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SERUM PROTEIN ELECTROPHORESIS UNDER EFFECTIVE CONTROL OF HIV-1 DISEASE PROGRESSION

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ABSTRACT

In this report, we compared the serum protein electrophoresis (SPE) patterns in a subset of HIV-1-infected subjects who did not progress to AIDS without antiretroviral treatment with those in whose control of disease progression was achieved by highly active antiretroviral therapy (HAART). SPE and immunofixation electrophoresis were performed on Helena Electrophoresis System according to manufacturer's instructions. The percentage of SPE abnormalities, resembling chronic inflammation, was significantly higher in HIV-1-infected subject without HAART compared with those under HAART ($p = 0.001$). The majority of individuals under HAART showed evidence of oligoclonal bands on the γ-band against a polyclonal background compared with those without HAART but β-γ-band bridging was more evident. Immunofixation pattern was consistent with oligoclonal hypergammaglobulinaemia of IgG kappa type, which was found to be more intense in group without HAART. HIV clinical status did not show appreciable effect on the SPE pattern in subjects without HAART. However, under effective HAART, subjects with better CD4 T-cell count were associated with higher γ-globulin band. In group without HAART, acute infection was found to be associated the higher γ-globulin fraction compared with chronic infection. The opposite was the case under effective HAART. HIV infected subjects that did not progress to AIDS were associated with markedly abnormal SPE pattern. Overall results reflect the host ability compensate defective cellular immunity in HIV-1 infection with humoral immune responses. These findings underscore the usefulness of SPE monitoring HIV disease management and identifying individuals that may not progress to full-blown AIDS in the absence of treatment.

Keywords: HIV, HAART, SPE, IFE, clinical status, duration

INTRODUCTION

Human immunodeficiency virus type 1 (HIV-1) selectively infects immune cells, thus resulting in depletion of peripheral blood CD4 T-lymphocyte population (Post et al., 1996, Cloyd et al., 2000). According to the joint United Nations Programme on HIV/AIDS (UNAIDS) and World Health Organization (WHO), 42 million people

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lived with HIV/AIDS worldwide in year 2002, resulting in 3 million deaths and majority of cases occurred in sub-Saharan Africa. According to a later report on the global AIDS epidemic, 33 million people lived with the disease worldwide while about 2 million people died in 2007. Recent global reports show a decreasing new infections and AIDS-related deaths. Thus more (35 million) people lived with the disease in 2010 (UNAID, 2011). Similar trends were observed in Nigeria during the period.

The progression of HIV disease to full-blown AIDS is usually associated with progressive increase in HIV-1 viral load in addition to defects in cell-mediated immunity. Effective cell-medicated immune machinery is therefore found to stem the tide of disease progression (Rosenberg et al., 1997; Dyer et al., 2008). The rapid reduction in AIDS-related mortality and increase in people living with HIV are largely due to the introduction of highly active antiretroviral therapy (HAART). HAART has been shown to play critical roles in suppressing viral load and increasing CD4+ T lymphocyte counts, which translates to significant reduced AIDS related morbidity and mortality among HIV/AIDS patients (Palella et al, 1998; Arminio et al., 2005). However, subsets of people living with HIV in Nigeria have achieved control over disease progression without treatment and similar observations have been reported in therapy-naïve individuals elsewhere (Dyer et al., 2008). These observations show that lack of HIV disease progression can be independently obtained with the host's immune responses and HAART. Viral, genetic and immunological factors have been identified for this phenomenon (Poropatich and Sullivan, 2011) and some of the factors common with individuals who have been exposed to HIV infection but remained uninfected may also be associated with HIV-1-infected subjects naïve treatment and yet resist progression to AIDS (Lederman et al., 2010).

Unfortunately, none of the identified viral, genetic and immunological factors is routinely investigated to identifying and predicting HIV infected individuals that may resist progression to AIDS in the absence of treatment, most especially in resources-poor settings where mere blood CD4 T lymphocyte quantity is used to qualify candidates for antiretroviral therapy. Since all antiretroviral drugs have been shown to have both short-term and long-term adverse reactions (Montessori et al., 2004), the need to identify HIV-infected subjects that would not progress to AIDS in the absence of HAART becomes highly imperative. In the present research, we compared the serum protein electrophoresis patterns in a subset of HIV-1-infected Nigerian subjects who achieved control over disease progression without treatment with those in whom control of HIV disease progression was achieved by HAART, as this may reveal the precise usefulness of SPE in identifying HIV-infected individuals that may not progress to full-blown AIDS in the absence of treatment.

MATERIALS AND METHODS

Selection of subjects

HIV-1 infected subjects attending Living Hope Care, Ilesa, constituted the majority of subjects for this study. Others were selected from General Hospital, Iwo and Baptist Health Centre Ejigbo, South-western Nigeria. Two hundred and sixty (260) subjects were studied. 75 % of selected subjects had been receiving effective oral highly active antiretroviral therapy (HAART) [Lamivudine (300 mg/day), Stavudine (60 mg/day) and Nevirapine (400 mg/day)] while 25 % were not on antiretroviral treatment between year 2007 and 2010. In the group not on HAART, there was no history of AIDS diagnosis including a CD4 count < 200 cells/µl, or self-reported occurrence of any AIDS-defining illness or AIDS diagnosis. Pregnant women and individuals co-infected with tuberculosis and/or hepatitis virus were excluded from

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the study, because these conditions may obscure or aggravate any potential effect on the natural history of HIV disease on serum proteins. The HIV-1-infected subjects studied were asymptomatic and had CD4 T cell counts > 200 cells/µl as at the time of enrolment. Thirty age-matched apparently healthy volunteers were also selected as controls. The research was approved by the Research Ethics Committee of Living Hope Care and informed consent was obtained from all subjects.

Diagnosis of HIV Infection and enumeration of blood CD4 T-cell

The diagnosis of HIV-1 infection was performed by Enzyme-Linked Immunosorbent Assay (ELISA) and confirmation was done by HIV Western Blot using Immunetics Qualicode HIV-1/2 Kit (USA). Subjects with indeterminate results were excluded from the study. Control subjects were also confirmed to be HIV seronegative. EDTA-anticougulated blood CD4 T-cell was enumerated using Cyflow® Cytometer according to the manufacturer's instructions (Partec, Germany).

Electrophoresis and immunofixation

Serum protein electrophoresis was performed using Helena Electrophoresis system, according to manufacturer's instructions (Beaumont, TX; Sebia, Norcross, GA, USA). The electrophoresis gels were independently examined by two of the authors (TLO and ALA) and discrepancies were resolved by consensus. IFE was performed using commercial kit obtained from Bindingsite, Birmingham, United Kingdom. The facilities were stored and used according to the manufacturer's instructions.

Densitometry of electrophoregram

The strip was removed from the final rinse tray and excess rinse solution was allowed to drip off into rinse tray. The strip was carefully trimmed and immersed in 40 % aqueous N-methyl pyrolidone for 5 minutes. Strip was removed from cleaning solution and placed lengthwise on a glass slide. A second glass was gently pulled across the surface at a 45° angle to remove xcess fluid and bubbles. The end of strip was folded over the edges of the glass slide and excess trimmed to about 0.6 cm overlap. The slide was placed in an 80-90 °C oven for 15 minutes to produce a completely transparent background. Densitometry scanning was performed on a Helena Automatic Computing Densitometer at 525 nm according to the manufacturer's instruction (Helena Automatic computing Densitometer, Helena Laboratories, Beaumont, TX; Sebia, Norcross, GA.USA).

Statistical analysis

Descriptive analysis, and student t-test was used for the comparisons of data. Spearman correlation and Fisher's test were used to test association between variables, as appropriate, using Graphpad® 5 software (San Diego, CA). p-values < 0.05 were considered significant.

RESULTS

Study subjects

HIV-1 infected subjects naïve to HAART had a median CD4 lymphocyte count of 440 (IQR 321-510) cells/µl while that of HIV-1 infected subjects under HAART was 410 (IQR 300-605) cells/µl.

Although, the female to male ratio was about 3.3, the blood CD4 T-cell counts of the groups were not significantly different ($p > 0.05$). This implies that the HIV-1-infected subjects studied, HAART-naive and treated, had similar clinical status (Table 1).

Serum protein electrophoresis pattern

A typical HIV-1 infected subject, in this study, regardless of HAART status, exhibited a more intense staining at the gamma regions of the electrophoregrams than healthy HIV-1 uninfected controls. In most of the cases studied, increases in gamma globulin bands were accompanied by apparent decreases in albumin bands.

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Table 1: Characteristics of study subjects

CHARACTERISTICS	HIV-	HAART-	HAART+	p-Value
N	30	65	195	
Age (years)	37 (35, 45)	36 (34, 48)	35 (32, 47)	0.352
MAC(cm)	28 (24, 29)	29 (28, 30)	27 (25, 31)	0.505
Body Mass Index (Kgm^{-2})	24 (22, 25)	24 (21, 26)	25 (22, 27)	0.505
Sex (M/F)	3/7	7/24	8/23	0.605
Infection Duration (months)	NA	12 (4, 45)	16 (10, 55)	0.236
HAART Duration (months)	NA	0	14 (5, 48)	NA
CD4 T-cell count (/µl)	807 (688, 900)	440 (321, 510)	410 (310, 605)	0.558

Values are medium (25^{th} and 75^{th} percentile). p-values were determined by Student's 't' test and Fisher's exact test, as appropriate to compare HAART- and HAART+, $p < 0.05$ was considered significantly different. Not applicable (NA); Mid Arm Circumference (MAC).

Changes in other globulin bands were not apparent. It is pertinent to state that some of healthy HIV-1-uninfected control studied showed mild diffused rise in the γ-bands but with normal albumin band. The HIV-1 infected subjects were grouped either as HAART-treated (HAART+) or without HAART (HAART-). Visual examination of the electrophoregrams revealed a diffused rise in the gamma regions in both groups. However, the staining of the gamma regions of group without HAART was consistently more intense than that of HAART+ group. Densitometry scanning enabled a better appreciation of the behaviour of protein fractions. The majority of individuals under HAART showed evidence of oligoclonal bands on the γ-band against a polyclonal background compared with those without HAART but β-γ-band bridging was more evident. These features were not so obvious on visual examination of the electrophoregrams. Densitometry also revealed the relative proportions of serum protein fractions more clearly.

To confirm whether the increase in the gamma globulin fraction was due to homogeneous protein, immunofixation electrophoresis was performed on sera suspected to exhibit homogeneous protein in both groups. The patterns obtained were consistent with oligoclonal hypergammaglobulinaemia of IgG kappa type. The oligoclonal IgG κ-banding was found to be more intensely stained in subjects without HAART (Figure 1). No apparent abnormality was discovered in α_1- and α_2-globulin bands under effective control of HIV-1 disease progression. The percentage of SPE abnormalities was significantly higher in HIV-1 infected subject without HAART compared with those under effective HAART ($p = 0.003$). However, the patterns in both groups were consistent with chronic inflammation. The summary of visual comparison of SPE pattern is shown in Table 2.

Effect of clinical status and HAART/infection duration on SPE pattern

To ascertain the impact of clinical (CD4) status and HAART/infection duration on the SPE pattern, the subjects were divided into sub-groups defined by CD4 T lymphocyte counts (< 500 or > 500 cells/µl); and HAART/infection duration: Acute (< 12 months) or Chronic (> 36 months). In the group without HAART, HIV clinical status did not show appreciable effect on the SPE pattern. A representative pattern in subjects with a CD4 T cells count < 500/µl was found to be essentially similar to those with CD4 T-cell count > 500/ul. Under HAART, however, subjects with a better clinical status were associated with higher γ-globulin bands (Figure 2). In group without HAART, acute infection was found to be associated the higher γ-globulin fraction compared with chronic infection. The opposite was true under effective HAART. HIV infected subjects that did not progress to AIDS were associated with markedly abnormal SPE pattern (Figure 3).

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Table 2: Visual analysis of electrophoresis

SPE Pattern	HAART-	HAART+	p-value
N	65	195	-
Normal	16 (25)	90 (46)	0.001
Abnormal	49(75)	105(54)	0.001
Albumin ↓	37(60)	37(19)	0.001
α_1-globulins ↓↑	0(0)	0(0)	1.000
α_2-globulins ↓↑	0(0)	0(0)	1.000
β-globulins ↑	7(14)	16(8)	0.258
γ-globulins ↑	46(71)	96(49)	0.001

Values are findings (%). p-values were determined by Fisher's test and $p < 0.05$ was considered significantly different Arrows refer to increased (↑), or decreased (↓) regulation.

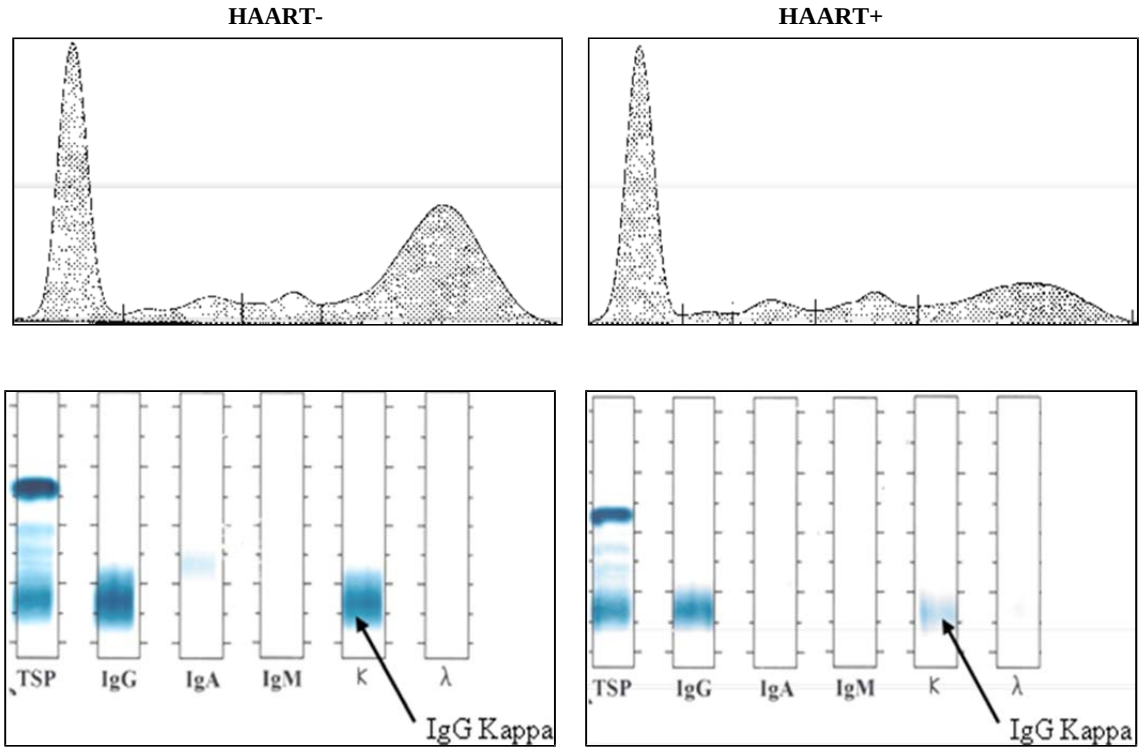

Figure 1: Representative serum protein electrophoresis (SPE) and immunofixation electrophoresis (IFE) under effective control of HIV-1-diasese progression. Comparison of densitometry scans of electrophoregram of HIV-1-infected subject naïve to HAART (HAART-) and under HAART (HAART+). SPE was performed under non-denaturing condition in diethylbarbital buffer, pH 8.6 and visualized with Ponceau S stain. Scanning was performed on a Helena Automatic Computing Densitometer at 525nm [Upper panel]. Comparison of IFE of sera from subjects suspected to exhibit homogeneous immunoglobulins (M-protein) of HAART- and HAART+. The gel was visualized with Acid Blue 29 after immunofixation with (left to right) acid fixation (TSP lane); anti-human IgG heavy chain(IgG lane); anti-human IgA heavy chain(IgA lane); anti-human IgM heavy chain (IgM lane), anti-human kappa light chain (κ lane) and anti-human lambda light chain (λ lane). The arrows highlight an oligoclonal IgG κ-banding [Lower panel].

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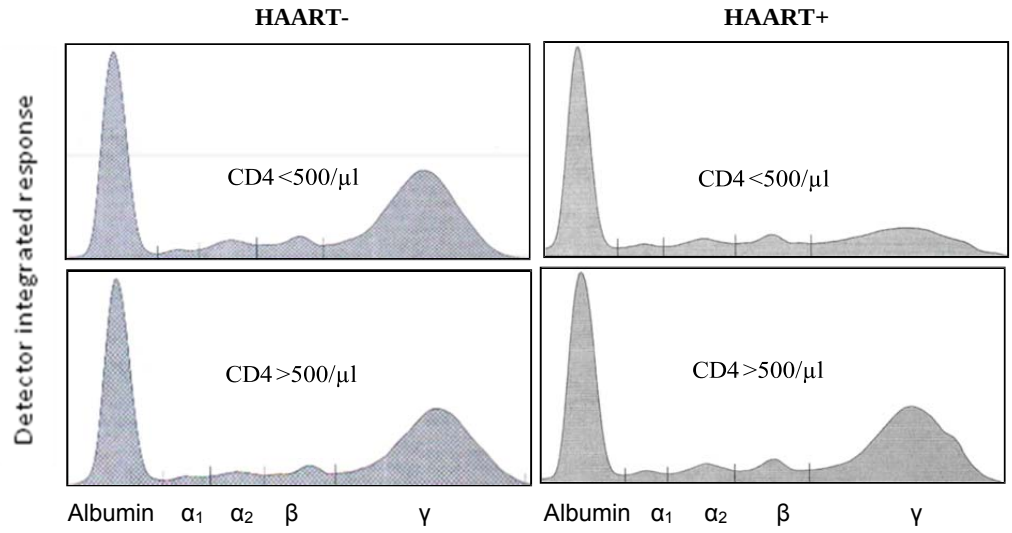

Figure 2: Impact of HIV clinical status on densitometry pattern of serum protein electrophoresis (SPE) under effective control of HIV-1-disease progression. Left panel compares the representative pattern of subjects with CD4 T cell blood count < 500/µl (upper) and > 500/µl (lower) naïve to HAART (HAART-) while right panel compares the representative pattern of subjects with CD4 T cell blood count < 500/µl (upper) and > 500/µl (lower) under HAART (HAART+). SPE was performed under non-denaturing condition using diethylbarbital buffer (pH 8.6) and visualized with Ponceau S stain. Densitometry scanning was performed on a Helena Automatic Computing Densitometer at 525 nm.

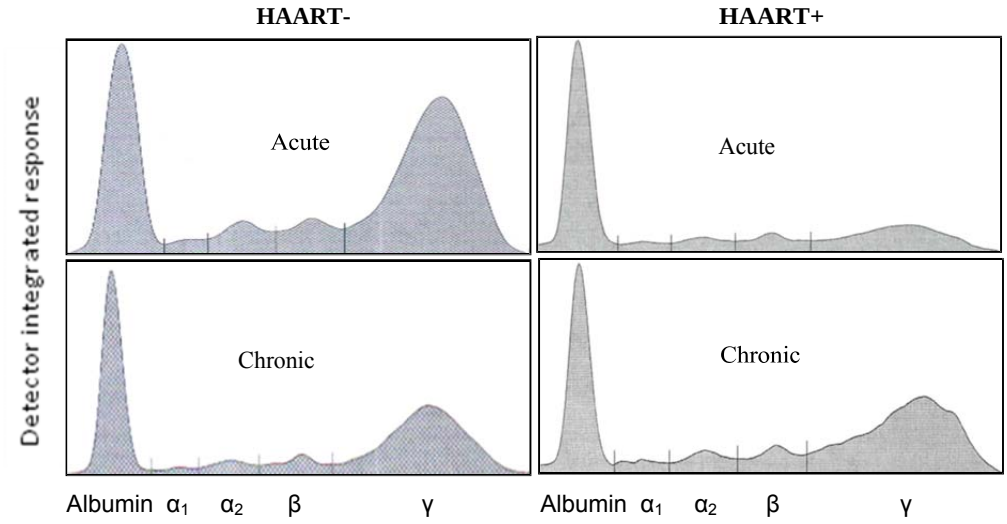

Figure 3: Impact of HAART and infection duration on densitometry pattern of serum protein electrophoresis (SPE) under effective control of HIV-1-disease progression. Left panel compares the representative pattern of subjects with acute (upper) and chronic infection (lower) naïve to HAART (HAART-) while right panel compares the representative pattern of subjects with acute (upper) and chronic infection (lower) under HAART (HAART+). SPE was performed under non-denaturing condition using diethylbarbital buffer (pH 8.6) and visualized with Ponceau S stain. Densitometry scanning was performed on a Helena Automatic Computing Densitometer at 525 nm.

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DISCUSSION

Previous studies plus personal experience with support groups for people living with HIV/AIDS have shown that control of disease progression in HIV-1 infection could also be achieved without antiretroviral chemotherapy (Resino et al., 2003). This shows that the host potent HIV specific and non-specific immunological responses in individual naïve to treatment play crucial roles in control of disease progression. Specific and non-specific humoral immune responses can be assessed immunological techniques including serum protein electrophoresis (SPE). In an earlier study, we observed a significant variability in the serum protein pattern of subjects infected with HIV-1, even within the same clinical (CD4 T-cell count) status (Adedeji et al., 2004). It was then speculated that individual genetic and intrinsic homeostatic variations (host factors) were responsible for the phenomenon. Introduction of antiretroviral therapy had drastically reduced AIDS related mortality in HIV infection. Since adoption of antiretroviral drugs and HAART have altered the outcome and prognosis for HIV-infection, we sought to study the peculiar association of SPE pattern with effective control of HIV disease progression and its possible usefulness as an adjunct to the standard protocol (CD4 T-cell count) in HIV/AIDS control.

In this study, we specifically compared the serum protein electrophoresis pattern of HIV-1 infected subjects under effective highly active antiretroviral therapy (HAART) as well as those latently infected but did not progress to AIDS despite absence of treatment (HAART naïve). The majority of the subjects studied were females (approximately 77 %), either in the HAART or HAART naïve group; giving a female: male ratio of about 3.3. This may imply and support the notion that females are more susceptible to HIV-1 infection (Glynn et al., 2001). Of course, other factors may be responsible for the high female: male ratio (Koblin et al, 2000). However, others reported a very low female: male ratio (about 0.3) in their study population (Konstantinopoulos et al., 2007). In this study, it was discovered that the HIV-1 infected subjects under effective HAART had a higher body mass index (BMI) than the HAART naive group. Since the majority of subjects in this study were females, the higher BMI in the present study is in agreement with McDermott et al. (2001) who reported a higher BMI in HIV infected women under HAART. Stavudine, a composite of HAART in this study is known to cause fat redistribution (Gervasoni et al., 1999) and may contribute to the change in BMI.

Case note information on the subjects showed that the individuals in HAART group were either in group ‘B’ or ‘C’ of the 1993 CDC criteria prior to initiation of HAART while the HAART naïve group ware in stage ‘A1’ and ‘A2’ (CDC, 1992). After effective control of HIV-1 disease control however, the HIV-1-infected subjects under HAART had a median CD4 lymphocyte count 410 cells/µl (IQR 300-605) which was not significantly different ($p = 0.427$) from those naive to HAART [440 cells/µl (IQR 321-510)]. Therefore, the subjects in the present study were apparently in group II of the 1986 CDC classification system for HIV infection (CDC, 1986). The overall characteristics of HIV-1 infected subjects under effective highly active antiretroviral therapy (HAART) was not significantly different ($p > 0.05$) from those latently infected but did not progress to AIDS despite absence of treatment.

SPE separates serum proteins based on their physical properties. Cellulose acetate and agarose are the widely used as supporting media. Their use permit resolution (after staining) of serum proteins into five bands designated albumin, alpha-1, alpha-2, beta and gamma factions respectively. The stained strip of cellulose acetate (or other supporting medium) is called an electrophoregram. The amount of these five bands can be assessed visually and quanti-

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fied by the use of densitometry scanning equipment. Characteristic changes in the amount of one or more of these five bands are found in many diseases (Vavricka et al., 2009).

The SPE pattern under effective control of disease progression was first compared with HIV-1-uninfected control. It was discovered that the typical HIV infected subjects under effective control of HIV disease had SPE pattern quite different from that of the HIV un-infected controls. There was diffused rise in the γ-band leading to a more intense staining at the gamma region of the electrophoregram implies a higher concentration of γ-globulin fraction and thus a more vigorous humoral immune responses in the subjects that did not progress to AIDS in the absence of treatment. This was also confirmed by the densitometry scanning of the electrophoregram. Since serum antibodies migrate at the γ-region, the present work supports that of Jacobson et al. (2002) who reported higher antibodies concentration in HIV-infected subjects compared with HIV-uninfected controls. The γ-globulin fraction of the electrophoregrams constitutes the primary arm of the humoral immune responses. The persistent abnormally high γ-globulin fractions in the face of effective control of HIV disease progression suggests a compensatory phenomenon for the deficient cellular immunity associated with HIV-1 infection. Since HIV is an intracellular pathogen and adoptive transfer of immune globulin is known to improve quality of life of AIDS patients (Durandy et al., 2009; Onyango-Makumbi et al., 2011), humoral responses would thus have indirect effects on viral replication. The broad appearance of the γ-globulin band suggests polyclonal responses. This depicts adaptive humoral immune responses not only against HIV antigenic determinants but also to possible complex antigenic epitopes on opportunistic infectious agents which usually contribute to disease progression. Humoral responses may also exert direct protective action in a number of ways such as recruitment of the complement pathway to the destruction or removal of a pathogen. Antibody binding to bacterial surfaces may promote opsonization, phagocytosis and killing by macrophages and neutrophils. Viruses can be bound and neutralized by antibody, even as the antibody marks the pathogen for removal from the body by phagocytes. By the initiation of antibody-dependent cell-mediated cytotoxicity, antibodies can also mediate the killing of target cells by cytotoxic cell populations such as natural killer cells (Roitt et al., 1989). Possible expression of potent endogenous suppressors of HIV such as inhibitors of serine proteases may also contribute to this phenomenon (Shapiro et al., 2001).

In a healthy individual, the total kappa to lambda ratio is roughly 3:1 in serum and the ratio may vary widely in pathologic conditions such as in HIV infection (Katzmann et al., 2002). In this study immunofixation electrophoresis confirmed that the elevated immunoglobulin was mainly due to IgG-kappa especially in HAART-naïve group. Although the immunofixation pattern did not reveal presence of IgG-lambda, it does not indicate that the subjects sera contained no IgG-lambda. Rather, it indicates elevated IgG-kappa as the sera were appropriately diluted before immunofixation. This dilution may have decreased the serum IgG-lamda below detection. The same explanation may also be applicable to IgA and IgM that were not demonstrated (Figure 1).

It is noteworthy that the majority of individuals under HAART showed evidence of oligoclonal bands on the γ-band against a polyclonal background compared with HAART-naïve but β-γ-band bridging was more evident under HAART-naïve. Oligoclonal banding has been reported to be a common feature of HIV infection (Ng et al., 1988; Tathiah et al., 2011). This is likely to be caused by hyperactivation of B-cells due to chronic antigenic stimulation by antigens of HIV itself or other opportunistic infections. Tathiah et al. (2011) have

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reported increased polyclonal gammopathy and oligoclonal bands in HIV infection and that the majority of oligoclonal bands were present on a background of polyclonal gammopathy, suggesting simultaneous polyclonal B-cell activation and selective B-cell oligoclonal proliferation. This condition may have developed before the initiation of treatment. IgA migrate at the γ-band but very close to β-band (Mayne, 1994) elevated level of IgA may cause the β-γ-band bridging. However, immune-fixation pattern did not support elevation serum IgA under HAART (Figure 1).

Changes in other globulin bands were not apparent. Significant changes must have occurred before there could be obvious changes as the case in the γ-globulin region. This does not rule out significant variation that may have occurred in the concentrations of individual globulin fractions. Decrease in albumin fractions often accompanies increase in globulin fractions in many pathological cases such as in liver disease (Vavricka et al., 2009). HIV infection appears to be also associated with this phenomenon in our present study. The increased serum globulins concentration is essentially a compensatory mechanism for decreased albumin synthesis. It is noteworthy that some of healthy HIV-1 uninfected control studied showed mild diffuse rise in the γ-band with normal albumin band. Earlier studies speculated that endemicity of malaria as a possible reason for higher γ-globulins bands in African compared with European (Schofield, 1957, Kasper et al, 1970). Other intrinsic host factors may also be responsible as only a minority of apparently normal Nigerians living in Nigeria exhibited this relative abnormal electrophoresis pattern. The possible intrinsic immunologic host factors that may contribute to this phenomenon are currently under investigation in our laboratory.

Stratification on the basis of HAART/ infection duration and HIV clinical status added interesting dimensions to the present study. We compared the SPE pattern in groups defined by the blood CD4 count (CD4 <500 or >500/μl). While no apparent difference was observed in the densitometry pattern in HIV-1-infected subjects naïve to HAART, subjects with better clinical status exhibited a higher γ-globulin bands under effective HAART. The densitometry patterns were remarkable showing evidence of oligoclonal bands on the γ-band against a polyclonal background. This shows that the host ability to resist progression is not a function of HIV clinical status in HAART naïve subjects. Under effective HAART however, improved clinical status sequel to treatment enhanced the host ability to mount humoral immunity against opportunistic infections. Without HAART, they would normally progress to AIDS and the γ-band would decrease and approaches a normal pattern as evident in our previous work (Adedeji et al, 2004).

We similarly compared the SPE pattern in groups defined by infection and HAART duration (Acute or chronic). In this study we considered infection duration or HAART duration less the 12 months as acute while those with greater than 36 months were considered chronic. It is generally difficult to estimate the HIV infection duration precisely (Cohen et al., 2010, Skar et al., 2013). HIV infection date in HAART-naïve subjects was estimated to be approximately two months prior the date they were first tested positive. Since most of them accompanied their spouses to hospital on AIDS-related conditions, the infection date might even been underestimated. Despite this uncertainty, we discovered that acute infection was associated the higher γ-globulin band compared with chronic infection in HAART-naïve subjects. While we were silent on the infection duration before the initiation of HAART because of wide variability, we were able to estimate HAART duration with high degree of certainty as adequate treatment records were available. Unlike the HAART naïve group, chronic HAART was associated with higher γ-globulin band

compared with acute treatment. This shows that the effectiveness of HAART can be estimated from the trend of SPE pattern overtime. We were unable to determine the viral load to compare the extent of viral replication control more importantly in the HAART-naïve group. This would have enabled us to classify our HAART-naïve subjects to either long-term non-progressors or elite controllers (Poropatich and Sullivan, 2011). This would have added a further interesting dimension to the present study.

In the foregoing, we have shown that persistent abnormally high γ-globulin fractions is associated with effective control of HIV disease progression and that humoral responses may have indirect effects on viral replication. Furthermore, the broad appearance of the γ-globulin band suggests polyclonal responses, not only against HIV antigenic determinants but also possible complex antigenic epitopes on opportunistic infectious agents. Apart from these, we also discovered that the host ability to resist progression to AIDS without treatment is not a function of HIV clinical status. In conclusion, the overall results demonstrate the host ability to compensate defective cellular immunity with humoral immune responses in HIV-1 infection. These findings underscore the usefulness of SPE in HIV disease management and identifying individuals that may not progress to full-blown AIDS in the absence of treatment.

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