International Research Journal of Pharmacy and Medical Sciences
ISSN (Online): 2581-3277

Assessment of Follicle Stimulating Hormone (FSH) And Luteinizing Hormone (LH) Levels in Patients Undergoing Haart Regimen in Nigeria

Dr. Victoria Ngozi Mokwenye (Ph.D)
Department of Medical Laboratory Sciences, College of Health Sciences, Igbinedion University, Okada, Edo State, Nigeria.
Email address: veekmok@yahoo.com

Abstract*—The main objective is to establish the effect of Highly Active Antiretroviral Therapy (HAART) on the Follicle Stimulating Hormone (FSH) and Luteinizing Hormone (LH) of young reproductive women during therapy. The research was prospective and targeted at younger reproductive aged women attending University of Benin Teaching Hospital, Edo State, Nigeria, a premier and multi-specialty healthcare provider in West Africa and a referral centre for HIV treatment and control in the southern part of Nigeria and the rest of the country. A targeted population of one hundred (100) HIV seropositive women between 18-40 years old (mean = 29years) starting their first onset of HAART regimen were monitored for nine (9) months after initiation. The FSH and LH were monitored at three (3) monthly intervals and checked at the different phases of their menstrual cycles. Another fifty (50) HIV sero-negative women of same age range (mean = 32years) were used as controls. Pregnant women were excluded. All subjects were recruited at the point of their first awareness of their HIV status. After counselling and with informed consent, their specimens were collected for analysis before they embarked on HAART regimen and monitored thereafter. HIV infection significantly ($p<0.05$) raised FSH and LH in both the Follicular and Luteal phases of their menstrual cycles leading to amenorrhea in some cases. HAART led to significant reductions in the first few months of administration with subsequent increases by the ninth (9th) month, in both phases. This study has shown that while HAART is effective in improving the CD4+ TCell Counts in the sero-positive patients and reducing the raised FSH and LH in the first few months, its ability to control with prolonged administration is doubtful and could explain the adverse menstrual disorders experienced by these category of women*.

***Keywords*—** *HAART, FSH, LH, CD4TCell Count, Follicular Phase, luteal phase*.

I. INTRODUCTION

Human Immunodeficiency Virus (HIV) is a retrovirus with an estimated 33.3million (31.4-35.3 million) people living with the virus at the end of 2009 (UNAIDS, 2010) as compared with 26.2 million (24.6-27.8million) in 1999, a 27% increase. As at 2018, global estimate was 38million (UNAIDS, 2019), shockingly portraying an increase in infections still. As at 2022, the global estimate stands at 39 million (33.3-45.7million) infections with 37.5million (31.8-43.6million) being adults of 15years and above and 53% of the total population as women and girls (UNAIDS, 2023). Nigeria's prevalence rate as at year 2022 stands at about 2.1% (corresponding to about 2million of global estimate) with Akwa Ibom state leading with 5.5% of its population according to Nigerian HIV/AIDS Indicator and Impact Survey (NAIIS, 2023). About 29.8million of the infected global population are assessing anti-retroviral therapy as at 2022 (UNAIDS, 2023).

HIV is a lentivirus (Int. Comm. on Taxonomy) and lentiviruses are noted for long duration illnesses with long incubation periods (Levy, 1993). However, the rate of clinical disease progression varies widely between individuals from two weeks up to twenty years. Many factors can however affect the disease progression, example, the infected person's general immune function (Clerici *et al*.,1996; Morgan *et al*.,2002); age, access to health care and the existence of co-existing infections such as tuberculosis and genetic inheritance such as the CCR5-Δ32 mutation which confers resistance to certain strains of HIV (Tang and Kaslow, 2003). The use of Highly Active Antiretroviral Therapy (HAART) prolongs both the median time of progression to AIDS (Acquired immune deficiency syndrome) and the median survival time.

Women on HAART continue to show diverse menstrual irregularities (Grinspoon *et al*, 1997; Chirgwin *et al*, 1996) and yet nothing much has been done in this area of research. HAART allows the stabilization of the patient's syndromes and viremia but however, neither cures nor alleviates disease. The study of Santoro *et al* (2007) was based on substance-using middle-aged women and established increased levels of follicle stimulating hormone (FSH), Luteinizing hormone (LH) and estrogen (E_2) with HAART. Nothing much is known about the effect of HAART on women who are still undergoing their monthly periods or cycles. Assessment of the hormonal changes and biochemical variations have become inevitable and absolutely necessary in order to initiate the processes for addressing the raised issues. Revelation of the underlying factors would serve as a necessary tool for the women in this African sub-region where the research is carried out and for the generality of women undergoing HAART regimen. This study will also establish the changes in the female sex hormones.

The use of HAART will continue for as long as HIV infections lasts. However, it is a suppressive but not a curative regimen. Changes in endocrinological function including those of the hypothalamic-pituitary-gonadal axis have *been* described for men with HIV infection (Collazos, 2007; Collazos *et al*, 2002) however, similar data in HIV-infected

Dr. Victoria Ngozi Mokwenye, "Assessment of Follicle Stimulating Hormone (FSH) And Luteinizing Hormone (LH) Levels in Patients Undergoing Haart Regimen in Nigeria," *International Research Journal of Pharmacy and Medical Sciences (IRJPMS)*, Volume 6, Issue 6, pp. 40-44, 2023.

women are lacking and this study has been directed on those of reproductive ages and their menstrual irregularities.

Women make up an increasing percentage of HIV-infected patients especially in the sub-Saharan Africa (UNAIDS 2009, UNAIDS 2023) and they have not been adequately represented in clinical studies and there is limited literature and findings in this area of research. This lack of adequate information in the literature as regards data on the effects of HAART on HIV-infected women and menstrual irregularities stimulated this study. The usefulness of the result obtained will help in the management plans of these patients and cannot be overemphasized.

II. Subjects and Methods

This study was done at the University of Benin Teaching Hospital (UBTH) which is a tertiary healthcare institution located in Midwestern part of Nigeria. It has a bed compliment of over eight hundred (800) patients and serves as a referral Centre for four states in the country and beyond. Subjects were female HIV-seropositive women of reproductive age, seen at the hospital clinics. The age range is between 18yrs and 40yrs. (mean 29yrs) in order to eliminate menopausal women.

Ethical standards permission was obtained from hospital management committee before the commencement of study. Only willing patients were included in the study. Adult females who were confirmed HIV-seropositive and referred to the infectious diseases clinic for treatment and monitoring were recruited. The females selected were not above 40years with adequate information taken. Blood samples were collected from these patients on their visits to the clinic. All ethical standards as regards specimen collection were observed. Verbal and written consents were obtained using prepared forms which were attached to their case notes. Information on age, marital status and last menstrual periods (LMP) were obtained. The follicular and luteal phases were calculated using the information from LMP. Blood collection was done using appropriate vacutainers for the various tests required. Blood specimen separation was done with a centrifuge, registered and stored in the refrigerator which was maintained at -70°c and CD4+TCell Count was analyzed using Cyflow SL-3 Green Method. FSH and LH estimations were done using ELISA method of kits from Fortress Diagnostics, United Kingdom. Results were also obtained using the Microplate Reader at the stipulated wavelength.

III. Statistical Analysis

The data were analysed with Statistical Package for Social Sciences (IBM SPSS) software version 23.0. Continuous variables were expressed as means ± standard deviations while categorical variables were expressed in frequency and percentages. Tests of association at different phases were done using analysis of variance (ANOVA) while student t-test was adopted for the association between patients and control. The values are presented as Means ± SEM in bar chart using Microsoft excel. Correlation coefficient was also adopted to establish the strength of relationship between CD_4+ Count Cells and other variables and the level of significance was set at $p<0.05$.

IV. Results

TABLE 1a: Age Distribution of the respondents

Age(years)	Frequency	Percentage
<20	1	1.0
21-25	11	11.0
26-30	50	50.0
31-35	25	25.0
36-40	13	13.0
Total	100	100.0

Mean Age ±SD: Patient – 29.13±4.41; Control – 32.32±4.36

TABLE 1b: Baseline CD_4^+ Count of respondents' age group

Age Group	Mean	Std. Deviation
<20	201.00	0.0
21-25	109.73	94.37
26-30	144.24	88.07
31-35	165.84	102.59
36-40	129.46	76.04

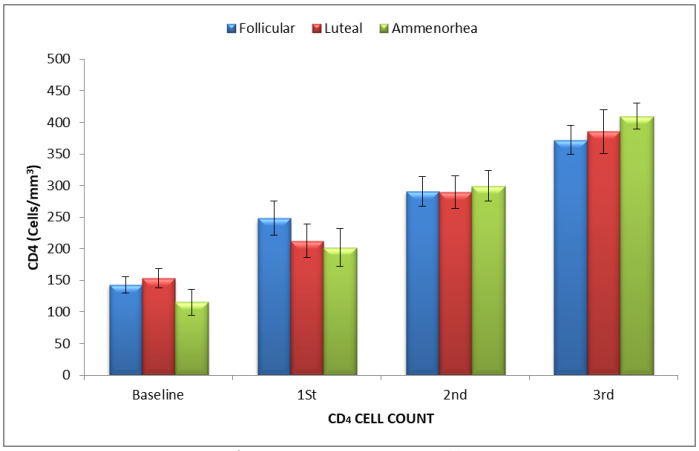

Figure 1: CD_4^+ statistical analysis at different phases

Baseline Pre-HAART HIV patients
1ST FOLLOW-UP AT 3-MONTHS.
2ND FOLLOW-UP AT 6-MONTHS
3RD FOLLOW –UP AT 9-MONTHS

HAART significantly increased CD4+ Count in all phases was recorded to the end of study

TABLE 2: FSH statistical result analysis at all phases

Menstrual Phases		N	Mean	Std. Deviation	Std. Error	P-value
Follicular	Baseline	53	14.13	6.92	0.95	0.002*
	1st	18	10.17	4.32	1.02	
	2nd	26	9.07	4.72	0.92	
	3rd	29	12.20	4.94	0.92	
Luteal	Baseline	38	12.55	8.16	1.32	0.052*
	1st	14	9.07	4.86	1.30	
	2nd	20	7.81	5.70	1.27	
	3rd	23	11.09	4.46	0.93	
Ammenorrhea	Baseline	9	34.51	53.68	17.89	0.711
	1st	10	27.54	24.77	7.83	
	2nd	11	19.52	9.90	2.98	
	3rd	6	20.13	16.46	6.72	

Dr. Victoria Ngozi Mokwenye, "Assessment of Follicle Stimulating Hormone (FSH) And Luteinizing Hormone (LH) Levels in Patients Undergoing Haart Regimen in Nigeria," *International Research Journal of Pharmacy and Medical Sciences (IRJPMS)*, Volume 6, Issue 6, pp. 40-44, 2023.

International Research Journal of Pharmacy and Medical Sciences
ISSN (Online): 2581-3277

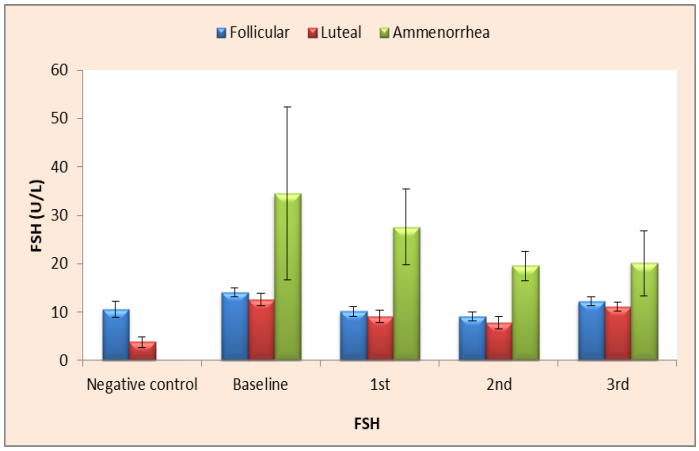

Figure 2: FSH statistical analysis at different phases

Baseline (Pre-HAART HIV patients); 1st (follow-up at 3months); 2nd (follow-up at 6months); 3rd (follow-up at 9months)

TABLE 3: LH statistical result analysis at all phases

Menstrual Phases		N	Mean	Std. Deviation	Std. Error	P-value
Follicular	Baseline	53	8.34	6.83	0.94	<0.001*
	1st	18	8.66	9.86	2.32	
	2nd	26	6.02	2.92	0.57	
	3rd	29	18.50	10.75	2.00	
Luteal	Baseline	38	8.19	4.96	0.80	<0.001*
	1st	14	7.35	4.33	1.16	
	2nd	20	9.17	5.64	1.26	
	3rd	23	18.42	16.75	3.49	
Ammenorrhea	Baseline	9	13.13	13.64	4.55	0.123
	1st	10	31.13	26.97	8.53	
	2nd	11	37.75	27.92	8.42	
	3rd	6	34.82	16.65	6.80	

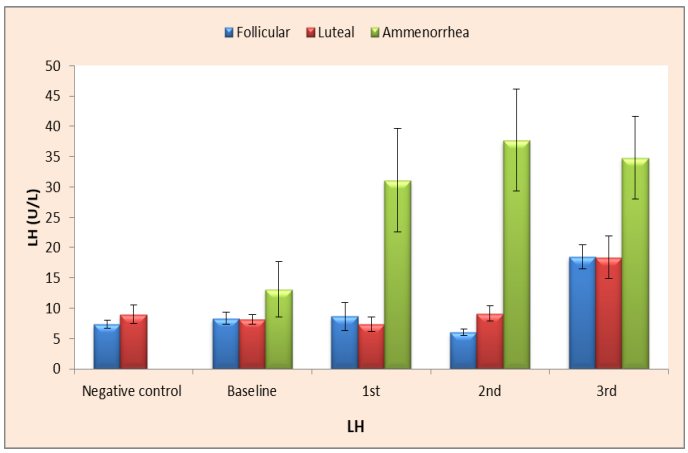

Figure 3: LH statistical analysis at different phases.

Baseline (Pre-HAART HIV patients); 1ST (Follow-up at 3-months); 2nd (follow-up at 6-months); 3rd (follow-up at 9months)

TABLE 4: Correlation matrix showing relationship between CD_4 count cell, FSH and LH across all phases at different follow-up of the participants.

Menstrual Phases			Baseline		1st		2nd		3rd	
			FSH	LH	FSH	LH	FSH	LH	FSH	LH
Follicular	CD4	*r*	0.160	0.251	0.445	-0.035	-0.128	0.255	0.080	-0.079
		p	0.253	0.070	0.064	0.889	0.534	0.208	0.679	0.685
Luteal	CD4	*r*	-0.064	-0.132	0.054	0.079	.485*	0.110	0.204	0.291
		p	0.703	0.428	0.856	0.787	0.030	0.644	0.349	0.179
Ammenorrhea	CD4	*r*	0.429	0.029	-0.043	-0.120	0.538	0.363	0.215	0.360
		p	0.249	0.941	0.907	0.741	0.088	0.273	0.683	0.483

*r = Pearson Correlation; p = p-value; *Statistically Significant.*

There was a weak positive correlation between CD4 and FSH (r = 0.160, p = 0.253) and LH (r = 0.251, p = 0.070) at baseline; FSH (r = 0.445, p = 0.064) at 1st follow-up, LH (r = 0.255, p = 0.208) at 2nd follow-up and FSH (r = 0.080, p = 0.679) at 3rd follow-up respectively. While a weak negative correlation was established between CD4 and LH (r = -0.035, p = 0.889) at 1st follow-up, FSH (r = -0.128, p = 0.534) at 2nd follow-up and LH (r = -0.079, p = 0.685) at 3rd follow-up respectively though all the parameters were not statistically significant for follicular phase of the participants.

V. Discussion

The spectrum of changes observed in this study in the levels of FSH and LH at the different phases and stages of monitoring are of great interest. The use of HAART has shown great improvements in the management of HIV/AIDS. There is an upward increase in the CD_4TCell Count values as therapy continued (fig.1) in compliance with studies carried out in other parts of the world (Lok *et al*, 2010; Mrudala *et al*, 2012; Bowen, R. 2019).

This study targeted at HIV-infected women of reproductive ages, determined the effects of HAART on their FSH and LH hormones at the follicular, luteal and those experiencing amenorrhea, phases in their menstrual cycle. It is evident that HAART disrupts the array of hormone patterns. A study on substance-using middle-aged women related HAART to higher levels of FSH, LH and Estrogen (Santoro *et al*, 2007). In this study, however, FSH that was raised by HIV infection (Fig.2) was significantly reduced by HAART to the sixth month and rose again by the ninth month at the follicular phase (table2 & fig.2). The same pattern of events was made for luteal phase (table.2 & fig.2). The FSH that was raised by HIV infection, was reduced by HAART for about six months only during therapy. It is notable that some of these young women experienced amenorrhea at some time or the other with both HIV-infection or HAART regimen and some regained their cycles with change of HAART combination though. For the amenorrhic women, sharp reductions in FSH obtained within the first six months of HAART started rising by the end of the nine months study period (fig 2). This interesting observation shows that HAART was effective in reducing the FSH for the first six months only while by the ninth month, all phases started experiencing further elevations.

Dr. Victoria Ngozi Mokwenye, "Assessment of Follicle Stimulating Hormone (FSH) And Luteinizing Hormone (LH) Levels in Patients Undergoing Haart Regimen in Nigeria," *International Research Journal of Pharmacy and Medical Sciences (IRJPMS)*, Volume 6, Issue 6, pp. 40-44, 2023.

Luteinizing Hormone (LH), on the other hand, experienced a significant increase with HIV infection in the follicular phase and significant reduction in the luteal phase (fig.3). HAART administration, however, led to significant reductions of LH in the follicular phase within the first six months (fig 3) and an abnormally raised value by the 9th month, way above the control levels. Same sharp increase was observed in the luteal phase while the amenorrhic patients recorded highly raised values to the end of study. The changes observed in the follicular and Luteal phases were highly statistically significant ($p<0.05$).

Gonadotropin-Releasing Hormone (GnRH) produced in the brain and secreted in the anterior pituitary gland, triggers the production of FSH at the follicular phase of the menstrual cycle. In this phase, which lasts for about 12 days from the first day of menstrual bleeding and called the "Proliferation phase", FSH proliferates the lining of the uterus (Losos et al., 2002), stimulating a few ovarian follicles, which compete with each other for dominance. The follicle(s) that reaches maturity forms the ovum and the non-dorminant follicles atrophy and die (Losos et al., 2002). When FSH production is triggered by GnRH, the development and growth of the Follicles are stimulated in the ovary (Follicular Phase), which fosters the growth of a woman's eggs (Cahorean *et al*, 2015). However, elevated levels of FSH above normal ranges are menopausal and disrupts the feedback signals as well. FSH and LH act synergistically in reproduction (Bowen, R., 2019). Fluctuations of FSH at this period in time are bound to have serious repercussions at this stage as the case may be since most of the actions and feedback mechanisms of these hormones are based on their levels.

The luteinizing Hormone (LH), as recorded in this study, was significantly ($P<0.05$) reduced by HAART up to the sixth month of monitoring, but by the ninth month, had shut up to more than twice the baseline value (Fig 3). The elevation of FSH by the ninth month, on the other hand, only tended towards the baseline value.

The mature follicles at this follicular phase secrete increasing amounts of Estradiol (E_2) which stimulates crypts in the cervix to produce fertile cervical mucus. Increasing amounts of E_2 subsequently stops menstrual flow and thickens the lining of the uterus (Losos *et al*, 2002). E_2 further suppresses the production of LH from the anterior pituitary gland but when the egg matures, levels of E_2 reach a threshold above which they stimulate the production of LH through the dual feedback E_2/LH loop in the hypothalamus (Hu *et al*, 2008). There is then a sharp surge of LH about this time that lasts for about 48 hours and called the ovulatory phase (Losos *et al*., 2002), usually about the middle of the menstrual cycle. The produced LH matures the egg and weakens the wall of the follicles in the ovary releasing the ovum in normal circumstances. If fertilization does not occur, the egg disintegrates or dissolves in the fallopian tubes initiating the luteal phase.

Having highlighted expected hormone interplay at this phase and also noting our findings, one can only imagine the series of events that can take place as compensatory measures and how many other systems in the body that might be affected by these events.

The corpus luteum, which continues to grow after ovulation, produces significant amounts of progesterone (Losos *et al*., 2002). This induces production of E_2, suppressing the production of FSH and LH. In the absence of fertilization, the levels of FSH and LH fall quickly overtime and the corpus luteum atrophies, causing sharp drops in the levels of progesterone and E_2. This event triggers menstruation and the beginning of the next cycle. The length of Luteal phase in an individual is normally constant (Weschler, 2002), about 14 days, while the follicular phase fluctuates in length.

In this study, diverse alterations were also obtained on women who experienced amenorrhea at one point or the other. While some recovered from their amenorrhic status and regained normal menstrual cycles, other patients became amenorrhic as HAART progressed. However, there were significant decreases in FSH throughout the study period (fig 2) while LH significantly improved throughout the study period (fig.3) with HAART, for them. This can be considered welcome developments since elevated FSH are characteristic of reproductive cycle cessation. FSH and LH act synergistically in reproduction and there is a bidirectional communication network which links the immune and reproductive endocrine systems (Dorshkind *et al*, 2001; Da Silva,2006) and this makes the observed spectrum of fluctuations of the hormones with HAART in our study most worrisome. FSH and LH directly affects ovulation, production with growth of eggs and menstrual cycles and work synergistically in reproduction. There is a weak correlation between the levels of CD_4TCell Counts and the measured hormones (table 4) depicting that the changes are not quite factors of their improved levels. This study has opened a new page on the swings observed with HAART administration and can easily explain the diverse spectrum of menstrual irregularities experienced by these women.

VI. Conclusion

The administration of HAART will continue as long as HIV infection exists. Although it neither cures nor alleviates disease, it stabilizes patient's syndrome and viremia by improving their CD_4 TCell Counts. With the observed fluctuations in this study on FSH and LH as HAART progresses, I believe that timely interventions can be fashioned out to help these young women who will be tied to these drugs for as long as HIV persists.

Recommendation:

With the revelations achieved in this research, further and extensive studies are highly recommended in order to fashion out adequate hormonal interventions to ameliorate the effects of HAART and improve the well-being and reproduction of these women.

References

[1]. Bowen R. (2019). "Luteinizing and Follicle Stimulating Hormones".www.vivo.colostate.edu. Retrieved 2019-05-06.

[2]. Cahoreau C, Klett D., Combarnous Y. (2015-02-26). "Structure-function relationships of glycoprotein hormones and their subunits' ancestors". Frontiers in Endocrinology. **6**:26
[3]. doi:10.3389/fendo.2015.00026. PMC 4341566. PMID 25767463
[4]. Clerici M, Balotta C, Meroni L, Ferrario, Riva C, Trabattoni D, Ridolfo A. Villa, M., Shearon GM, Moroni M, Galli M (1996) "Type-1 Cytokine production and low prevalence of viral isolation, correlate with long-term non progression in HIV infection." AIDS Res. Hum. Retroviruses 12 (11): 1053 – 1061.
[5]. Chirgwin KD; Fieldjah J, Muneyyurci DO, Landesman S; Minkoff H. (1996) "Menstrual function in HIV-infected women without AIDS" JAIDS 12: 489-494.
[6]. Collazos J. (2007). "Sexual dysfunction in HAART era". AIDS Rev. 9(4): 237-245.
[7]. Collazos J, Mastinez E, Mayo J, Ibara S (2002) "Sexual Hormones in HIV-infected patients; the influence of antiretroviral therapy". AIDS 16 (6):934-936.
[8]. Da Silva JA (2006): "Sex hormones and glucocorticoids: interactions with the immune system". Annals of the New York Academy of Sciences. 876; 102-118.
[9]. Dorshkind K, Horseman ND (2001). "Anterior pituitory hormones, stress and immune system homeostasis". Bioessays 23(3): 288-294.
[10]. Grinspoon S, Carr A (2005). "Cardiovascular risk and body-fat abnormalities in HIV-infected adults". N Engl J. Med 352: 48-62.
[11]. Hu L, Gustofson RL, Feng H, Leung PK, Mores N, Krsmanovic Z, Catt KJ (2008). "Converse regulatory functions of estrogen receptor-alpha and beta subtypes expressed in hypothalamic gonadotropin-releasing hormone neurons". Mol. Endocrinol 22 (10): 2250-2259.
[12]. International Committee on Taxonomy of Viruses 61.0.6 Lentiviruses. National Inst. Of Health.
[13]. Lengi AJ, Philips RA, Karpuzughi E, Ahmed SA (2007). "Estrogen selectively regulates chemokines in murine splenocytes." J. Leukocyte Biol. 81:1065 – 1074.
[14]. Levy JA (1993). "HIV pathogenesis and long-term survival". AIDS 7 (11): 1401 - 1410.
[15]. Lok J. J, Bosch R. J, Benson C.A, Collier A.C, Robbins G.K, Shaffer R.W and Hughes M.D (2010). Long term increase in CD4+ T-Cell counts during combination antiretroviral therapy for HIV – 1 infection. 24(12) 1867-1876.
[16]. Losos JB, Raven PH, Johnson JB, Singer SR (2002). Biology, New York: McGraw-Hill pp 1207-1209.
[17]. Morgan D, Mahe C, Mayanja B, Okongo JM, Lubega R, Whitworth JA, (2002). "HIV-1 infection in Rural Africa: Is there a difference in median time to AIDS and survival compared with that in industrial countries?" AIDS 16(4): 597-632.
[18]. Mrudala ND, Suwarna U.P.,Khadse RK., Minal P., and Shubhangi K. (2012). Statistical Analysis & Evaluation of CD4 Count after 6 months on ART. Indian J. Community Med. 37(4): 266-267
[19]. Nigeria HIV/AIDS Indicator Impact Survey (NAIIS, 2023) https://www.naiis.ng
[20]. Pierce JG, Parsons TF (1981). "Glycoprotein hormones: structure and function". Annual Review of Biochemistry. **50** (1): 465-95. doi:10.1146/annurev.bi.50.070181.002341. PMID 6267989.^ "CGA glycoprotein hormones, alpha polypeptide [Homo sapiens (human)]". NCBI. Retrieved 2 January 2016.
[21]. Santoro N, Lo Y, Moskaleva G, Arnsten JH, Floris-Moore M, Howard AA, AdelG, ZeithlanG, Schoenbaum EE (2007). "Factors affecting reproductive hormones in HIV affected, substances-using middle-aged women". Menopause 14(5); 859-865.
[22]. Tang J, Kaslow RA (2003). "The impact of host genetics on HIV infection and disease progression in the era of Highly active antiretroviral therapy." AIDS 17 (Sullp 4): S51 -S60.
[23]. UNAIDS (2009): UNAIDS report of global AIDS epidemic.
[24]. UNAIDS (2010). "UNAIDS report on the global AIDS epidemic". Genev
[25]. UNAIDS (2020). UNAIDS DATA 2020, 06 July 2020.
[26]. UNAIDS (2023): Global HIV Statistics; Fact Sheet 2023
[27]. Weschler T (2002). Taking charge of your fertility (Revised ed.), New York: Harper Collins. pp 359-361. ISBN 0-06-093764-5.