

In vitro analysis of apoptosis in hepatic stellate cells cultured under steatogenic conditions

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Introduction and Objectives: Hepatic stellate cells (HSC) are responsible for the development of fibrosis during chronic liver disease. Cell death is among the most common cellular causes of increased tissue damage. From the different types of cell death, apoptosis and necrosis of hepatocytes are associated with the development of inflammation and the progress of liver disease. We aimed to assess cell death by apoptosis and necrosis in LX-2 hepatic stellate cells in an *in vitro* model of steatosis.

Materials and methods: LX-2 HSC were cultured under different conditions: control (C), mild steatosis (MS), severe steatosis (SS) and activation (TGF β). Cell death was identified by trypan blue staining. Apoptosis and necrosis were analyzed by flow cytometry at 24, 48 y 72h. Data: Mean $\pm$ SD, analyzed by one-way ANOVA, $p < 0.05$ was considered significant.

Results: Cell death was similar between HSC cultured under control or activation conditions at the different times studied. Increased cell death was observed in MS at 72h and in SS from 24h. Accordingly, percentage of apoptotic cells was significantly increased in MS at 72h compared with other conditions at that time (C72h=14.1 $\pm$ 6.1, TGF β 72h=11.8 $\pm$ 5.2, MS72h=57.7 $\pm$ 8.4, SS72h=3.8 $\pm$ 2.7 %, $p < 0.05$). In contrast, SS group showed its peak in apoptosis at 24h (C24h=27.4 $\pm$ 2.8, TGF β 24h=21.7 $\pm$ 7.5, MS24h=25.7 $\pm$ 3.2, SS72h=50.8 $\pm$ 9.5 %, $p < 0.05$), showing that apoptosis begins early at 24h but is evidenced by trypan blue up to 48h. Activation of HSC was not associated with changes in apoptosis. No differences were observed in necrosis.

Conclusions: Apoptosis in LX-2 HSC was associated with the severity of the steatogenic condition, the higher the amount of free fatty acids in the medium, the higher the mortality at short term. Apoptosis explained most of the mortality observed by trypan blue in HSC; however, other death processes, including pyroptosis or necroptosis, should not be discarded yet, since they might also contribute to HSC cell death during steatosis.

Ethical statement

The protocol was registered and approved by the Ethics Committee.

Declaration of interests

None

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Prevalence of Immunoglobulin G against Hepatitis A Virus and Hepatitis E Virus in healthcare personnel

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Introduction and Objectives: The hepatitis A and hepatitis E viruses are organisms that stand out for their high resistance to acid

and alkaline media, as well as to freezing temperatures. Despite presenting an approximate mortality of 1% for both viruses, a seroprevalence of up to 81.3% has been reported in previous decades, so it is important to know the current epidemiological status of both diseases.

Materials and Patients: Cross-sectional, descriptive, prospective and observational study. Individuals over 18 years of age were recruited, who were studying or had the degree of gastroenterologists at the Hospital de Especialidades Centro Médico Nacional Siglo XXI, in a period of time between June 01 and June 30, 2023. Blood samples were collected for detection of immunoglobulin G against hepatitis A and hepatitis E viruses and a demographic questionnaire was conducted to each of the participants.

Results: 23 individuals were recruited, 60.9% men ($n=14$) and 39.1% women ($n=9$), with a median age of 29 years, 13.0% corresponding to individuals from Mexico City ($n=3$) and 86.9% from other states of the Mexican Republic ($n=20$). A seroprevalence of 17.3% ($n=4$) and 4.3% ($n=1$) was reported for hepatitis A virus and hepatitis E virus, respectively.

Conclusions: There is a lower seroprevalence for hepatitis A and hepatitis E viruses than reported, so it is vitally important to take preventive measures in populations at risk of infection, such as health personnel.

Ethical statement

The protocol was registered and approved by the Ethics Committee. The identity of the patients is protected. Consentment was obtained.

Declaration of interests

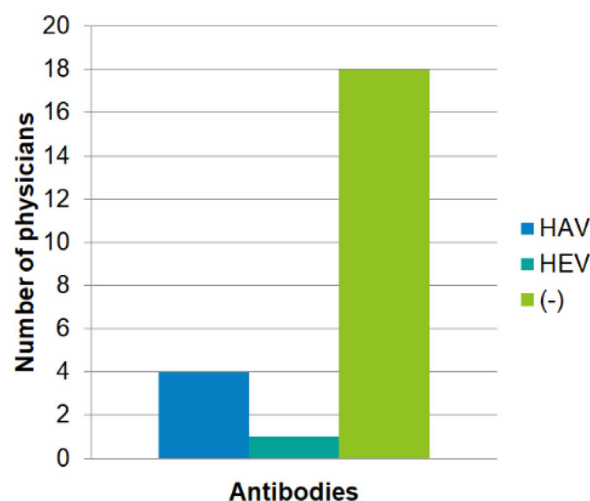
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Table 1.

Number of physicians with antibodies against hepatitis A virus (HAV), hepatitis E virus (E) or a negative test (-).



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Noninvasive markers of hepatic fibrosis and their clinical application in coronary artery disease.

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Introduction and Objectives: Coronary artery disease (CAD) is a leading cause of morbidity and mortality worldwide. Reliable knowledge of the prevalence of occult CAD, particularly anatomically confirmed CAD is limited and cardiovascular risk (CVR) models only predict the risk of an acute coronary event within a set period. It has been described that a FIB-4 score is associated with a higher CVR. Determine what is the utility of noninvasive markers of liver fibrosis in CAD.

Materials and Patients: A cross-sectional study was conducted in two tertiary centers in central and western Mexico from March 2019 to April 2023. Patients who required percutaneous coronary angiography were studied and demographic data and coronary angiographic were recorded. Noninvasive fibrosis indexes were calculated. Continuous variables were subjected to a distribution analysis and equality of variances to subsequently perform a mean comparison analysis with U-Mann-Whitney test between patients with monovascular, bivascular and trivascular involvement. A correlation analysis was also performed between the invasive markers and the Syntax index.

Results: A total of 168 patients were included with a mean age of 66 ± 12 years with a predominance of male sex with 75.6% (n= 127). Angiographic findings included 37.5%, monovascular, 32.7%, bivascular and 29.8% trivascular involvement. Comparison of means of non-invasive markers of fibrosis demonstrated a significant difference in HFS between patients with monovascular (0.17 ± 0.18), bivascular (0.27 ± 0.18) and trivascular (0.30 ± 0.25) coronary artery disease, $p < 0.001$. A correlation was also demonstrated between non-invasive markers and Syntax score: FIB-4 ($r = 0.820$, $p < 0.001$), APRI ($r = 0.766$, $p < 0.001$), HFS ($r = 0.869$, $p < 0.001$), ($r = 0.820$, $p < 0.001$), NFS ($r = 0.807$, $p < 0.001$)

Conclusions: The score of noninvasive tools to assess liver fibrosis correlates positively with the complexity of CAD and could be considered as noninvasive tools to be used in the assessment CVR.

Ethical statement

The protocol was registered and approved by the Ethics Committee. The identity of the patients is protected. Consentment was obtained.

Declaration of interests

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Correlation between steatosis and fibrosis in patients with metabolic syndrome

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Introduction and Objectives: MAFLD is a highly prevalent cause of chronic liver disease, present in 70% of overweight people, 70% of diabetics, and 90% of morbidly obese people. It is the hepatic manifestation of the metabolic syndrome, defined by the presence of central obesity, insulin resistance, hyperlipidemia, hyperglycemia, and hypertension. The development of liver fibrosis is secondary to several factors, steatosis being one of them. To evaluate the correlation of steatosis with hepatic fibrosis in patients with metabolic syndrome using transition elastography.

Materials and Patients: Patients older than 18 years who met MALFD criteria were included, transition elastography was performed to calculate CAP and kilopascals, steatosis degree and fibrosis degree were calculated according to the myfibrosan application, for statistical analysis Pearson's bivariate correlations were used between CAP and kilopascal values. The association between the degree of steatosis and fibrosis was performed using the chi-square test. Was considered significant at $p < 0.05$.

Results: 94 patients were included, 20 men (21.3%), 74 women (78.7%), mean age 40.5 ± 10.02 , CAP 300.6 ± 63.4 , kilopascals 6.4 ± 2.7 , steatosis grade S0: 8, S1: 8, S2: 20, S3: 58, degree of fibrosis F0: 58, F1: 14, F2:14, F3: 6, F4:2. The correlation between CAP and kilopascals was moderate and significant $RHO=0.343$ $P=0.001$. A significant association was found between the degree of steatosis and that of fibrosis chi-square (12) =25.1, $p=0.015$. The proportions were 50% (S0:F0), 16% (S1:F3), 50% (S2:F3), 100% (S3:F4).

Conclusions: The correlation between steatosis and fibrosis is moderate, implying that there are other factors that influence the development of fibrosis and its progression, so metabolic control and other factors in patients with MALFD are highly relevant to prevent fibrosis progression.

Ethical statement

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Acute liver failure secondary to co-infection of Hepatitis B and HIV

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Introduction and Objectives: We present the case of a man with hepatitis B and HIV coinfection diagnosed by serological studies which presented acute liver failure; the diagnostic approach, its treatment and outcome are described.

Materials and Patients: Provide information on the association of hepatitis B virus and HIV. When hepatotropic viruses are identified, intentionally find the association with other factors that cause acute