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A Cross-Sectional Study on the Clinical Presentation of Vitamin D Deficiency at a Family Medicine Specialist Center in Kenya

Mazaher Jaffer[1*], Fatma Abdullahi Ahmed[2], Zamanali Khakhar[3] and Erolls Sigei[4]

[1]Associate Professor of Clinical Medicine at MKU university, at MP Shah and Avenue Hospitals, and HRA clinics in Nairobi, Kenya

[2]Associate Physician (MAHSA, Malaysia), HRA clinic, Nairobi, Kenya

[3]Medical Student at University of Nairobi, Kenya

[4]Associate Professor of Lab Sciences and Microbiology at KMTC, Kenya

ABSTRACT

Background and Aims: The pandemic of vitamin D deficiency bears many an enigma. Clinically, many a primary care clinician prescribes vitamin D to anyone suspect of the deficiency, whilst the exact range of symptoms associated with the deficiency is yet to be fully defined. Newly associated symptoms have been proposed, and the most clinically relevant normal reference range for the deficiency and insufficiency of the vitamin is a question that is yet to be satisfactorily answered in the Kenyan population. The local populations with higher risk despite sunlight exposure is an additional question that warrants further looking into.

Method: Considering the above we conducted a cross-sectional study at a family medicine specialty center in Nairobi over a period of 3 months from 15th April 2023 to 15th July 2021, and the clinical findings of newly proposed symptoms by family physicians to be associated with vitamin D deficiency were correlated to the respective serum vitamin D levels in 168 patients with informed consent. They were then called in 3 months after supplementation to confirm resolution of symptoms through Vitamin D supplementation.

Results: Overall deficiency prevalence was 37%. It was a female-dominated study representing mainly the African and Indian-Asian ethnicity. Symptoms strongly associated were undue fatigue, bowel irregularities, hormone imbalance, myalgia, and arthralgia. Urolithiasis had a weaker association and mood disorders were not associated significantly. Most associations persisted till around the serum level of 75 ng/ml, thereby implying higher optimal levels of vitamin D rather than the current globally accepted reference range.

Definitions: For Vitamin D, 30ng/ml is equivalent to 75 nmol/L

***Corresponding author:**
Mazaher Jaffer (MFM, MSc. ID), Associate Professor of Clinical Medicine, HRA Clinics, Nairobi, Kenya.

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Introduction

Vitamin D deficiency has emerged as a global health concern due to its role in physiological processes such as bone health, immune function, and overall well-being. Its prevalence has led to its classification as a pandemic, with 30-90 % prevalence reported in different continents and regions [1]. Its deficiency has been linked to several health issues, from osteoporosis and rickets to immune system dysfunctions, and has been recognized as a factor contributing to multiple chronic conditions [2]. Multiple individual studies are showing the possible benefits of Vitamin D correction for varying clinical presentations, but the exact clinical utilization of vitamin D and its deficiency correction is not well understood. [3]. Some of these studies report cases with possibly fatal illness due to vitamin D deficiency in children with sudden infant death syndrome [4]. In Kenya, especially in urban environments like Nairobi, vitamin D deficiency was reported with a prevalence of around 70%, with discrepancies in populations such as exclusively breastfed babies [5,6]. We also have some discrepancies with osteopenia despite normal vitamin D levels in elderly in Kenya, suggesting the normal range in use possibly being clinically inappropriate [7].

The prevalence of vitamin D deficiency in Africa, using the reference range of less than 30ng/ml being deficient, is high despite sufficient sunlight exposure. This potentially challenges the concept that vitamin D deficiency is only associated with climates that lack sunlight. [8].

Both plasma 25(OH)D and 1,25(OH)2D levels have shown to decrease with age due to the age-related factors such as reduced capacity to produce vitamin D and intake, decreased sunlight

exposure, and secondary causes [9]. A study in the UK showed that vitamin D deficiency was most common among participants with Asian ancestry (57.2% in winter/spring and 50.8% in summer/autumn), followed by those with Black African (38.5% and 30.8%, respectively), mixed (36.5%, 22.5%), Chinese (33.1%, 20.7%), and White European (17.5%, 5.9%) ancestry respectively. These were despite similar sunlight exposure amongst the groups compared [10].

The most common symptoms found in vitamin D deficiency patients were fatigue 187(55.8%), muscle cramps 131(39.1%), generalized myalgia 125(37.31%), bone and joint pain 111(33.13%) [11]. A meta-analysis done in 2023 found that vitamin D deficiency was associated with suicidal behaviors [12].

Recent clinical discussions about changes to the normal reference ranges for serum vitamin D to make more relevant changes to these thresholds have raised concerns regarding the clinical significance of different levels of deficiency and their associated health risks [13].

Vitamin D naturally exists in two forms: D2 (ergocalciferol), which is derived from yeast and plants, and D3(cholecalciferol), which is derived from the diet by consuming foods high in vitamin D (such as fatty fish and eggs), vitamin D- fortified milk or margarine, and/or multivitamins [14].

With the above there is an evident need to identify the high-risk populations, the clinical presentations and the clinically appropriate reference normal reference ranges for serum vitamin D levels.

Objectives

Broad Objective

To analyze the prevalence and stratify risk of patients with vitamin D deficiency attending the HRA clinic in Nairobi over a three-month period (15th April 2023 to 15th July 2023) and to further determine the association of clinical symptoms proposed to be associated to vitamin D deficiency at specific categorized levels of serum vitamin D.

Specific Objectives

- To quantify prevalence of vitamin D deficiency in the population according to both old and newly proposed reference ranges of Vitamin D deficiency levels.
- To stratify the demographic risk of vitamin D deficiency in the population being studied.
- To identify clinical presentations associated with the varying levels of vitamin D deficiency.
- To propose a more clinically relevant normal vitamin D reference ranges based on patient clinical outcomes.

Ethical Considerations

Ethical clearance was obtained from KMTC (Kenya Medical Technical College)'s ethical review committee (ERC) for the research. NACOSTI (National Commission of Research Technology and Innovation) certification was then attained with the research license number: NACOSTI/P/23/24166. Written consent was taken from the participants who agreed to be part of the study. The data was collected in a confidential registry with unique identification numbers allocated participants that was not revealed to the staff working on their laboratory result for blinding purposes. This document was then destroyed after the study.

Methodology

A cross-sectional study evaluated the prevalence and symptomatology of Vitamin D deficiency among patients presenting with symptoms aligning with this deficiency. This study was conducted at the HRA (Health Risk Assessment) Clinic in Nairobi, a private specialist family medicine physician clinic in Nairobi. The Vitamin D levels were tested using the immunofluorescence immunochemistry technique through the I-chroma immunofluorescence machine as per their recommended standard operating procedures having a sensitivity and specificity of 96% and 94%, respectively [15].

Exclusion criteria were anyone who had taken vitamin D supplements over the last 1 month, and anyone who had a proven malabsorption syndrome, chronic kidney disease or anyone with a genetic disorder that would interfere with Vitamin D metabolism. Informed consent was sought from each participant prior to enrolling the patients in the study.

A non-probability sampling method was used, specifically convenient sampling, as participants were selected based on their availability at the clinic during the study period. The sample size was calculated using the Fischer's formula leading to a minimum sample size of 103 patients [16].

Venous blood samples were collected in a test tube with a clot activator and centrifuged. Separated serum was stored in cryovials with light protection to prevent disintegration until they were processed for serum vitamin D levels by immunofluorescent chemistry within 24 hours.

Over the period of three months (from 15th April 2023 to 15th July 2023), the clinic gathered information on the participants' presenting symptoms and laboratory findings. Informed consent was obtained from each participant to use their medical information for research purposes. During each patient's initial visit, serum Vitamin D levels were assessed through phlebotomy for the collection of blood samples, to determine deficiency or sufficiency status. The set of symptoms they presented with was also listed, with special focus on the symptoms that were assumed to be attributed to vitamin D deficiency. These were: undue fatigue, myalgia, incomplete bowel emptying, history of urolithiasis, pseudogout/arthralgia, hormonal imbalance, and mood disorders. The symptoms were defined as follows:

Undue fatigue was defined as feeling tired even after a good night sleep persistently for more than 10 days without an attributable cause, myalgia was defined by muscle pains that were not attributable to significant exertion, incomplete bowel emptying was a subjective feeling of heaviness in the bowel even after passing stool that would be accompanied by excessive flatulence and gastro-esophageal reflux disorder, urolithiasis was confirmed through previous urinalysis or imaging of the kidney, ureter and bladder (KUB ultrasound or CT scan) in the past, arthralgia was defined by joint pains that were not attributable to significant exertion or injury , hormonal imbalance was defined through presence of irregular, painful or unusually heavy periods and in men evidence of low testosterone levels, and mood disorders were defined by a PHQ-9 score and GAD-7 score of more than or equal to 10 [17-21].

Osteopenia and osteoporosis were excluded from these associations that was already known. The results of the vitamin D levels were classified between <15 ng/ml, 15-30 /ml, 30-45 ng/ml, 46-60 ng/ml, 61-75 ng/ml and above 75 ng/ml.

Patients were then given supplementation of their vitamin D levels with 60,000 IU weekly and were followed up over 3 months after supplementation regarding the improvement or complete resolution of their symptoms to confirm that their symptoms reported were fairly attributed to the deficiency and not due to any other cause [22].

Data was analyzed using Microsoft Excel. Correlation was derived using the Linear Correlation Coefficient (Pearson's Formula), Odds ratios, Line graphs and Pie charts were derived to show trends of relevant data for correlation. The CRISP reporting checklist tool was used to write this report [23].

Results

A total of 168 out of the 400 patients who attended the clinic consented to partake in the study. Participants comprised of male (76) and female (92) patients, with a predominance of female patients. Among the participants, 94 were Indian-Asian, 67 were African and 7 were other ethnic backgrounds. An overall prevalence of the deficiency of 37% was found. All participants were residing in Kenya and were evaluated upon their visit to the clinic. The loss to follow up for the review calls in 3 months was 8%.

Trends Found Between Vitamin D Deficiency (Old and New Reference Range) and Gender

Of all who attended, around 90 participants were in the equivocal range of vitamin D between the new proposed normal range and the old proposed normal range. These had relatively equal representation of both females and males. The deficient group as per the old normal reference range had almost two times the number of females than males, and the sufficient group has the smallest number with equal representation of males and females.

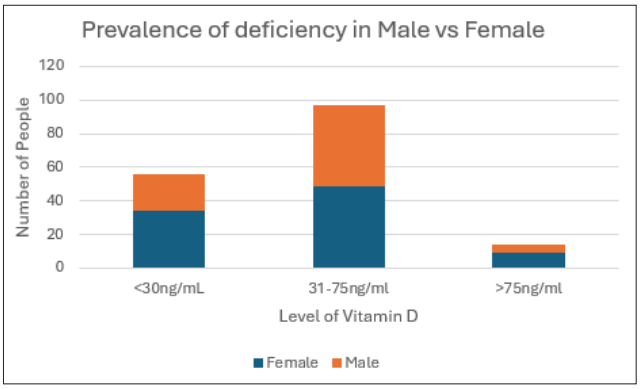

Figure 1: Prevalence Categorized Between the Old and Higher Normal Ranges of Different Genders

Trends found between Vitamin D deficiency (old and new reference range) and Age

Most patients who participated in the study were above the age of 50, and the trend from the population was progressive in representation of age groups from young to old. The old normal reference range would have missed most deficient patients (around 65%) who were in the newer deficiency range, and this effect was most pronounced in the over 50-year-old patients. The sufficient level patients also had a proportionately higher number in the same group.

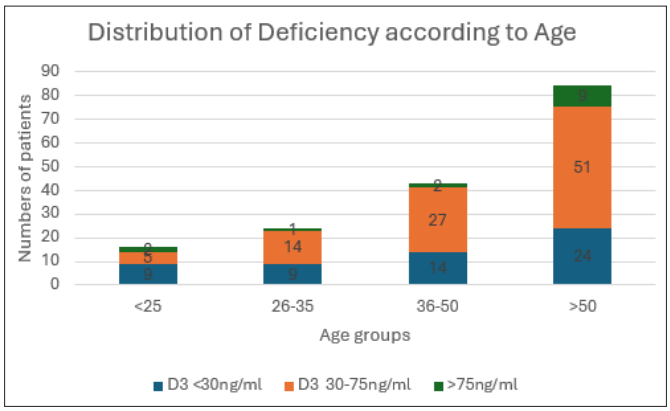

Figure 2: Prevalence Categorized Between the Old and Higher Normal Ranges of Different Age Groups

Trends Found Between Vitamin D deficiency (Old and New Reference Range) and Ethnicity

The population being studied had a far wider representation of the Indian-Asian and African ethnicities than any other. A similar trend is found in this population where the largest numbers are found in ranges between the old and the new proposed reference ranges, and the smallest are those with sufficient levels. The other ethnicities too had the same trend but with such large numbers there is a chance of missed irregularities.

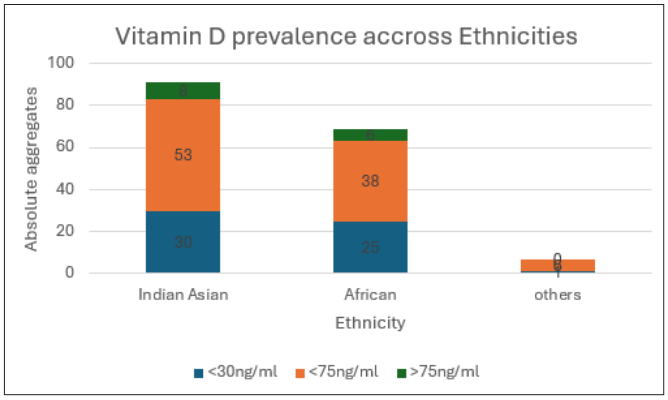

Figure 3: Prevalence Categorized Between the Old and Higher Normal Ranges of Different Ethnic Groups

Correlation Between Vitamin D Deficiency and Each Symptom Attributed to it

Each proposed associated symptom that was proposed by some family medicine clinicians in referral hospitals in Kenya to have been possibly associated with vitamin D deficiency was correlated with the prevalence at each category of level of deficiency. A linear correlation co-efficient by Pearson's method was made toward sufficiency and not deficiency, thereby meaning that the closer a positive correlation was found, the closer the value would be to -1.0, and an inverse correlation or antagonistic relationship. The trends give an idea when the relationship changes in the trend to allow us to estimate the correct normal reference ranges for vitamin D deficiency regarding the symptom in question. In some cases, the trends persisting till levels above normal ranges may be explained by the presence of other factors that led to the symptom persisting. The odds ratio of each symptom would indicate the strength of the correlation such that the higher the odds ratio above 1, the stronger the correlation. The percentage proportion of reversibility by supplementation of vitamin D alone was probably the strongest measure confirming causality and

indicates some of these symptoms were solely due to vitamin D deficiency in specific cases. The symptoms with their linear correlations were as follows:

Undue Fatigue

With an Odds Ratio of 3.7, a linear correlation coefficient at vitamin D levels below 75 ng/ml of -0.987, and Linear correlation coefficient at vitamin d levels above 75ng/ml: 1 – there was a very strong association between this symptom and low vitamin D deficiency up to 75ng/ml and this was not the case after this level. The confirmation was reinforced by the percentage that resolved upon supplementation within 3 months of 95%.

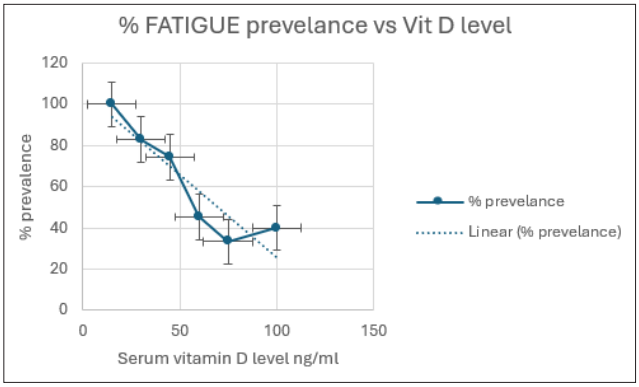

Figure 4: Prevalence of Fatigue at Different Levels of Serum Vitamin D Concentration

Myalgia

Myalgia had an odds ratio(OR) of 1.9 with vitamin D deficiency, a linear correlation coefficient(CC) at vitamin D levels below 75 ng/ml of 0.7592497, and Linear correlation coefficient at vitamin d levels above 75ng/ml: -0.82345 – there was a very strong association between this symptom and low vitamin D deficiency even after the 75ng/ml implicating that there are many other factors effecting it that led to its continuity even upon reaching higher levels then the proposed normal levels. There was however a high percentage (90%) that reported significant improvement even without resolution of the symptoms within 3 months through the supplementation.

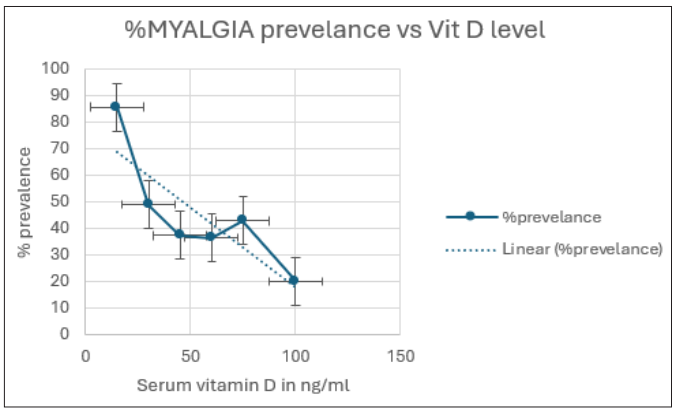

Figure 5: Prevalence of Myalgia at Different Levels of Serum Vitamin D Concentration

Arthralgia

There seemed to be a weak association between vitamin D deficiency and arthralgia with only an OR of 0.91, and a CC at below 75ng/ml of -0.48. There was a interesting trend here though in the CC being very much stronger (-0.89) at levels below 30ng/ml (the old reference range), and there was a reversal of association above the range of 75ng/ml. The percentage resolution after 3 months was also comparatively low at 45%, likely due to osteoarthritis being prevalent in damaged joints after recurrent pseudo-arthritic reactions.

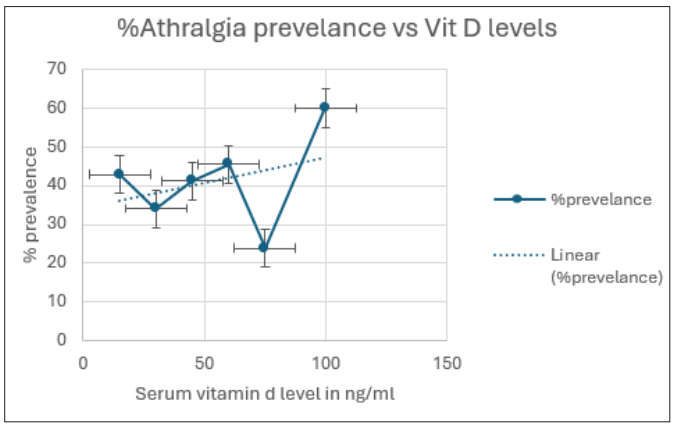

Figure 6: Prevalence of Arthralgia at Different Levels of Serum Vitamin D Concentration

Urolithiasis

Since about 70% of kidney stones had calcium origin in some studies of the past (17), vitamin D deficiency would be expected to be associated with kidney stones. However, with an increase in sufficiency, there was an increase in prevalence, contrary to the findings. This may be because other stones exist such as uric acid stones from patients with hyperuricemia that have also increased prevalence globally after 2015(18). Despite this, an overall weak association was found with OR 0.98 and a percentage resolution of recurrent urolithiasis of only 15% since most cases had not had recurrent symptoms more frequent than 3 months apart.

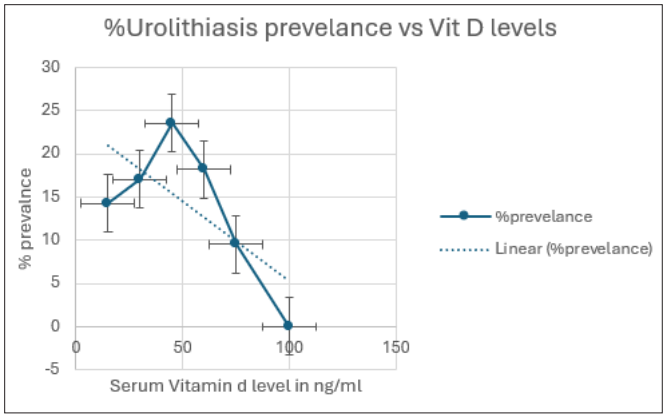

Figure 7: Prevalence of Urolithiasis at Different Levels of Serum Vitamin D Concentration

Mood Disorders

The mood disorders that were examined were specifically anxiety and depression and patients were not on prior medication. However, the factors that would be able to affect mood were too variable in the population and it being done in a clinic setting with chronic fatigue and unexplained pains in many of the study population, it was doubtful if the chronic symptoms led to the mood disorder or there was a correlation directly with the vitamin deficiency. None the less, a statistically insignificant odds ratio of 1.02 was found with a borderline CC below 75ng/ml of -0.42 that extended normal levels in the same way making it hard to be sure of association. The percentage resolved in 3 months also was quite insignificant. The association is therefore unlikely as per this result.

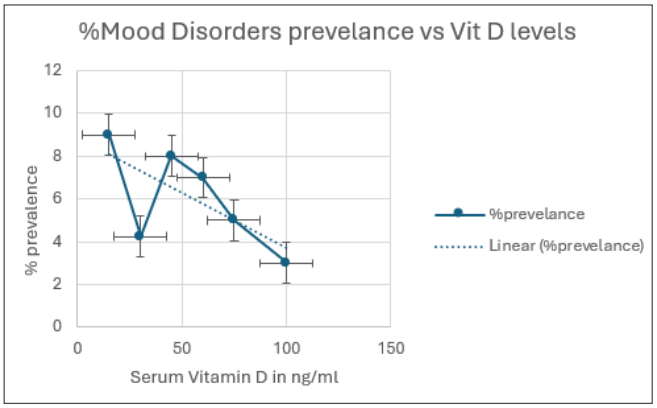

Figure 8: Prevalence of Mood Disorders at Different Levels of Serum Vitamin D Concentration

Hormonal Imbalance

Period irregularities, and hormonal imbalance was seemingly very much affected by the deficiency of vitamin D, with OR of 1.6, CC at levels below 75ng/ml of -0.81, and above 75 till 100ng/ml of 1. The % that resolved in 3 months was less than other stronger associated symptoms but was still significant at 65%. This again may be attributed to other factors that can play a role in effecting the hormone balance especially in females.

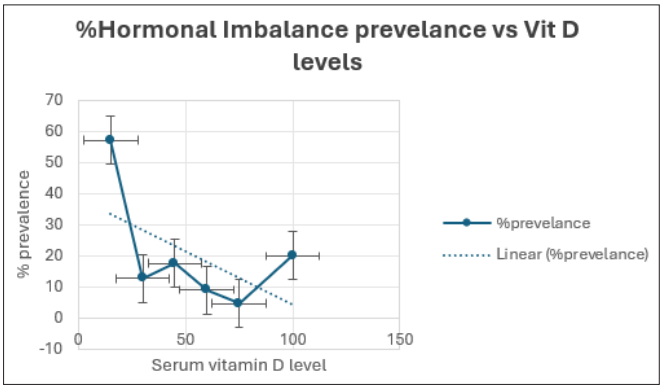

Figure 9: Prevalence of Hormonal Imbalance at Different Levels of Serum Vitamin D Concentration

Incomplete Bowel Emptying

A strong correlation of bowel irregularities with vitamin D deficiency was found, which was not proportional to the mood improvements so if stress was a factor to IBS, this mechanism cannot explain the bowel improvement in the population. We found an OR of 3.2, CC at levels less than 75ng/ml of vitamin D of -0.92, and above 75ng/ml of +0.95. The percentage with symptoms resolution by supplementation alone in 3 months was 67% - again likely to be affected with the need for avoidance of dietary insults that was in commenced by the patients to experience the benefit properly.

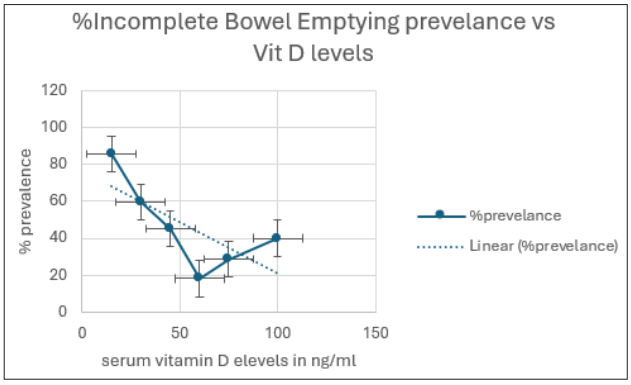

Figure 10: Prevalence of Fatigue at Different Levels of Serum Vitamin D Concentration

Discussion

The prevalence of the deficiency at the current reference range of above 30ng/ml was not very high and was more prevalent in females than males. This tendency is reduced through the 90 additional patients within the range below 75ng/ml, meaning that higher ranges would confirm the occurrences have less gender bias. More elderly people participated in the study, but the percentage who were considered deficient by percentage would increase about threefold in each group, except for the under-represented age group of those below 25 years of age. Ethnicity was not representative of the general population in Kenya as a risk factor because the clinic mainly attended to ethnicities of Indian- Asian as well as African origin in their regular patients seen. The deficiency seemed equally ethnically distributed in the patients, proportional to their attendance rate at the clinic.

The association between the proposed symptoms and the vitamin deficiency was tested in three ways. These were: the odds ratio between the prevalence of the symptoms and the level of vitamin D, secondly a Pearson's linear correlation co-efficient between the levels of vitamin D and the presence of symptoms and thirdly to reinforce association without the effect of confounding factors, the correction of deficiency was followed up for reversal of the same in 3 months, and the percentage of this was used as an indicator strongly toward association to the deficiency.

The Linear correlation curve would confirm a positive correlation in proportion to the negative gradient since it was measured in comparison to the level of vitamin D present rather than deficiency. The change point of the overall gradient was also an indicator of the approximate serum vitamin D level that should be used for clinically normal reference ranges in the population.

Table 1: Summary of Associations Calculated by Different Methods Between Symptoms and Deficiency

Symptom	Fatigue	Bowel Irregularities	Myalgia	Hormone Imbalance	Arthralgia	Urolithiasis	Mood Disorder
Odds Ratio	3.7	3.2	1.9	1.6	0.91	0.98	1.02
CC < 30	-0.97	-0.96	-0.91	-0.97	-0.89	0.95	-0.94
CC < 75	-0.98	-0.92	-0.76	-0.81	-0.45	-0.98	-0.41
CC 75-100ng/ml	1	0.95	-0.90	1	1	-0.75	-0.63
Symptom Reversal %	95%	67%	92.5%	65%	45%	15%	7.5%

By the methods above, it's quite apparent that there is a strong correlation between unexplained fatigue, hormone imbalance, arthralgia, and bowel irregularities. The association with myalgia and urolithiasis was not as strong as expected, likely because of the possibility of multiple other factors causing the same symptoms. Mood disorders were very hard to assess accurately, with a very low prevalence amongst subjects, recall bias, and relative subjectivity of the information gathered that led to an unsatisfactory representation of the possible association. As per the result, there was no correlation.

In the symptoms with a strong association, the reversal point was deficient above 30ng/ml vitamin D serum concentrations and referred to levels above 60ng/ml or even 75ng/ml being more appropriate for clinical use as the normal reference range.

Conclusion and Recommendations

The study confirms an association between some previously unrelated symptoms and the deficiency of vitamin D that should be considered in clinical practice and further suggests that a higher normal reference range is appropriate for clinical use in the population studied.

A larger-scale study with more representation from other ethnic groups and countries, especially as a prospective cohort study, would help confirm the findings.

Limitations

The data collected without double blinding and the application of convenience rather than random sampling may introduce a selection bias. The new list of potentially associated symptoms was not exhaustive and other additional postulated symptoms could be added to this list through similar studies in the future. The absence of significant representation from the population of ethnicity other than African or Indian Asian, and the female-predominance of the samples, limit the generalizability of the findings to the overall population in Nairobi. The lack of a past medical history defining the exposures to vitamin D supplements more than 6 months prior to the study, dietary information, genetic predisposition (alluded to by a family history) and other variables prevent understanding of the possible factors leading to the deficiencies detected may allow us to miss some etiological insights to the findings. Patients with liver and kidney disease were not actively screened for and excluded from the sample, though there was no clinical evidence history of the same report. Finally, the upper limit of the normal reference range that would lead to toxicity thereafter was also not investigated in this study.

Acknowledgments

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