

## Contributions to the study of pathogenic mechanisms of Hepatitis C Virus.

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Understanding the mechanisms of hepatitis C virus (HCV) infection and pathogenesis is an important part of HCV research [1]. Detection and localization of HCV components in the target cells is important to study the host-viral interactions at the cellular level [2]. In this study, the expression of HCV components in liver biopsies and peripheral blood mononuclear cells (PBMC) from patients seropositive for HCV antigens but negative for HCV-RNA in serum was examined. Samples were analyzed by an in situ hybridization (ISH) assay using specific probes for detection of HCV-RNA of negative and positive polarity. In addition, an immunofluorescence assay using antibodies specific for HCV core (HCcAg) antigen (anti-HCcAg) was performed. Besides, the expression of HCcAg was analyzed in the liver of 150 HCV-infected patients using electron microscopy studies. Samples of liver biopsies were fixed in 0.1% glutaraldehyde and 4% paraformaldehyde in 0.1% OsO<sub>4</sub> in the same buffer, dehydrated in ethanol, embedded in lowicryl. Ultrathin sections were incubated with anti-HCcAg antibodies. The ultrathin sections were examined with a JEOL JEM 2000EX Transmission Electron Microscope.

The findings show that HCV-RNA of both and negative polarity was carried in the hepatocytes of more than 80% of cases. No hybridization signals were observed in either the liver biopsy specimens from the negative control subjects. In addition, reaction products suggestive of HCV core in the cytoplasm and nucleus of some hepatocytes were observed by immunofluorescence. No hybridization signals were observed in the liver biopsy specimens from the control subjects. 120 out of 150 patients with apparent clinical and histological resolution of hepatitis, contain HCV-RNA in either liver or PBMC. The intermediate replicative form of the HCV genome can persist in hepatocytes and PBMC after apparently complete resolution of chronic hepatitis C.

On the other hand, immunostaining with anti-HCcAg antibodies revealed the presence of this protein in different liver cell types such as lymphocytes, Kupffer, pit, endothelial, stellate, and fibroblast-like cells. Interestingly, HCcAg was immunolabeled not only in the cytoplasm but also in the nucleus of these cells. Remarkably, HCcAg co-localized with large lipid droplets present in stellate cells and with collagen fibers in the extracellular matrix. Data suggest that nonparenchymal liver cells may constitute a reservoir for HCV replication. Besides, HCcAg may contribute to modulate immune function and fibrosis in the liver as well as steatosis.

### Referencias

[1] Radkowski, M., Gallegos-Orozco, J.F, Hepatology, 41 (2005) 106-114.

[2] M Comar, G Dal Molin, Journal of Clinical Pathology, 59 (2006) 526-529.