World Wide Journal of Multidisciplinary Research and Development

WWJMRD 2016; 2(2): 49-52
www.wwjmrd.com
e-ISSN: 2454-6615

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Serum calcium and phosphorus levels of HIV/AIDS positive patients

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Abstract
The management of serum calcium and phosphorus related diseases such as tuberculosis, renal, bone etc. has posed lots of challenges to practicing physicians over the years. HIV/AIDS is another of such that must be accorded with unperturbed attention, especially in developing countries where poverty, gender inequality and unemployment has placed women, youths and children under the most vulnerable group. Serum calcium and phosphorus levels of a total population of 100 subjects (50 HIV positive and 50 HIV negative individuals of age range of**:** 18 years and above) were determined spectrophotomerically using cresophthaline complexone and phosphorus molybdate methods for calcium and phosphorus respectively. Mean calcium was 1.84 (+2SD) mmol/L (HIV positive); 2.06(+2SD) mmol/L (HIV Negative) while phosphorus was 1.64 (+2SD) mmol/L; 1.12 (+2SD) mmol/L for HIV negative and positive respectively at $P < 0.05$. Significant difference in mean values confirms lower serum calcium and phosphorus levels HIV positive subjects than those negative.

Keywords: Serum Calcium, Phosphorus, HIV-AIDS, Nigeria

Introduction
Diseases of calcium, phosphate and skeleton metabolism are among the most common group of disorders that the practicing physicians encounter [1]. They can involve abnormalities in the serum concentrations of bone and abnormalities of the major regulating Organ systems, especially the parathyroid gland, kidney and gastrointestinal (GI) tract. The serum calcium concentration can be abnormally high as in malignancy and primary hyperparathyroidism or abnormally low as it is in renal failure and hypoparathyroidism [2]. There is a loss of focus over time of immune cell function which allows intrusion by several different infectious agents, the result of which is loss of the ability of the body to fight infection and the subsequent acquisition of disease such as pneumonia, tuberculosis, diarrhea, etc. [3]. The ongoing investigations on HIV have provided several observations that revised the initial conclusions about their biological properties, first their presumed preferences to be too limiting a definition since their replication has been reported in macrophages, promyelocytes and more recently in a variety of different susceptible human cells including those of the bowel, skin and connective tissues [4]. AIDS caused by HIV was first clinically observed in 1981 in the United States. Both HIV -1 and HIV -2 are believed to have originated in non-human primates in West – central Africa and have transferred to humans (a process known as zoonosis) in the early 20th century. Infections with HIV occur by the transfer of blood, semen, vaginal fluid, pre-ejaculate or breast milk within those body fluids. HIV is present as either free virus particles or virus within infected immune cells [5]. The biological facts that Women in developing countries like Nigeria are at risk of human Immunodeficiency virus (HIV) infection than male counterpart for biological and socio-cultural reasons with rate of transmission of HIV from male to female is 2-3times higher than from female to male, owing to greater susceptibility of the cervicovaginal tissue to heterosexual contraction of human immunodeficiency virus amplify the high prevalence of non-consensual sex, sex without condom use and the high risk behaviors of sex partners. Gender inequalities, poverty, less access to education and lack of employment opportunities, force many women to commercial sex at a very high risk of human immunodeficiency virus (HIV) infection [6]. The major factors that influence the serum calcium concentration are parathyroid hormone (PTH), vitamin D, the calcium ion itself [7].

Physiological electrolytes include N_a^+ K^+, $Ca^{2+,}$ mg^{2+}, CI, HCO_3, H_2, H_2PO_3 HPO^{2-}_4 and SO^2_4, as well as some organic anions such as lactate, and trace elements. In blood, approximately 50% of the plasma calcium is free, 40% is protein bound, and 10% is complexed. About 80% of protein –bound calcium is associated with albumin and 20% with globulins. Calcium binding is PH dependent, Phosphorous in the form of inorganic phosphate is an important and widely distributed element on the human body, 85% in skeleton, 15% in soft tissue, <0.1% in extra cellular fluid. In the soft tissues, most phosphate is organic or incorporated into cellular macromolecules [8].Multiple biological functions of calcium includes - cell signaling, neutral transmission, Muscle function, Blood coagulation, Activation of enzymes, Formation of skeleton, biomineralisation, and as a component of blood. Phosphorus in the other hand plays an essential part in the basic biochemical mechanism by which energy is obtained for the process of life. It is an essential component of the blood and a constituent of certain enzymes and hormones, which control the working of the body while Phosphorous plays an essential part in the basic biochemical mechanism by which energy is obtained for the process of life. It is an essential component of the blood and a constituent of certain enzymes and hormones, which control the working of the body [9], [2]. Calcium metabolism is a critical health issue among patients receiving dialysis, but it is also quite complicated. When protein concentrations (particularly albumin) fluctuate substantially, total calcium levels may vary, whereas the ionized calcium, whose level is hormonally regulated, remains relatively stable. Thus, total serum calcium concentrations may not accurately reflect the physiologically important ionized (or free) calcium concentration. As an example, in volume overload, chronic illness, and malnutrition or nephrotic syndrome (where serum protein can be reduced), total plasma calcium is low but the ionized calcium is normal. This phenomenon is called pseudohypocalcemia [10].

Calcium and phosphorous occurs as a compound they are important in bone metabolism, they go hand in hand if one goes up the other goes down and vice versa. Calcium phosphate $\{Ca_3 (Po_4)_2\}$ is the principal organic constituent of bones and of bone ash. It occurs as phosphorite $\{Ca_3 (Po_4)_2\}$ in Florida; Tennessee, part of the western United States and northern Africa [11]. Phosphorous is more widely distributed than calcium and also serves as a variety of biological functions [12], [13]. Calcium and phosphorous, and magnesium are transported to blood from bone, renal and GI cells, and vice versa [14, 15,].These transport mechanism can be trans-cellular and around para-cellular. The cellular transport is mediated by the membrane structure and by binding transport proteins [16], [17]. Mineral homeostasis requires the transport of calcium, magnesium and phosphate across their target cells in bone, intestine and kidney: this transport can also be trans-cellular and around cells (pericellular). The pericellular transport is usually diffusional, down a gradient ("downhill"), and not hormonally regulated. Diffusion can also occur through cell channels which can be gated. Transport across cells is more complex and usually hydrolysis or electrochemical gradients and involves membrane structures that are generally termed porters, exchangers, or pumps [18]. For kidney tubules, PTH is the key regulator in a corresponding manner for the transport of phosphate and calcium [19], while for bone; PTH and Ct are the major regulators of cellular calcium and phosphate transport, while vitamin D provides appropriate concentrations of these minerals through its renal and GI actions [20]. Much of the current discourse on infections and drug resistance as it affects sub-Saharan Africa is limited to the pressing problems associated with HIV ,TB and other emerging and re-emerging resistant organisms [21]. HIV infection leads to numerous immunologic abnormalities and eventually to severe depletion of $CD4^+$T cells. As a result, both humoral and cell – mediated immunity is severely depressed in AIDS patients. Since less than 0.1% of the $CD4^+$T cells in an HIV – infected individual are actually infected with the virus, the extensive depletion of T_H cells cell is observed means that uninfected $CD4^+$ T_H cells also are destroyed. Several mechanisms proposed to account for destruction of infected T_H cells involve gp 120, a viral envelope protein that is shed into the blood and lymph of HIV – infected individuals. The binding of soluble gp 120 to CD4 on uninfected cells may lead to their destruction, inhabit their functioning, or interfere with T – cell maturation [22]. Epidemiological data indicates that Human Immunodeficiency virus (HIV) is transmitted mainly through un- protected sexual intercourse (heterosexual or homosexual), transfusion of infected blood and blood products and from an infected mothers to her child (vertical transfusion). Transmissions via injection, drug abuse and other routes seldom occur worldwide. More than 90% of all adolescent and adults HIV infection results from unprotected sex (heterosexual) and over 90% of the infection in children result from mother to child transmission [23]. Measurements of total serum calcium and albumin levels in which parameters of calcium homeostasis were determined in a subgroup of 21 hypocalcaemic AIDS patients showed that Mean serum calcium was 2.34 +/- 0.13 mmol L-1 in the HIV group vs. 2.46 +/- 0.10 mmol L-1 in controls ($P < 0.0001$). After adjusting for serum albumin, hypocalcaemia was present in 6.5% of the HIV group vs. 1.1% of controls. Mean serum calcium was declining according to CDC groups and differed significantly from controls in each group. Regression coefficients of calcium vs. albumin were 0.147 amongst HIV-infected patients and 0.106 for controls. In the subgroup of hypocalcaemic patients with AIDS, 10/21 had vitamin D deficiency, six of these with low ionized calcium levels. Low serum PTH was found in 2/21 patients [24].

Aims and Objectives

This work discusses calcium and phosphorous level of body fluid concentration in HIV positive patients in comparison with HIV negative individuals.

Materials and Methods:

5ml of blood samples was collected each from a total population of 100 subjects consisting of 50 HIV positive and 50 HIV negative individuals of age range of 18 years and above, attending 3rd Armored Division MRS (Hospital), Rukuba Barracks, Jos between February and July 2003. Samples were collected from the anticubital vein via venipuncture between the hours of 8:30am and 12noon. The blood sample was centrifuged at 3,000 rpm for 5 minutes. Serum was dispensed into a clean dry plane tube anaerobically to avoid CO_2.

Calcium and Phosphorus Determination

Calcium was measured using Cresophthaline Complexone method and Phosphorus by Phosphorus molybdate method. These assays were performed spectrophotomerically using kits supplied by Randox (Randox UK) on a SP 1800 UV Spectrophotometer (Pye Unicam, Cambridge England).

Results & Discussion

Table 1: Sum values, means, and standard deviation of HIV positive individuals.

n = 50	**∑x** mmol/L	**x_1** mmol/L	**S_1**
Ca^{2+}	92.03	1.84	0.56
PO^{-}_4	83.55	1.64	0.86

Key:
∑x = Sum of individual values
X = mean
S = Standard deviation

Table 2: Sum values, means, and standard deviation of HIV negative individuals.

n = 50	**∑x** mmol/L	**x_1** mmol/L	**S_1**
Ca^{2+}	103.22	2.06	0.29
PO^{-}_4	55.89	1.12	0.39

Table 3: Significant Test – Comparison of mean and standard deviation of serum Calcium of both HIV positive and negative individuals.

Calculated d value	**Tabulated d value**	**inference**
2.47	1.96	significant

Table 4: Significant Test – Comparison of mean and standard deviation of serum Phosphorus of both HIV positive and negative individuals.

Calculated d value	**Tabulated d value**	**inference**
4.28	1.96	significant

Table 5: The reference ranges for both Calcium and Phosphorous of HIV position and negative individuals.

Group	**Calcium**	**Phosphorous**
HIV positive	0.72 – 2.96	0.03 – 3.31
HIV negative	1.48 – 2.64	0.34 – 1.96

Key:
Reference Ranges = mean + 2S.D
= X + 2S.D

Discussion

The reference value obtained for calcium in the HIV positive individual is 0.72-2.96 mmol/L; and HIV negative individual 1.48-2.6mmol/L respectively (table 5), thus, a significant difference between the calculated "d" value and the tabulated "d" value (table 3), although, the value differ slightly, the lower limits of HIV positive to the HIV negative compare to the established ranges (2.20mmol/L Stanley S. Raphael 1983).

The obtained reference ranges for phosphorous in HIV positive and negative are 0.03-3.31mmol/L and 0.34-1.90 respectively (table 5) also showed a significant difference between the calculated "d" value and the tabulated "d" value (table 3). When this was compared with an established range, the lower and upper limit values are lower and higher than the already reviewed own lower and upper limit (0.81-1.55mmol/L Stanley S Raphael 1983).

Conclusion

Mean calcium was 1.84 (+2SD) mmol/L (HIV positive); 2.06(+2SD) mmol/L (HIV Negative) while phosphorus was 1.64 (+2SD) mmol/L; 1.12 (+2SD) mmol/L for HIV negative and positive respectively at $P < 0.05$. Significant difference in mean values confirms lower serum calcium and phosphorus levels HIV positive subjects than those negative.

Conflict of Interest: None

Ethical Clearance: This was obtained from the 3rd Armored Division Headquarters via the MRS (Hospital), Rukuba Barracks, Nigeria.

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