

IN VIVO DOWN REGULATION OF HIV REPLICATION AFTER HEPATITIS C SUPERINFECTION

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Abstract There are increasing molecular and clinical evidences that the effects of human immunodeficiency virus (HIV) infection can be modified by coinfection with other viruses. The objective was to investigate the viral interaction between HIV and hepatitis C virus (HCV) after HCV superinfection. A 16 year-old pregnant woman was evaluated because of icteric acute hepatitis. Admission laboratory tests showed the following results: ALT 877 IU/L; AST 1822 IU/L; bilirubin 6.79 mg/dl. Diagnosis of acute HCV was based on detection of serum HCV RNA by PCR and anti-HCV seroconversion. ELISA for anti HIV testing was positive and confirmed by western blot. Serum markers for other viruses were negative. The patient was followed during 19 months; serum samples were taken monthly during this period for detection of plasma HIV and HCV RNA. Levels of plasma HIV-RNA were positive in all samples tested before and after the onset of acute hepatitis C. Six months later and a for two month period, and 13 months later for a period of one month HIV viremia was undetectable; then HIV-RNA in plasma was detectable again. In conclusion, HCV superinfection may have temporarily interfered with HIV replication in our patient. The following observations support our hypothesis: it has been demonstrated that HIV-1 replication is suppressed by HCV core protein which has transcriptional regulation properties of several viral and cellular promoters. Clinical implications of this event are not generally known and the interaction between these two viruses in dual infections is worth considering.

Resumen *Inhibición de la replicación de HIV luego de una superinfección por el virus de la hepatitis C.*

Existen evidencias clínicas y moleculares que muestran que el curso de la infección por el virus de la inmunodeficiencia humana (HIV) puede ser modificado por la coinfección con otros virus. El objetivo del presente estudio fue investigar el efecto de la interacción viral entre el HIV y el virus de la hepatitis C (HCV) después de una superinfección por HCV. Una mujer de 16 años embarazada fue evaluada por presentar hepatitis aguda e ictericia. El laboratorio de admisión mostró: ALT 877 UI/L; AST 1822 y bilirrubina de 6.79 mg/dl. Se detectó en suero la presencia del ARN del HCV por PCR y seroconversión de anti-HCV por lo que se diagnosticó hepatitis aguda por virus C. Los marcadores virales para otros virus hepatotrópicos fueron negativos. Un ELISA fue positivo para anti-HIV y confirmado por Western blott. La paciente fue seguida durante 19 meses con muestras de sangre tomadas mensualmente para la detección plasmática del ARN del HIV y HCV. Los niveles del ARN-HIV fueron positivos en todas las muestras estudiadas antes y después del inicio de la hepatitis C. Seis meses después y por un lapso de 2 meses, y 13 meses después y por un lapso de un mes la viremia del HIV fue indetectable. En conclusión la superinfección por el HCV pudo haber interferido temporariamente en la replicación del HIV. Las implicancias clínicas de este evento se desconocen, sin embargo, la interacción entre estos virus debería ser considerada en futuros estudios virológicos y moleculares.

Key words: acute hepatitis C, HIV-HCV coinfection, HIV replication, viral interference, HCV core protein, down regulation HIV replication

There are increasing molecular and clinical evidences that the effects of human immunodeficiency virus (HIV) infection can be modified by coinfection with other viruses^{1,2}. HIV and hepatitis C virus (HCV) have similar parenteral routes of transmission including intravenous drug abuse and exposure to blood products³. This common epidemiology explains the high frequency of combined infection by both viruses. However, the influence of

HCV infection on HIV-seropositive patients is unclear. In the present study, we report the case of an HIV infected patient whose plasma levels of HIV-RNA became temporarily undetectable after HCV superinfection.

A 16 year-old pregnant woman was referred to the Liver Unit in April 1997 because of acute hepatitis-like illness, with jaundice, nausea, abdominal pain, fever, diarrhea, itching and elevated serum alanine amino-transferase (ALT) and aspartate amino transferase (AST) levels. She was at 30 weeks gestation and her prenatal course had been uneventful until this episode. She had a history of intranasal cocaine use since she was 14 years old, sharing devices to inhale the drug and had an intra-

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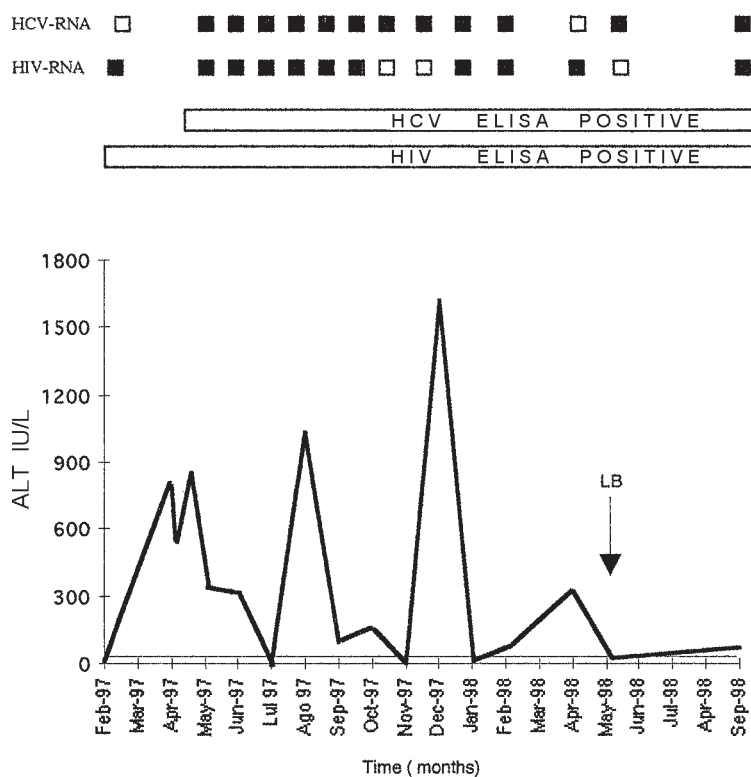


Fig. 1.— Virological, serological and biochemical profile of HIV/HCV coinfecting patient. LB: Liver Biopsy: ■ Detectable RNA level, □ Undetectable RNA level.

venous drug user (IVDU) sexual partner. There was no history of blood transfusion, hepatotoxic drugs exposure or other hepatocellular injuries. Liver function tests performed in February 1997 during routine ante natal care showed normal ALT (19 IU/L), AST (23 IU/L) and bilirubin (0.28 mg/dl) level. Serum sample collected in March 1997 was retrospectively tested for HCV antibodies (ELISA-2, Abbot Laboratories, Chicago, USA) being negative. HIV antibodies testing by ELISA was positive in this sample and the result was confirmed by western blot. At the onset of the hepatitis symptoms in April 1997, admission laboratory tests showed the following results: ALT 877 IU/L (normal value < 40); AST 1822 (normal value < 37 IU/L); bilirubin: 6.79 mg/dl and hemoglobin 10.2 g/dl. Serum markers for hepatitis B virus revealed a pattern of resolved HBV infection (HBsAg negative, anti-HBc positive and anti HBs positive) and for hepatitis A, IgM antibodies were negative. Testing of anti-HCV was positive indicating seroconversion, and diagnosis of acute hepatitis C was established.

HCV-RNA was detected in serum concomitantly with the increase of ALT (Figure 1) by polymerase chain reaction (PCR) with a set of primers derived from the highly conserved 5'non-coding region of the HCV genome⁴. The serum sample collected from the patient in March 1997 was retrospectively tested for HCV RNA being undetectable. All serological markers were determined

by commercially available ELISA tests according to the manufacturer's instructions (Abbott Laboratories, Chicago, USA). Non-organ-specific autoantibodies (anti smooth muscle, antinuclear and anti liver-kidney microsomal) were negative. Abdominal ultrasound examination was normal.

CD4 lymphocyte subsets were measured by flow cytometry and the results were 360 and 240 cells per cubic millimeter in July 1997 and September 1998 respectively. After resolution of the jaundice she had intermittent elevation of ALT levels (see Figure 1). Fifteen months after the initial episode of hepatitis, liver biopsy was performed and showed chronic hepatitis, total Knodell score 5, and no fibrosis was found.

The patient was followed during 19 months from March 1997 to September 1998; serum samples were taken monthly during this period for detection of plasma HIV and HCV RNA. Plasma HIV-1 RNA was assayed by nested PCR using a set of primers from the polymerase region gene⁵. All plasma samples taken previous and after the onset of acute hepatitis C and until October 1997 were HIV-1RNA positive. In November 1997 and for to months period and in May 1998 for a period of one month HIV viremia was undetectable; then HIV-RNA in plasma returned detectable again (see Figure 1). No antiretroviral therapy was implemented during the study period because the patient denied to be treated.

At all times, informed consent was obtained from the patient and the study conforms to the ethical guidelines of 1975 Declaration of Helsinki and was approved by a local ethics committee.

Discussion

The interference among viruses is a well-documented biological phenomenon whereby one virus particle can restrict the replication of another⁶. It has been demonstrated that patients with HIV infection are frequently coinfecting with other viruses, including hepatitis viruses. Moreover, there is strong evidence that biological interaction between two viruses in the same host could modify the clinical course or transmissibility of viral infection³. Both HCV and HIV are transmitted by parenteral routes, and coinfection with these two viruses is common among patients with a history of IVDU or transfusion. The virological findings of the present study showed that there was transitory clearance from plasma of HIV-RNA after HCV superinfection in a patient previously infected by HIV. Therefore, we postulate that HCV superinfection may have temporarily interfered with HIV replication in our patient. The following observations support our hypothesis: a) it has been experimentally demonstrated *in vitro* that HIV-1 replication is suppressed by HCV core protein which might act as a strong repressor of the long terminal repeat of HIV-1⁷, b) the HCV core protein is a major component of viral nucleocapsid with transcriptional regulation properties of several viral and cellular promoters⁸ and c) it has been found that HCV superinfection in a patient with chronic HBV infection exerted a viral interference that was followed by spontaneous seroconversion and termination of the HBV carrier state⁹.

We cannot rule out the possibility of a spontaneous decrease of HIV viral load below the detection level of our PCR. However, HIV RNA clearance seems to be induced by HCV superinfection since plasma HIV viral load in untreated patients vary from 10^2 to 10^6 copies/ml and the sensitivity of the HIV-PCR used in our study was 10 copies/ml^{5, 10, 11}.

Noteworthy, despite the high frequency of this viral association, the role of HCV in modulating HIV replication has not been studied extensively. Different facts may give an explanation for this lack of information. First, the transitoriness of this event would require a close follow-up of virological assays in order to be evidenced. Second, this phenomenon might only occur during the first stages of HCV infection when acute HCV infection is usually subclinical and any change in HIV viral load is not suspected of being produced by HCV superinfection.

One may speculate that viruses that interfere with HIV replication should infect the same cells¹. HCV, like HBV, is regarded as predominantly hepatotropic, neverthe-

less HCV related RNA has also been detected in lymphoid cells from the bone marrow and peripheral T cells, indicating that HCV is also lymphotropic¹².

Alternatively, immunological host responses triggered by HCV superinfection might influence the viral dynamics of HIV replication. In line with this possibility is the observation that HIV replication seems to be influenced by cytokines¹³.

To our knowledge, this is the first report about *in vivo* down regulation of HIV-1 replication in an infected human by HCV superinfection. The clinical implications of this event are not known; however, the interaction between these two viruses in dual infection is worth considering. For that purpose, we are currently performing in our patient a thorough virological and immunological characterization of HIV-HCV interaction at the molecular level.

References

1. Ho WZ, Harouse JM, Rando RF, Gonczol E, Srinivasan A, Plotkin SA. Reciprocal enhancement of gene expression and viral replication between human CMV and HIV-1. *J Gen Virol* 1990; 71: 97-103.
2. Gendelman HE, Phelps W, Feigenbaum L, et al. Transactivation of the human immunodeficiency virus long terminal repeat sequence by DNA viruses. *Proc Natl Acad Sci USA* 1986; 83: 9759-63.
3. McNair AN, Main J, Thomas HC. Interactions of the human immunodeficiency virus and hepatotropic viruses. *Semin Liver Dis* 1992; 12: 188-96.
4. Abate ML, Manzini P, Negro F, et al. Detection of hepatitis C virus RNA by reverse transcriptase and polymerase chain reaction: clinical applications of quantitative analysis. *Clin Diag. Vir.* 1994; 1: 289-97.
5. Vandamme AM, Fransen K, Debaisieux L, et al. Standardisation of primers and an algorithm for HIV-1 diagnostic PCR evaluated in patients harbouring strains of diverse geographical origin. *Clin Diag Vir* 1995; 51: 305-16.
6. Csatory LK, Romvary JJ, Kasza L, Schaff S, Massey JR. *In vivo* interference between pathogenic and non-pathogenic viruses. *J Med* 1985; 16: 563-73.
7. Srinivas RV, Ray RB, Meyer K, Ray R. Hepatitis C virus core protein inhibits human immunodeficiency virus type 1 replication. *Virus Res* 1996; 45: 87-92.
8. Ray RB, Lagging LM, Meyer K, Steele R, Ray R. Transcriptional regulation of cellular and viral promoters by the hepatitis C virus core protein. *Virus Res* 1995; 37: 209-20.
9. Liaw YF, Lin SM, Sheem IS, Chiu CM. Acute hepatitis C superinfection followed by spontaneous HBeAg seroconversion and HBsAg elimination. *Infection* 1991; 250: 54-5.
10. Havlir DV, Richman DD. Viral dynamics of HIV: Implications for drug development and therapeutic strategies. *Ann Intern Med* 1996; 12: 948-94.
11. Saag MS, Holodny M, Kuritzkes DR. HIV viral load markers in clinical practice. *Nature Medicine* 1996; 2 (6): 625-9.
12. Cohen OJ, Kinter, Fauci AS. Host factors in the pathogenesis of HIV disease. *Immunol Rev* 1997; 159: 31-48.
13. Zignego AL, Macchia D, Monti M, et al. Infection of peripheral mononuclear cells by hepatitis C virus. *J Hepatol* 1992; 15: 382-86.