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Case report

DOES TESTOSTERONE INTAKE AFFECT DIAGNOSING PRIMARY HIV INFECTION?

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ABSTRACT

Diagnosis of primary HIV infection may be disrupted by many factors, including drugs such as exogenous steroids. We report a rare case of a 37-year-old male patient undergoing testosterone treatment due to Klinefelter syndrome. He presented with the general symptoms of fever, weakness and diarrhea lasting for 2 weeks. He had also been hospitalized twice before he was diagnosed with HIV. The fourth generation ELISA HIV test was negative. The HIV real time polymerase chain reaction test was performed and showed very high plasma viral load, over 10^7 copies/ml. We discuss the connection between the androgen replacement therapy used in Klinefelter syndrome and the diagnosis of PHI. This case report illustrates the importance of obtaining a detailed medical history, especially of chronic diseases and medications, and applying appropriate diagnostic tests.

KEYWORDS: Exogenous steroids; HIV diagnostics; Klinefelter syndrome; Primary HIV infection; Testosterone.

1. Introduction

Primary HIV infection (PHI) is a condition that develops up to six months after human immunodeficiency virus (HIV) infection. According to European AIDS Clinical Society (EACS) guidelines, two types can be distinguished: acute and recent. The infection might be asymptomatic, further complicating the HIV diagnosis, or symptomatic [1]. However, even the symptomatic course is nonspecific because it has a wide array of clinical manifestations (flu-like illness) such as fever, lymphadenopathy, pharyngitis, maculopapular skin rash, headache, chronic diarrhea, muscle pain, or general fatigue [2]. This usually leads to a suspicion of viral upper respiratory tract infection, rather than HIV infection.

During this infection period, recognition is complex due to nonspecific clinical symptoms and diagnostic problems. In enzyme-linked immunosorbent assay (ELISA) test, false-positive results can occur, e.g. due to autoimmune diseases, pregnancy, or infections such as acute malaria [3-5]. Additionally, false-negative results were reported in association with agammaglobulinemia and testing within the window period. However, they can also occur in the early HIV infection stage because the antibodies may still be absent [6]. That is why negative or doubtful result for HIV antibodies cannot be the basis for excluding the infection [2].

To facilitate the diagnosis, the Fiebig scale has been developed. According to Fiebig et al. who distinguish six stages of PHI, the amount of HIV ribonucleic acid (RNA) copies increases at stages I to III. At stage I with undetectable p24 antigen and HIV Immunoglobulin M (IgM), the maximum viraemia level is between 10^4 to 10^5 copies/ml [7]. At this stage the determination of HIV RNA may be crucial as it appears as the first infection marker within the first ten days [7,8]. The peak viraemia (above 10^6 copies/ml) occurs at stage III, which is when antibody seroconversion occurs [9]. This is also when the symptoms may begin [10].

Many mechanisms seem to be important in controlling the immune response of an individual infected with HIV. They include: increased production of interleukine-2 (IL-2), interferon-1 (IFN-1) and interferon-γ (IFN-γ), cluster of differentiation 4+ (CD4+) T-cell proliferative responses and cluster of differentiation 8+ (CD8+) T-cell activity. They also increase synthesis of CD8+ T-cell suppressive factors and ß-chemokines, antibody-mediated mechanisms dependent on Fc receptors (FcRs) (e.g. antibody dependent cell-mediated

cytotoxicity), requiring natural killer cells (NK-cells), macrophages or neutrophils, NK-cell mediated lysis [10,11].

As previously mentioned, the diagnosis of HIV infection may be disrupted by other infections, pregnancy, autoimmune diseases, medications, and exogenous steroids taken by the patient [3,12,13]. Exogenous testosterone is broadly used by individuals with decreased hormone levels, as in Klinefelter Syndrome, and those who want to improve their muscle mass and body image. Its impact on immunological system cannot be neglected.

Androgens, including testosterone, are known to have an immunosuppressive role [14]. They play a significant role in HIV infection as they affect crucial mechanisms deployed against it. They decrease the activation of dendric cells and macrophages, production of cytokines released by macrophages, T helper 1 (Th1) T-cells, IFN-1, IFN-γ and interleukine-12 (IL-12). They also hamper T and B cell development and regulate the number of immunosuppressive neutrophils. Moreover, it has been found that, compared to females, plasmacytoid dendric cells in males produce less IFN-1 in reaction to HIV infection, which results in higher viral loads and is likely related to anabolic steroids [15].

2. Case presentation

We would like to present the case of a patient, a 37-year-old man having sex with men (MSM) who has been taking exogenous testosterone for fourteen years due to Klinefelter syndrome. He had unprotected sexual encounters that took place two weeks earlier. Although the patient has shown signs of an acute retroviral disease and has had high plasma viral load, neither third and fourth generation HIV tests nor Western blot confirmed the infection.

Our patient came to the Emergency Department presenting diarrhea, general weakness and fever (up to 38.5°C) lasting for fourteen days. He had a medical history of Klinefelter syndrome treated with exogenous testosterone (100 mg intramuscular/once a week), but no further documentation of endocrine treatment was accessible. Laboratory tests revealed thrombocytopenia (71 000 G/l) and leukopenia ($2.5x10^3/mm^3$). The patient did not give consent to hospitalization. After five days, he was admitted to another hospital due to the deterioration of his general condition. Laboratory tests were performed and showed thrombocytopenia (115 000 G/l) and leukopenia ($3.3x10^3/mm^3$) once again. Two consecutive third-generation HIV tests were performed, but both were inconclusive and difficult to interpret. However, the elevated levels of certain liver transaminases were concerning, namely alanine transaminase (ALT): 181 U/l, aspartate transaminase (AST): 316 U/l, as well as gamma-glutamyltranspeptidase (GGTP): 233 U/l and creatine kinase (CK): 1449 U/l.

Because of the suspicion of hepatitis of unclear etiology, the patient was referred to an infectious disease hospital for further investigation. There, the laboratory findings for Hepatitis A virus (HAV), Hepatitis B virus (HBV) (anti-HBc-total antibodies were negative, anti-HBs antibodies: 48 mIU/ml), Hepatitis C virus (HCV) (anti-HCV IgM negative), Epstein–Barr virus (EBV), and Cytomegalovirus (CMV) (anti-CMV IgM negative, anti-CMV IgG positive) infections were negative. The patient had been vaccinated against hepatitis B several years before.

The first test of the fourth generation *VIDAS*®HIV DUO Ultra assay was negative. Due to testosterone intake, clinical symptoms, and unprotected sexual encounters, it was decided that the *VIDAS*®HIV DUO Ultra assay should be repeated, despite the negative result. A second test was performed two days later and was very weakly positive for p24 antigen. Antibodies were still nonindicative. HIV Western blot confirmation test was negative. HIV viral load was assessed and revealed more than 10^7copies/ml, confirmed by real time polymerase chain reaction (PCR). The CD4 T-cell count was 351 cells/µl, CD8 T-cell count was 680 cells/µl, and CD4/CD8 ratio was 0.52.

There were no abnormalities in his clinical examinations or in the abdominal and pelvic ultrasound, except for a slightly enlarged spleen. During his stay in the hospital, the patient was also diagnosed with herpes labialis and treatment was initiated (aciclovir). After HIV infection was diagnosed, he received an antiretroviral therapy (ART) (emtricitabine, tenofovir disoproxil, raltegravir). The treatment was well-tolerated. Within a few days, the symptoms subsided, and transaminase levels decreased. The patient was discharged from the hospital. After four months of antiretroviral therapy, the viral load decreased to less than 20 copies/ml (i.e. was undetectable) and the CD4 T-cell count increased to 580 cells/µl. A follow-up abdominal ultrasound revealed that the spleen remained enlarged, despite antiretroviral treatment.

Table 1. Patient's results and reference values.

Test	Result	Reference values
Viral load	$>10^7$ copies/ml	undetectable
CD4 + T-cell	351 cells/mm^3	410-1590 cells/mm^3
CD8 + T-cell	680 cells/mm^3	190-1140 cells/mm^3
CD4+/CD8+ ratio	0.52	>1
Platelet count	71 000 G/l	120 000 – 350 000 G/l
Leukocytes	2.5 x$10^3/mm^3$	4.0 - 10.0 x$10^3/mm^3$
ALT	181 U/l	10 – 70 U/l
AST	316 U/l	10 – 59 U/l
GGTP	233 U/l	15 - 73 U/l
CK	1449 U/l	55 - 170 U/l
HAV IgM	negative	negative
Anti-HBc – total	negative	negative
Anti-HBs	48 mIU/ml	>10 mIU/ml after vaccination
Anti-HCV IgM	negative	negative
Anti-CMV IgM	negative	negative
Anti-CMV IgG	positive	negative

3. Discussion

Our case report highlights the importance of conducting a thorough medical history to prevent misdiagnosing PHI. Although in Fiebig's research, at

stages II and III p24 antigen was detected in all samples, our patient had negative *VIDAS*®HIV DUO Ultra test results for antigen p24 and HIV antibodies. Despite the fact that antigen p24 test should be positive after approximately seventeen days of an HIV acquisition, it was very weakly detectable after about three weeks since the symptoms had begun [7]. This raises a suspicion that the detectability of the infection was somehow delayed.

Moreover, according to Ananworanich *et al.*, the number of CD4+ before ART is higher at stage I than at later stages (508 *vs.* 340 cells/mm^3) and so is the CD4+/CD8+ ratio (1.1 *vs.* 0.7) [16]. Despite this, our patient had low CD4+ cells count (351 cells/mm^3) and a low CD4+/CD8+ ratio (0.52). According to Koçar's research, androgen deficiency enhances B-cell response, whereas androgen replacement treatment inhibits immunoglobulin (IgG, IgM, IgA) synthesis. Furthermore, testosterone replacement therapy in Klinefelter syndrome patients led to a decrease of not only serum antibodies but also interleukins IL-2, IL-4 and total T and B cells levels. Additionally, it significantly reduced the number of CD4+ and CD4+/CD8+ ratio [17].

Considering the immunosuppressive character of exogenous testosterone used by the patient as a person with Klinefelter syndrome, we assume that this might have caused an abnormal reaction of the patient's immunological system to primary HIV infection which resulted in high viral load and a low number of CD4+ cells. These diagnostics findings can be associated with the severity of the patient's symptoms [18].

Individuals with PHI harbor a high level of viraemia that increases the risk of spreading the virus [19]. Early diagnosis and treatment of people newly infected with HIV would give the benefit of lesser seeding of viral reservoir in the patient and reduce the possible further transmission of the virus [20]. As our case has shown, the diagnosis of primary HIV infection can be hard to obtain, e.g. due to anabolic steroid usage. Testosterone's impact on the immunological system fighting HIV infection cannot be neglected.

4. Conclusion

The PHI diagnosis of patients with testosterone intake requires special attention. To prevent misdiagnosis, it should be considered to take a real time PCR test at the beginning of diagnosis of HIV infection as the *VIDAS*®HIV DUO Ultra test and Western blot might be negative. Clinicians must keep in mind that the severity of primary HIV infection varies depending on the patient's other diseases and treatment.

Author(s) contribution

TM contributed to the conception and gathered materials for this study. OG and GK collected the data, processed it and wrote the manuscript. OG, GK, TM contributed to the literature review and design of the case report. TM and AWD made the supervision and critical review. All authors contributed to manuscript revision, read and approved the submitted version.

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Informed Consent Statement

Informed consent was obtained from the patient.

Conflict of interest

The authors declare no conflict of interest.

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