Pediatr Nephrol (2004) 19:547–549
DOI 10.1007/s00467-003-1405-9

BRIEF REPORT

Douglas M. Silverstein · Diego H. Aviles · V. Matti Vehaskari

False-positive human immunodeficiency virus antibody test in a dialysis patient

Received: 30 September 2003 / Revised: 10 December 2003 / Accepted: 11 December 2003 / Published online: 26 February 2004

Abstract A patient developed end-stage renal disease secondary to p-anti-neutrophil cytoplasmic antibody (p-ANCA) positive rapidly progressive glomerulonephritis. He subsequently had human immunodeficiency virus (HIV)-1 antibody screening performed as part of a pre-transplant evaluation. The HIV-1 enzyme immunoassay (EIA) antibody test was repeatedly reactive. The HIV-1 western blot was indeterminate. The western blot pattern revealed "non-specific staining obscuring bands in that region." Another sample of serum was sent and the results were identical to the first result. An HIV-1 proviral qualitative polymerase chain reaction test was then performed several months later and no HIV-1 DNA was detected. One year later, an HIV-1 RNA test was negative. Thus, the positive antibody EIA test and the indeterminate western blot represent a false-positive result, most likely due to cross-reacting antigens in the patient's serum with various HIV antibodies. Throughout this period and thereafter, the patient has exhibited no symptoms of HIV infection.

Keywords End-stage renal disease · Human immunodeficiency virus · Enzyme immunoassay · Western blot · Polymerase chain reaction

D. M. Silverstein · D. H. Aviles · V. M. Vehaskari
Louisiana State University Health Sciences Center,
New Orleans, LA 70112, USA

D. M. Silverstein (✉)
Department of Pediatrics,
Louisiana State University Health Sciences Center,
Research and Education Building, Suite 4241,
Children's Hospital,
200 Henry Clay Avenue, New Orleans, LA 70118, USA
e-mail: dsilve@lsuhsc.edu
Tel.: +1-504-8929238
Fax: +1-504-8929240

Introduction

All patients who are eligible for renal transplantation must have screening for human immunodeficiency virus (HIV)-1 performed. This is generally accomplished by screening the serum of the patient with an enzyme immunoassay (EIA) test for HIV-1. It has been estimated that up to 0.5% of seropositive patients have false-positive results for HIV-1 [1]. The standard test performed in patients in whom a false-positive EIA result is suspected is western blot analysis. Alarmingly, up to 20% of patients with false-positive EIA tests for HIV-1 will have an indeterminate or inconclusive pattern on western blot analysis [1]. If this occurs, it is now possible to test for the HIV-1 DNA and RNA by performing a polymerase chain reaction (PCR) test of the patient's serum [2].

There are various patients who may have a false-positive HIV-1 result by EIA and/or an indeterminate western blot result, including those who have received blood products or who have circulating antibodies related to another condition such as an autoimmune disease [3]. Indeed, circulating immune complexes have been detected in adult patients receiving hemodialysis [4]. In addition, autoantibodies to red blood cells, cytokines, and phospholipids have been detected in hemodialysis patients [5, 6, 7]. In one report of 520 adult hemodialysis patients, 9% were EIA positive, and half of those had indeterminate western blot results, suggestive of false-positive reactions. A 5-year follow-up of those with indeterminate western blot results revealed that none became western blot positive for HIV-1 or HIV-2 [8]. Finally, there was one report of a false-positive HIV antibody test following therapy with alpha-interferon in a patient receiving chronic hemodialysis [9], indicating that various conditions may result in false-positive HIV EIA results.

We report an adolescent with end-stage renal disease (ESRD) whose HIV EIA antibody test was repeatedly reactive, although the western blot was inconclusive.

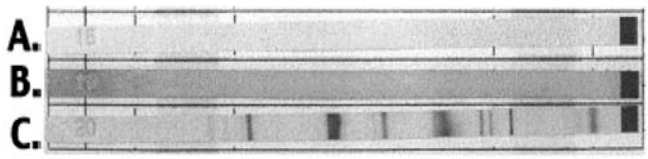

Fig. 1 Representative western blot for human immunodeficiency virus. *Lane A* represents a negative result. *Lane B* represents an indeterminate result because of the presence of diffuse, non-specific staining. *Lane C* represents a true positive result because of the presence of specific staining for several bands. Reprinted with permission from C.M. Litwin (from reference [15])

Case summary

The patient is an 18-year-old male who originally presented to our service in July 2000 with hematuria and proteinuria detected on a routine physical examination. He was asymptomatic at that time. His vital signs revealed a normal blood pressure and no other abnormalities. His physical examination was unremarkable. A random urine protein/creatinine ratio was 1.36. His serum creatinine was 1.6 mg/dl, with an estimated glomerular filtration rate of 75.5 ml/min per 1.73 m^2. Light microscopy of a renal biopsy revealed four crescents (two cellular, one fibrous, and one fibrocellular) in 19 total glomeruli. Several glomeruli showed fibrinoid necrosis. The tubules showed some areas with resorption granules and the interstitium revealed a mild mononuclear infiltrate. Immunofluorescence revealed mild staining for IgM (1+). Electron microscopy showed numerous foam cells, probably of visceral epithelial origin, with many lysosomes, occasional endothelial swelling, and focal foot process effacement. P-anti-neutrophil cytoplasmic antibody (ANCA) and myeloperoxidase antibody (77.5 EU/ml, normal <5.0 EU/ml) were both positive. This was all consistent with a pauci-immune rapidly progressive glomerulonephritis (RPGN), and the patient received high-dose intravenous methylprednisolone followed by intravenous cyclophosphamide. His renal function rapidly deteriorated and he developed ESRD.

In February 2001, he sustained a moderate gastrointestinal bleed and received a blood transfusion. He began receiving hemodialysis in May 2001. As part of a pre-transplant evaluation he was tested for HIV-1 antibodies. The result by EIA was repeatedly reactive, although the western blot result for various antigens, including p18, p24, p31, p40, gp41, p51/55, p65, gp120, and gp160, revealed "non-specific staining obscuring bands in that region" (Fig. 1). The patient was asymptomatic at that time. A repeat EIA antibody and western blot analysis 1 month later yielded similar results. Two months thereafter, a PCR test revealed that no HIV-1 DNA was detectable. An HIV-1 PCR RNA test was performed 1 year later and was negative.

Discussion

This report demonstrates that patients receiving hemodialysis may have concomitant conditions that may alter the normally reliable HIV-1 EIA and western blot tests. This patient had a positive EIA antibody test and an indeterminate western blot analysis for multiple antigens. Specifically, the indeterminate western blot result was reported to reveal "non-specific staining obscuring bands in that region." Subsequent PCR tests revealed that no HIV-1 DNA or RNA could be detected in the patient's serum, effectively ruling out the possibility of HIV-1 positivity.

It is important to briefly review the current tests available to detect HIV-1 in patients′ serum. The standard tests used in most laboratories are the EIA and western blot tests. These tests detect IgG antibodies to several antigens in the serum of HIV-1-infected patients, most commonly gp160, gp41, and RT (or p66) [10]. Traditionally, the detection of other HIV-1 antigens (gag proteins) in HIV-1-infected individuals, such as p24 and p17, was less certain; however, recent advances in EIA testing have improved the detection of these antigens [11].

Most of the data regarding the time period for seroconversion (window) have been generated from studies of patients who were infected by HIV-1 from blood transfusions. These studies show that the window for the antigens is generally between 5 and 8 weeks [12]. The advent of HIV-1 RNA and DNA (PCR) testing has provided an adjunct for the detection of HIV-1 infection. The window for detection of the HIV-1 RNA or DNA is narrower than that for antigen detection. Murthy et al. [13] studied the window for detection of HIV-1 RNA, HIV-1 DNA, and HIV-1 antigens in chimpanzees inoculated with HIV-1. They reported that the window for detection of HIV-1 RNA or DNA was about 5 weeks, whereas anti-HIV and HIV p24 antigen were not detected until 8 weeks.

In the study by Vardinon et al. [8] of 46 adult hemodialysis patients who were EIA positive with no signs of clinical disease, the western blot was negative in 22 patients, "indeterminate" in 23 patients, and positive in 1 patient. Their description of the western blot results as indeterminate was based upon the appearance of bands on western blot representing several antigens, including p24, p31, p55, and p66, two (p24 and p66) of which are generally detected in "true" positive HIV patients. Thus, we believe that their patients had a false-positive, not an indeterminate result. In contrast, we used the term "indeterminate" to describe the western blot result in our patient, since no specific band could be detected, although the result clearly could not be determined to be negative or positive (Fig. 1). We thus described the western blot in our patient as indeterminate, according to Willman et al. [14, 15]. Although differences in clinical implication between the terms "false-positive" and "indeterminate" may not be apparent, these are nevertheless distinct variations of a negative or positive result. It is also important to note that Vardinon et al. [8] concluded that 45 of 46 of their patients had a false-positive HIV EIA test based upon the lack of conversion of the western blot to a true positive result with subsequent re-testing, but not on the basis of a negative HIV PCR test. We believe that the PCR test, which was repeatedly negative in our patient, is more conclusive and confirmatory of a false-positive EIA test. An alternative to HIV EIA testing would be to perform a rapid p24 antigen assay (Elecsys HIV Ag test) that has been shown to be more sensitive than EIA and shows high specificity in combination with neutralization assays [16].

The cause of "indeterminate" HIV western blot results was recently explored by Willman et al. [14, 15]. In their studies, they used a multiplex immunoassay to simultaneously measure IgG antibodies to animal IgG and bovine serum albumin in patients with a history of indeterminate HIV western blot results. Heterophil antibodies were detected in about 50% of these patients. Furthermore, preabsorption with animal IgG removed heterophil antibodies, as detected by the multiplex immunoassay, and also reversed the false-positive EIA and inconclusive western blot results in some patients, suggesting that heterophil antibodies are the cause of some inconclusive HIV test results [14, 15].

Our patient had a pauci-immune RPGN. His serum revealed p-ANCA and myeloperoxidase, all consistent with p-ANCA-associated RPGN. The false-positive EIA and indeterminate western blot results may have been due to circulating p-ANCA and/or myeloperoxidase, although to our knowledge this has not been reported. Alternatively, he may have developed circulating cross-reactive antibodies from the blood transfusion he received. It is likely that the cause of the false-positive result may never be identified with certainty. However, this case highlights the necessity of scrutinizing all HIV-1 EIA results in dialysis patients and confirming any positive result by PCR.

References

1. MacDonald KL, Jackson JB, Bowman RJ, Polesky HF, Rhame FS, Balfour HH Jr, Osterholm MT (1989) Performance characteristics of serologic tests for human immunodeficiency virus type 1 (HIV-1) antibody among Minnesota blood donors. Public health and clinical implications. Ann Intern Med 110:617–621
2. Chen MY, Lee KL, Hung CC, Chuang CY, Chou MJ (1997) Strategies for diagnosing HIV-1 infection in atypical Western blots. Microbiol Immunol Infect 30:135–144
3. Ranki A, Kurki P, Riepponen S, Stephansson E (1992) Antibodies to retroviral proteins in autoimmune connective tissue disease. Relation to clinical manifestations and ribonucleoprotein autoantibodies. Arthritis Rheum 35:1483–1491
4. Perez GO, Glasson P, Favre H, Wauters JP, Benzonana G, Jeannet M, Lambert PH (1984) Circulating immune complexes in regularly dialyzed patients with chronic renal failure. Am J Nephrol 4:215–221
5. Katoh S, Ida K, Mizunoya A (1991) Irregular isoantibodies in the sera of patients undergoing chronic hemodialysis. Int J Artif Organs 14:136–140
6. Chew SL, Lins RL, Daelemans R, Zachee P, De Clerck LS, Vermylen J (1992) Are antiphospholipid antibodies clinically relevant in dialysis patients? Nephrol Dial Transplant 7:1194–1198
7. Sunder-Plassmann G, Sedlacek PL, Sunder-Plassmann R, Derfler K, Swoboda K, Fabrizii V, Hirschl MM, Balcke P (1991) Anti-interleukin-1 alpha autoantibodies in hemodialysis patients. Kidney Int 40:787–791
8. Vardinon N, Tust I, Katz O, Iaina A, Katzir Z, Modai D, Burke M (1999) Anti-HIV indeterminate western blot in dialysis patients: a long-term follow up. Am J Kidney Dis 34:146–149
9. Sungur C, Akpolat T, Ozkuymcu C, Yasavul U, Turgan C, Caglar S (1994) False-positive HIV antibody test following alpha-interferon therapy in a chronic hemodialysis patient. Nephron 67:251
10. Hashida S, Hashinaka K, Nishikata I, Oka S, Shimada K, Saito A, Takamizawa A, Shinagawa H, Yano S, Kojima H, Izumi T, Ishikawa E (1995) Immune complex transfer enzyme immunoassay that is more sensitive and specific than western blotting for detection of antibody immunoglobulin G to human immunodeficiency virus type 1 in serum with recombinant pol and gag proteins as antigens. Clin Diagn Lab Immunol 2:535–541
11. Hashida S, Ishikawa S, Hashinaka K, Nishikata I, Oka S, Ishikawa E (2000) Earlier detection of human immunodeficiency virus type 1 p24 antigen and immunoglobulin G and M antibodies to p17 antigen in seroconversion serum panels by immune complex transfer enzyme immunoassays. Clin Diagn Lab Immunol 7:872–881
12. Lackritz EM, Satten GA, Aberle-Grasse J, Dodd RY, Raimondi VP, Janssen RS, Lewis WF, Notari EP 4th, Petersen LR (1995) Estimated risk of transmission of the human immunodeficiency virus by screened blood in the United States. N Engl J Med 333:1721–1725
13. Murthy KK, Henrard DR, Eichberg JW, Cobb KE, Busch MP, Allain JP, Alter HJ (1999) Redefining the HIV-infectious window period in the chimpanzee model: evidence to suggest that viral nucleic acid testing can prevent blood-borne transmission. Transfusion 39:688–693
14. Willman JH, Hill HR, Martins TB, Jaskowski TD, Ashwood ER, Litwin CM (2001) Multiplex analysis of heterophil antibodies in patients with indeterminate HIV immunoassay results. Am J Clin Pathol 115:764–769
15. Willman JH, Martins TB, Jaskowski TD, Hill HR, Litwin CM (1999) Heterophile antibodies to bovine and caprine proteins causing false-positive human immunodeficiency virus type 1 and other enzyme-linked immunosorbent assay results. Clin Diagn Lab Immunol 6:615–616
16. Weber B, Muhlbacher A, Michl U, Paggi G, Bossi V, Sargento C, Camacho R, Fall EH, Berger A, Schmitt U, Melchior W (1999) Multicenter evaluation of a new rapid automated human immunodeficiency virus antigen detection assay. J Virol Methods 78:61–70