EXPRESSION OF THE IRF1 PROTEIN IN PATIENTS INFECTED WITH THE HEPATITIS C VIRUS IN TREATMENT WITH ALPHA INTERFERON PLUS RIBAVIRIN.

Jacqueline Monroy Granados, Ma. de la Luz Martínez Rodríguez, Florencia del Carmen Salinas Pérez, María del Socorro Ruiz Palma. Unidad de Investigación Médica en Inmunología e Infectología del Hospital de Infectología, CMN "La Raza" México, D.F. CP 02990
socorroruizpalma@yahoo.com.mx, Maria del Socorro Ruiz P

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Introduction. The hepatitis C virus infects aproximately 170 million people around the world, representing a viral pandemic.[1] In the treatment for HCV an interferon α 2a or 2b plus ribavirin are used for 24 to 48 weeks depending on the genotype.[2] This treatment realizes its function by the regulatory Factor of the IFN 1 (IRF1) and by the efector mechanisms of the antiviral response. The presence of this protein in lymphoid infected cells with HCV indicates that the treatment is efficient and this can distinguish responder patients from nonresponders.

The aim of this study was to determine the IRF1 expression protein in patients with the hepatitis C virus, responders and nonresponders treated with interferon alpha and ribavirin.

Methods. Samples of peripheral blood of patients with hepatitis C at 2nd, 5th, 8th and 12th week of treatment were taken. The samples were processed to obtain lymphocytes from the Ficoll technique, with the lymphocytes obtained various techniques were standardized including the concentration of polyacrylamide gels, the concentration of protein loaded on the gel, the electrophoresis and Western blot technique. When the techniques were standardized, quantification of proteins of a sample from a patient responder was performed by BCA method, the concentration of protein was adjusted to 250 µg, subsequently an electrophoresis on polyacrylamide gels at 12% was performed and the Western blot technique was performed too.

Results. Different concentrations of polyacrylamide gels were tested, observing that bands to 12% were seen with a better resolution. The suitable concentration of protein to load on polyacrylamide gels was 250 µg and the preparation of the sample at a 5:1 dilution. Besides of that, the running of the electrophoresis was more appropriate to an initial voltage of 80 mV/20 min and subsequently voltage of 120 mV/120 min.
After the technique of Western blott was performed with a responder patient sample, the presence of the IRF1 protein was observed with a molecular weight of 55 kDa (Figure 1) to the second week of treatment at a dilution of the primary antibody to 1:500 and the second antibody at 1:2000; because to a 1:200 dilution a nonspecific band was located.

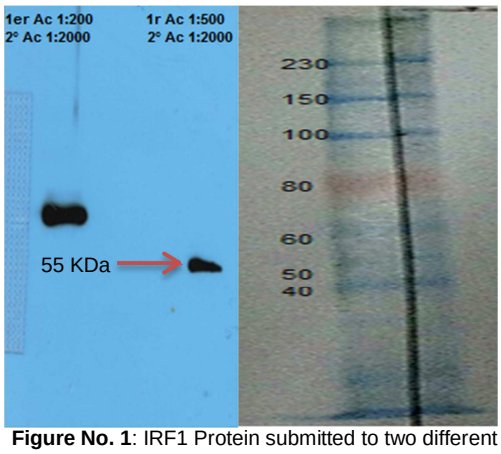

Figure No. 1: IRF1 Protein submitted to two different concentrations of 1st Ab (1:200 and 1:500) in the second week of treatment.

Conclusions. IRF1 protein is present at the second week of treatment in patients with hepatitis C, although it is necessary to run all samples at different times to determine the kinetics of protein synthesis and compare it between responders and nonresponders patients.

Referencias.

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