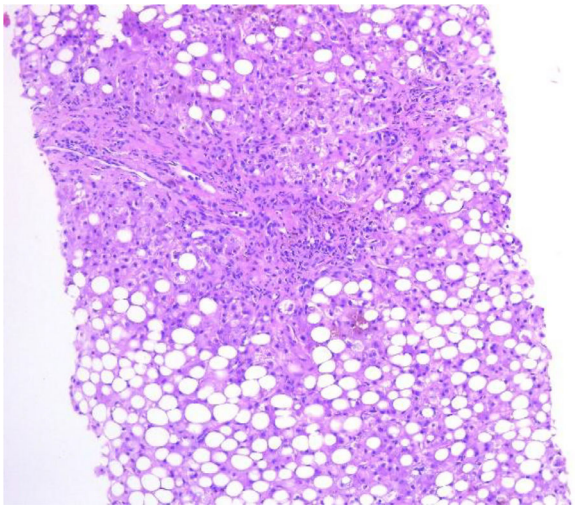

Figure 1. Liver tissue with widened portal spaces, mild inflammatory portal infiltrate, thick droplet steatosis, intracytoplasmic and ductal cholestasis.

Table 1
Control of laboratory studies

Leflunomide is discontinued

PARAMETER	11-Feb	23-Feb	23-Mar	21-Apr	24-May	5-Jul
Total bilirubin (mg/dL)	3.8	4.1	8.61	14.5	8.15	0.76
Direct bilirubin (mg/dL)	2.47	3.3	6.64	7.81	-	0.34
Indirect bilirubin (mg/dL)	1.33	0.8	1.97	6.77	-	0.42
AST (U/L)	65	95	277	236	168	48
ALT (U/L)	18	29	41	64	101	50
Alkaline phosphatase (U/L)	253	142	569	217	146	132
GGT (U/L)	245	413	322	702	607	154
TG (mg/dL)	-	-	803	411	225	155
Cholesterol (mg/dL)	-	-	324	289	296	194

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Evaluation of the effect of Cinnamomum cassia oil on markers of oxidative stress and its modification in gene expression in a diabetic rat model induced with alloxane.

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Introduction and Objectives: Diabetes Mellitus (DM) is a chronic hyperglycemia disorder presenting alteration of biochemical markers, proinflammatory activity, and oxidant stress (OS). There are treatments for DM but they can have adverse effects, so plants are an alternative to new therapeutic compounds. Cinnamomum cassia (cinnamon) has been shown to have antidiabetic and antioxidant activity. The objetive of this study was to evaluate the effect of Cinnamomum cassia oil (CCO) on oxidative stress markers and their modification in gene expression in a diabetic rat model induced with alloxan.

Materials and Methods: Experimental, prospective and comparative study with female and male Wistar rats. Groups (n=6): Sham (SH), Diabetic (D), CCO, D with CCO (D+CCO) and D with metformin (D+MET). From serum and liver tissue, biochemical and antioxidant markers were measured respectively, as well as gene expression. Ethics Committee approval under HI17-00002 registry.

Results: No significant difference in ALT and AST was observed between the SH and CCO groups at the dose used (300 mg/kg) (Figure 1A y B). Group D presented an increase in glucose (GLU) compared to SH (Figure 1C). A significant decrease in GLU, urea nitrogen (BUN),

AST and ALT were observed in the D+CCO group compared to D group, but not in cholesterol (COL), triglycerides (TG), creatinine (CREA) (Figure 1D-J). No significant changes were observed in the levels of malondialdehyde (MDA), glutathione (GSH) and superoxide dismutase (SOD) when comparing the D+CCO group with respect to D (Figure 2A-C), but there was a significant decrease in the expression of *Rela* and *Gpx* in the D+CCO group with respect to D (Figure 2 E and D).

Conclusions: CCO at the dose used and during the study period was not hepatotoxic, had a hypoglycemic effect from the first dose and decreased ALT, AST and BUN levels. CCO has an anti-inflammatory effect by decreasing *Rela* gene expression.

Ethical statement

The protocol was registered and approved by the Ethics Committee.

Declaration of interests

None

Funding

This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

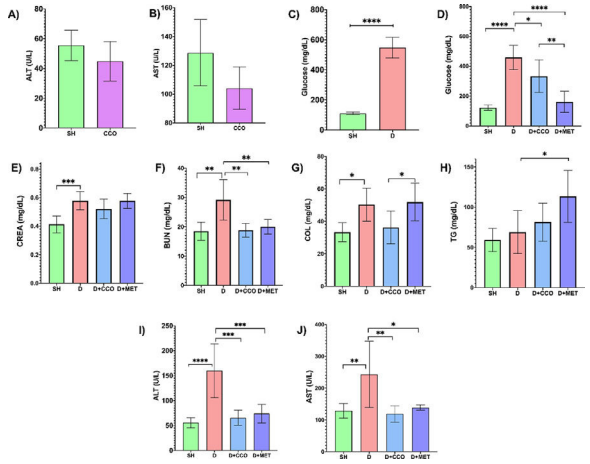

Figure 1. Results of the biochemical markers in the different study groups. (A and B) ALT and AST levels in SH groups and D. (C) Glucose levels after one week of administration with alloxane. (D-J) Glucose, CREA, BUN, COL, TG, ALT and AST levels in the various study groups after the administration period. Values are expressed as mean ± SD. P values: * p<0.05, ** p<0.01, *** p<0.001 and **** p<0.001.

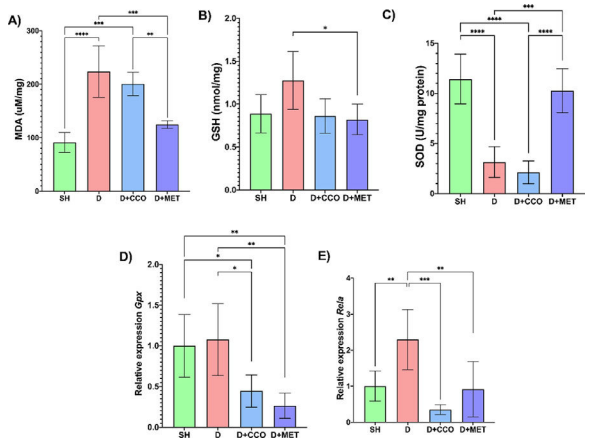

Figure 2. Results of oxidative stress and gene expression markers in the different study groups. (A, B and C) MDA, GSH and SOD oxidative stress markers. (E and D) Relative expression of *Gpx* and *Rela* in the various study groups. Values are expressed as mean ± SD. P values: * p<0.05, ** p<0.01, *** p<0.001 and **** p<0.001.

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Hepatoprotective effects of N-acetylcysteine prevents hepatocellular carcinoma development induced experimentally.

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Introduction and Objectives: Hepatocellular carcinoma (HCC) development involves imbalance of cellular processes such as oxidative stress, inflammation, fibrogenesis and cell proliferation. N-acetylcysteine (NAC) is an effective drug used clinically to treat drug-induced liver injury, but its ability to modulate molecular mechanisms activated during HCCestablishment is unknown.

This study aimed to evaluate antioxidant, antifibrogenic, and antiproliferative NAC properties in the HCC induced experimentally.

Materials and Patients: Male Fisher 344 rats divided into 3 groups: 1. Control (CTL); 2. HCC: Diethylnitrosamine (DEN) + 2-acetylaminofluorene (2-AAF). 3. HCC/NAC: DEN+2-AAF and NAC. Liver damage, oxidative stress, fibrogenesis and proliferation markers were evaluated by colorimetrics methods, Western blot, Dot blot, immunofluorescence, immunohistochemistry, respectively. H&E and Masson's Trichrome stains were also performed. This project was conducted in accordance with the guidelines of the University of Guadalajara under the approval number of the bioethics, research, and ethics research committees CI-01723.

Results: NAC exerts hepatoprotective effects, by preserving hepatic micro and macrostructure, slowing dysplastic nodules formation, and preventing an increase in ALT and GGT enzymatic activity. This drug also is able to exert anti fibrogenic effects by repressing extracellular matrix accumulation through to inhibition of α-SMA and TFG-β expression. Likewise, NAC demonstrated antiproliferative capacity by reducing Glypican-3 and Ki-67 expression.

Furthermore, NAC exerts its antioxidant effects by regulating Nrf2 signaling pathway, modulating CAT and SOD expression, and GSH levels. Finally, this drug prevents DNA oxidative damage through increasing enzyme 8-oxoguanine-DNA glycosylase (OGG1/2) expression, and therefore, reducing 8-oxoguanine (8oxoG) levels.

Conclusions: In this work, we demonstrate that NAC exerts antioxidant, antifibrogenic and antiproliferative effects useful in the prevention of the development of this disease. It is necessary to carry out additional analyzes that allow a more precise clarification of NAC hepatoprotective mechanisms, and that allow it to be repositioned as an adjuvant therapy in HCC treatment.

Ethical statement

All experimental procedures were approved by the Research Committee, the Research Ethics Committee, and the Biosafety Committee of the University of Guadalajara, with the approval number CI-01723.