

DETECTION OF HUMAN HERPESVIRUS-8 ANTIGEN IN HIV-INFECTED PATIENTS IN EAST JAVA, INDONESIA

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**Abstract**

**Background:** Co-infection of human herpesvirus-8 (HHV-8) in HIV-positive people might cause Kaposi's sarcoma. Early detection of HHV-8 may prevent the onset of clinical manifestations. In Indonesia, detection of HHV-8 antigen in HIV-positive patients has yet to be reported. The intention of this research was to examine the presence of HHV-8 antigen in HIV-positive patients in East Java, Indonesia.

**Material and Methods:** 103 serum samples were collected from HIV-positive patients in Surabaya and Tulungagung, East Java, Indonesia. Serums were then tested for the presence of HHV-8 antigen by using sandwich ELISA.

**Results:** Human Herpesvirus-8 antigen was detected in 15 samples (14.5%). The presence of HHV-8 infection in HIV-positive patients did not present differently in males and females and among different age groups. Human immunodeficiency virus-positive serum samples were collected from 23 homosexual men, 25 intravenous drug users (IVDUs) and 52 heterosexuals. In the male homosexual group, HHV-8 antigen was detected in 21.7% (5/23) of the samples, while in the intravenous drug user group (IVDUs), 16% (4/25) of the samples were found to have HHV-8 antigen.

**Conclusion:** This research found the presence of HHV-8 antigen in 14.5% of patients in East Java, Indonesia. It is recommended that patients with a positive result should receive further examination to detect any clinical manifestations related to HHV-8 infection, especially in the form of Kaposi's sarcoma lesions, so that the illness can be appropriately managed.

**Key Words:** HHV-8 antigen, HIV-positive, East Java, Indonesia.

**Introduction**

The human immunodeficiency virus (HIV) primarily attacks T helper (CD4+) lymphocytes. The human immunodeficiency virus (HIV) infection causes the immune system to weaken, so that the host becomes more susceptible to various infections and malignancies (Grant et al., 2005). One common co-infection found in people living with HIV/AIDS is a herpesvirus infection, including herpes simplex virus types 1 and 2 (HSV-1, HSV-2), varicella zoster virus, cytomegalovirus (CMV), Epstein-Barr virus (EBV), herpesviruses 6 and 7, and human herpesvirus 8 (HHV-8 or Kaposi sarcoma-associated herpesvirus [KSHV]) (Brooks et al., 2013). HHV-8 infection in HIV/AIDS patients may cause Kaposi's Sarcoma (Dourminishev et al., 2003). Kaposi's sarcoma is a tumor developed from cells on lymph nodes or in blood vessels, and can also develop in other parts of the body (Dourminishev et al., 2003).

HHV-8 or KSHV, the only known human herpesvirus (rhadinovirus), is the most recently discovered tumor virus (Viejo-Borbolla et al., 2003). HHV-8 has a diameter of 140 nm and a genome of between approximately 125 kb (Burbelo et al., 2015), 165 kb (Brooks et al., 2013) and 170 kb (Russo et al., 1996). The virus has a double stranded linear DNA and icosahedral capsid envelope (Viejo-Borbolla et al., 2003). It is covered by a tegument containing protein, and closed during

budding of the cell. The membrane is derived from outer envelope of lipid membrane from various host and specific virus glycoprotein (Swaminathan, 2003).

Kaposi's sarcoma is more prevalent in immunosuppressed patients than healthy people. It often occurs in people with Jewish, Mediterranean, African, and Middle Eastern origins. Sero-epidemiological evidence of HHV-8 infection can be used to reflect the epidemiology of Kaposi's sarcoma (Grulich et al., 2001). Ablashi and colleagues' research reports that in 1999, seropositive prevalence of HHV-8 infection in general population of USA, Northern Europe, and Asia was only around zero to five percent; this was also the case in a seroepidemiological study in Malaysia, Hong Kong, and Sri Lanka where the seroprevalence of HHV-8 was found to be around 3,7%. However, a higher prevalence was found in Southern Europe, around five to ten percent. (Ablashi et al., 1999). There has been no data regarding viremia detection using ELISA method and no epidemiological reporting on the prevalence of HHV-8 infection in Indonesia. The aim of this study was to correct this omission with an examination of the presence of HHV-8 antigen in HIV-positive patients in East Java, Indonesia.

## Materials and Methods

This study was approved by the Ethics Committee of Universitas Airlangga Hospital. A hundred and three serum samples were collected from HIV-positive patients in Universitas Airlangga Hospital, Surabaya and Dokter Iskak Tulungagung General Hospital, East Java, Indonesia. The inclusions criteria are positive HIV infected with naïve or treated with antiretroviral, all age (in this case 7-5 years old, and the mean age is 37 years old). Exclusion criteria in this study were patients who did not agree to follow the study. Informed consent was obtained from each patient prior to sample collection. Patients were also asked to complete a questionnaire to provide general information and the possible route of transmission. The patients were then subjected to an HHV-8 antigen test using sandwich ELISA (Abbexa, Cambridge, UK), according to manufacturer instructions. Data obtained were analyzed using statistical software (SPSS version 21.0).

## Results

HIV-positive patients involved in this study comprised of 64 males and 39 females, with a mean age of 37 years (range 7–64 years). This research showed that 15 of 103 samples (14.56%) were found to be HHV-8 antigen positive. In regards to patients' gender, ten men (15.6%) and five women (12.8%) tested positive. The presence of HHV-8 antigen did not show any significant differences in age ( $p=0.05$ ) and gender ( $p=0.78$ ). HHV-8 infection in homosexual men was 21.7%, while 16% intravenous drug users (IVDUs) were positive.

**Table 1.** Determinants of HHV-8 infection in HIV positive patients

Characteristic	N	HHV-8 positive N (%)	P value p<0,05, S (Significant), NS (Not Significant)
<b>Age group (years)</b>			
A: ≤ 30	31	4 (12,9%)	p= 0,5 (NS)
B: 31-40	35	7 (20%)	
C: >40	37	4 (10,8%)	
<b>Sex</b>			
Male	64	10 (15,6%)	p= 0,78 (NS)
Female	39	5 (12,8%)	
<b>Sexual orientation</b>			
Homo/bisexual	25	4 (16%)	p= 0,5 (NS)
Heterosexual	78	11 (14,1%)	
<b>Marital status</b>			
Married	68	10 (15,6%)	p=0,6 (NS)
Not married	35	5 (12,8%)	

## Discussion

The results of the study conducted in East Java, Indonesia are an interesting comparison with other studies around the world. It must be reiterated here that the participants in the study were not screened for Kaposi sarcoma and the ELISA method was not used to collect the data regarding HHV-8 viremia. While the presence of HHV-8 antigen was detected in 14.56% of HIV-positive patients in East Java, Indonesia, previous studies in Xinjiang, China (Zhang et al., 2012), Nigeria (Ogoina et al., 2011) and Cameroon (Mbondji-Wonje et al., 2013) showed a greater prevalence, that is

31.2%, 62% and 70% respectively. Human herpesvirus 8 antigen was detected in 21.7% HIV-positive homosexual men. A study in Thailand reported the presence of HHV-8 lytic antigen in 12% HIV-positive homosexual men, 16% heterosexual men, and 9% IVDUs (Ayuthaya et al., 2002). HHV-8 without KS in serum samples are detected 4% and AIDS-KS are 42.8% in Brazil (Machado et al., 2016). Detection of herpes virus DNA in lymphocytes could possibly represent latent infection, while detectable DNA in serum or plasma is usually associated with disease (Polstra et al., 2003). In HIV-positive patients without Kaposi Sarcoma, 4% were positive using PCR method. These patients certainly present a higher risk of developing AIDS-KS (Machado et al., 2016).

Kaposi's sarcoma was the most common neoplasm found in patients infected with HIV. Kaposi's sarcoma is a tumor of blood vessels, which appears as lesions on the skin, and then spreads to other organs. It is noted here that there has been a decrease in the incidence of Kaposi's with other opportunistic illnesses, since the introduction of highly active antiretroviral therapy (HAART) (Jacobson et al., 1999). This downward trend is most likely due to secondary effects of antiretroviral therapy on the immune system (Cattelan et al., 2001 dan De Milito et al., 1999) rather than direct effect to HHV-8 (Kedes dan Ganem, 1997).. A study utilizing polymerase chain reaction (PCR) demonstrated that HHV-8 was detected in more than 90% of Kaposi's sarcoma lesions (Mendez et al., 1998). Prospective studies indicated that the detection of HHV-8 could predict the development of Kaposi's sarcoma in asymptomatic subjects (Whitby et al., 1995 dan Moore et al., 1996). Several studies using different serological tests found that almost 100% patients with different clinical forms of Kaposi's sarcoma had a high titer of the HHV-8 antibody, which had been detected before the appearance of clinical lesions of Kaposi's sarcoma (Whitby et al., 1995; Gao et al., 1996 dan Moore et al., 1996). Current evidence suggests that HHV-8 infection is a prerequisite in the pathogenesis of Kaposi's sarcoma, but factors relating to the host immuno-deficiency may greatly increase the incidence of Kaposi's sarcoma among subjects infected with HHV-8.

The presence of HHV-8 infection in HIV-positive patients is possibly related to the risk of developing opportunistic diseases, including Kaposi's sarcoma (classic, AIDS-related KS, endemic and iatrogenic) and other proliferative diseases, such as primary effusion lymphoma and multicentric Castleman disease. Among immuno-competent people, only a small proportion of HHV-8 infections develop into Kaposi sarcoma. Various tests have detected HHV-8 antibodies in the majority of asymptomatic individuals with increased tumor risk, including HIV-positive homosexual men (Martin et al., 1998), adults from Italy and various regions of sub-Saharan Africa (Sitas et al., 1999; Calabro et al., 1998), and some low risk people, such as blood donors from the UK, USA, and Jamaica (Simpson et al., 1996; Engels et al., 1999).

## Conclusion

The presence of HHV-8 antigen was found in 14.5% of the HIV-positive patients who participated in this study which is the first of its kind in Indonesia. Based on these results found in patients in East Java, further investigations should be conducted on patients living with HIV. This would assist with early detection and subsequent management of any clinical manifestations related to HHV-8 infection, especially in the form of Kaposi's sarcoma lesion.

**Conflict of Interest:** The authors declare no conflict of interest.

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