

The Association between Vitamin D and Frequency of Streptococcal pharyngitis among School Students

Mohamed A. Fareid

Botany and Microbiology Dept., Faculty of Science, Al-Azhar University, Nasr City, Cairo, Egypt.

* Present address: Basic Science Dept., Health Colleges, H'ail University, Saudi Kingdom.

mohamedfareid73@yahoo.com

Abstract: The aim of the present study was to assessment the relationship between Vitamin D deficiency and Streptococcal pharyngitis among school students. Our study showed that, the proportion of vitamin D deficiency in females was significantly higher than the prevalence in males. It recorded 122 (64.89%) and 66 (35.10%), respectively. The frequency of Group A Streptococcal (GAS) isolate recorded the most higher percentage between enrolled students which have deficient vitamin D level in compared to sufficient vitamin D level. It recorded 115 (73.24%) and 31 (49.20%), respectively. Statistically, there is a higher significant different between vitamin D deficiency and frequency of GAS isolates, p-value < 0.05. Also, our data showed a higher significant positive correlation between vitamin D levels and the frequency of GAS ($r = 1.000^{**}$, p-value = 0.001).

[Mohamed A. Fareid. **The Association between Vitamin D and Frequency of Streptococcal pharyngitis among School Students.** *Nat Sci* 2019;17(3):81-91]. ISSN 1545-0740 (print); ISSN 2375-7167 (online). <http://www.sciencepub.net/nature>. 13. doi:[10.7537/marsnsj170319.13](https://doi.org/10.7537/marsnsj170319.13).

Keywords: Association, Vitamin D, Frequency, Streptococcal pharyngitis.

1. Introduction

Acute pharyngitis is a common illness in children and adults and its etiology include a wide variety of microbial agents. Group A Streptococci (GAS) are the most frequently isolated pathogens in acute pharyngotonsillitis cases in school-aged children. In children, approximately 20% of pharyngitis cases are caused by GAS (DuBose, 2002). Streptococcal sore throat is one of the most common bacterial infections of childhood. GAS is responsible for the great majority of such infections and frequently colonizes in the throat of an asymptomatic person. Pharyngeal carriage rates among normal school children vary with the geographic location and season of year. Among children, asymptomatic carriage rates of 15-20 % have been noted in several studies (Bisno and Stevens, 2000).

Vitamin D, the sunshine vitamin, has received a lot of attention recently as a result of associating vitamin D deficiency with many acute and chronic illnesses including disorders of calcium metabolism, autoimmune diseases, some cancers, type 2 diabetes mellitus, cardiovascular disease and infectious diseases. Vitamin D is not just a vitamin; it is also a prohormone with numerous functions in the body (Holick, 2004). "Prohormone" refers to a group of fat-soluble steroids hormones. The two major forms are vitamin D₂, or ergocalciferol, and vitamin D₃, or cholecalciferol (Norman *et al.*, 2007). It is well-established that vitamin D plays an important role in bone mineralization of the skeletal system. Serum levels of vitamin D may differ according to racial differences and seasonal changes. Afro-Americans and Hispanics have been reported to have lower vitamin D

levels than Caucasians (Esteite, *et al.*, 2010). In the winter season, lower amounts of vitamin D synthesis occur in the skin (Nseir *et al.*, 2012; Aydin *et al.*, 2011).

In addition, vitamin D has a critical role in the production of surface anti-microbial peptides (AMPs), which plays an important role in innate immunity (Reid *et al.*, 2011). These peptides have a wide spectrum antimicrobial activity and directly prevent proliferation of microorganisms in a tissue (Brogden, 2005). Anti-microbial peptides are not only produced from neutrophils, but also produced from macrophages and natural killer (NK) cells. Also, they have been shown to play a critical role in the respiratory defense system (Brogden, 2005; Ball *et al.*, 2007).

The major cause for vitamin D deficiency is the lack of appreciation that sun exposure has been and continues to be the major source of vitamin D for children and adults of all ages. Vitamin D deficiency among children has become a major health problem worldwide (Taheri *et al.*, 2014). Previous research findings have indicated that vitamin D deficiency is common among children in Western countries (Gordon *et al.*, 2008; Dyson *et al.*, 2014). Some studies examining vitamin D deficiency in Saudi Arabia have demonstrated a high prevalence of vitamin D deficiency among children and adolescents (Mansour and Alhadidi, 2012; Al-Ghamdi *et al.*, 2017). The present study aimed to assessment the relationship between Vitamin D deficiency and Streptococcal pharyngitis among school students.

2. Materials and Methods

A school-based cross-sectional study was conducted in Hail province, Saudi Arabia between September 2016 and March 2018. As the study aimed to assess the vitamin D deficiency as well as the association between vitamin D deficiency and Streptococcal pharyngitis among school students, a sample of the population was required, thus school-based survey was found to be a relevant approach to reach all age groups.

2.1. Data collection

A standardized and pre-tested questionnaire was used to collect data about background characteristics including age, gender, skin color, school level for students. Participants were classified into low income (those living in primitive houses), intermediate income (those living in apartments), and high income (those living in villas). Participants were also asked about their life style (exposure to sun and physical activity), as well as their nutritional habits (intake of multivitamins, intake of Omega 3, type of milk products consumed, carbonated soft drink consumption and body mass index).

2.2. Collection, transportation and processing of pharyngeal swab

Pharyngeal swabs samples were collected from the pharynx using sterile cotton swabs. Necessary care was taken not to swab the cheeks, tongues, lips or other areas of the mouth. The swabs were placed immediately in Amie's transport medium (Oxoid, England) and transported to Microbiology Laboratory and processed within 2 hours of collection (Carroll and Reimer, 1996; McDonald, *et al.*, 2006). Then, the pharyngeal swabs were inoculated onto 5 % sheep's blood agar plates and incubated for 24 hours at 37 °C in a candle jar, which can provide an atmosphere of 5 % CO₂. Culture plates negative for β-haemolytic colonies were incubated for additional 24 hours to allow the growth of slow growers. Identification of GAS isolates was made based on the standard microbiological techniques which include β-haemolytic activity on sheep's blood agar, small colony characteristics, Gram positive cocci, catalase production negative, 0.04-U bacitracin disc susceptible and PYR positive (Hardy Diagnostics, USA) tests (Carroll and Reimer, 1996; Cheesbrough, 2002; Brahmadathan and Gladstone, 2006).

2.3. Lancefield Grouping of Streptococci

The procedure of the manufacture company (OMEGA D. Ltd.) has been employed, as following: In a clean test tube, 0.3 ml of extraction enzyme has been placed; the five colonies have been suspended by using a loop. This suspension has been incubated in a water bath at 37 °C for 10 minutes (shaking after 5 min.), then by using pasture pipette, one drop of the extract has been added to each of the circles on the test

card. After that one drop one each latex reagent has been added also and mixed well by using a clean mixing stick each time. The card has been rotated gently for up to one minute to check the appearance of agglutination in corresponding group. Positive reaction was detected by red clumps of a green background (Facklam, 1980).

2.4. Vitamin D assessment

To assess vitamin D serum level, 5 ml of blood was collected from each individual as clotted sample and was kept on ice until centrifugation on the same day. Serum was kept in aliquots at -20°C until analysis. Serum levels of 25-hydroxyvitamin D were measured using chemiluminescent immunoassay (Liaison 25 OH Vitamin D Total Assay; DiaSorin, Stillwater, MN, USA). This analysis method measures the total vitamin D in the range of 10-375 nmol/L. The sensitivity of the assay is <10 nmol/l and the intra- and inter-assay coefficients of variation were 5% for the intra-assay and 10.4% for the inter-assay. We adopted the Institute of Medicine cutoff points for vitamin D levels (Ross *et al.*, 2011; Rosen *et al.*, 2012), classifying serum levels into sufficient (50-125 nmol/L), insufficient (25-50 nmol/L), deficient (<25 nmol/L), and toxic (>250 nmol/l).

2.5. Statistical analysis

To describe the study population and vitamin D serum level, we used frequencies and proportions for the qualitative variables for the quantitative variables. Significant differences in proportions of vitamin D deficiency in various population subgroups were assessed using Chi-square test. Pearson's correlation was done in order to assess correlation between vitamin D deficiency and the frequency of Streptococcal pharyngitis among school students. A *p*-value of <0.05 was defined as the level of significance. We used Statistical Package for the Social Sciences (SPSS Inc., Chicago, IL, USA), version 15 for data analysis.

2.6. Ethical considerations

This study was approved by the ethical committee of the College of Medicine (University of Hail, KSA) Research Board. All collected data were kept confidential. Participants were informed on the risks and benefits and their right not to provide information, or to withdraw from the study at any time without any sort of penalty. An informed consent was obtained from students' parents.

3. Results

The present study was conducted for a period from September 2016 to March 2018. A total of 350 participants with an average age 10.33 ± 3.68 , consisting of 172 (49.14%) males and 178 (50.85%) females were included in this study (Fig 1).

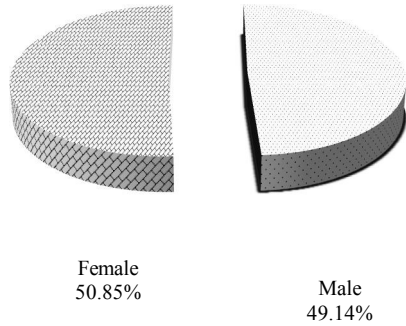


Fig (1): Study participants

males (49.14%) and females (50.85%), statistically there is no significant difference between the percentages of male and female, p-value recorded 0.748. Their ages ranged from 6 to 19 years. Mostly (52.86%) had light brown skin color, approximately one-third had white skin color (27.43%) and very few (7.14%) had black skin color. Most of the study participants were from the intermediate school (43.14%), and the rest were distributed between the primary school (18.86%) and secondary school (38.00%). Furthermore, approximately 50% were of high economic level, 36.29% were of intermediate economic level, and 14.57% were of low economic level. Statistically, there is a highly significant difference between the different age, skin colors and different economic levels between enrolled students, p-value < 0.05.

3.1. Characteristics of study participants

As shown in table 1, 350 school students were enrolled in the study with almost equal proportion of

Table (1): Characteristics of study participants

Characters		No. (%)	P- value
Gender	Male	172 (49.14)	0.748
	Female	178 (50.85)	
Age	6-9	48 (13.71)	0.000
	10- 14	89 (25.43)	
	15-19	213 (60.86)	
Skin color	White	96 (27.43)	0.000
	Light Brown	185 (52.86)	
	Dark Brown	44 (12.57)	
	Black	25 (7.14)	
School level	Primary	66 (18.86)	0.000
	Intermediate	151 (43.14)	
	Secondary	133 (38.00)	
Economic level	Low	51 (14.57)	0.000
	Intermediate	127 (36.29)	
	High	172 (49.14)	
Total		350 (100%)	--

3.2. Vitamin D deficiency among school students by background characteristics, life style, nutritional habits and status.

Table 2 showed the vitamin D deficiency among school students by background characteristics, life style and nutritional habits and status deficiency, life style, nutritional habits, and status among school students. The proportion of vitamin D deficiency in females was significantly higher than the prevalence in males. It recorded 122 (64.89%) and 66 (35.10%), respectively, p-value < 0.05. Daily exposure to sun for at least 20 minutes, being physically active, not drinking carbonated soft drink and taking multivitamin

supplements had lower risk of vitamin D deficiency. Vitamin D deficiency was significantly higher with the decrease in BMI, where the normal weight students had prevalence of 85 (46.44%) as compared to 68 (37.15%) in the overweight and 30 (16.39%) in obese students, p-value < 0.05. Students with light brown skin color, and those not taking omega 3 supplements and multivitamins appeared to be at higher risk of vitamin D deficiency, they recorded 95 (53.37%); 152 (85.39%) and 107 (59.77%), respectively. Moreover, there was significant difference between vitamin D deficiency and the type of milk products consumed and physical activity, p-value < 0.05.

Table (2): Vitamin D deficiency among school students by background characteristics, life style, nutritional habits and status

Variables	Vitamin D Deficiency No. (%)	P- value
Gender		
Male	66 (35.10)	0.000
Female	122 (64.89)	
Age (Years)		
6- 9	20 (10.58)	0.000
10- 14	46 (24.33)	
15- 19	123 (65.07)	
Skin color		
White	47 (26.40)	0.000
Light brown	95 (53.37)	
Dark brown	20 (11.23)	
Black	16 (8.98)	
Economic level		
Low	29 (16.38)	0.000
Intermediate	66 (37.28)	
High	82 (46.32)	
Daily sun exposure		
More than 20 min.	128 (39.62)	0.000
Less than 20 min.	195 (60.37)	
Physical activity		
Yes	71 (37.96)	0.000
No	116 (62.03)	
Milk product		
Skimmed	45 (24.75)	0.034
Low Fat	64 (35.16)	
Full Fat	73 (40.10)	
Carbonated soft drink		
Yes	151 (85.31)	0.000
No	26 (14.68)	
Multivitamins		
Yes	72 (40.22)	0.008
No	107 (59.77)	
Omega 3		
Yes	26 (14.60)	0.000
No	152 (85.39)	
Body mass index		
Normal	85 (46.44)	0.000
Overweight	68 (37.15)	
Obese	30 (16.39)	

3.3. The association between Vitamin D level and frequency Streptococcal isolates

Table 3 represents the association between vitamin D level and frequency Streptococcal isolates. The frequency of Group A Streptococcal isolate recorded the most higher percentage between enrolled students which have deficient vitamin D level in compared to sufficient vitamin D level. It recorded 115 (73.24%) and 31 (49.20%), respectively.

Followed by Group B, C and D Streptococci which recorded also a higher percentage between deficient vitamin D levels in enrolled students in compared to sufficient vitamin D level. It recorded 95 (60.50%), 21 (33.33%); 73 (46.49 %), 16 (25.39%) and 54 (34.39%), 9 (14.28%), respectively. Statistically, there is a higher significant different between vitamin D deficiency and the frequency of Group A, B, C, D, G and F Streptococcal isolates, p-value < 0.05.

Table (3): The association between Vitamin D level and the frequency Streptococcal isolates

Streptococcal isolates	Vitamin D level (No.) %		Frequency Streptococcal isolates (No.) %	P- value
Group A Streptococci	Sufficient	63 (18.00)	31 (49.20)	0.001
	Insufficient	130 (37.14)	91 (70.00)	
	Deficient	157 (44.86)	115 (73.24)	
Group B Streptococci	Sufficient	63 (18.00)	21 (33.33)	0.016
	Insufficient	130 (37.14)	72 (55.38)	
	Deficient	157 (44.86)	95 (60.50)	
Group C Streptococci	Sufficient	63 (18.00)	16 (25.39)	0.020
	Insufficient	130 (37.14)	55 (42.30)	
	Deficient	157 (44.86)	73 (46.49)	
Group D Streptococci	Sufficient	63 (18.00)	9 (14.28)	0.014
	Insufficient	130 (37.14)	42 (32.30)	
	Deficient	157 (44.86)	54 (34.39)	
Group G Streptococci	Sufficient	63 (18.00)	4 (6.34)	0.008
	Insufficient	130 (37.14)	25 (19.23)	
	Deficient	157 (44.86)	33 (21.01)	
Group F Streptococci	Sufficient	63 (18.00)	2 (3.17)	0.014
	Insufficient	130 (37.14)	11 (8.46)	
	Deficient	157 (44.86)	15 (9.55)	

Correlation between vitamin D levels and the frequency of Group A Streptococcal (GAS) isolates

Table 4 & Fig 2 showed a higher significant correlation between vitamin D levels and the frequency of Group A Streptococcal (GAS) isolates ($r = 1.000^{**}$, $p = 0.001$). Pearson's correlation

coefficient between vitamin D levels and the frequency of Group A Streptococcal (GAS) isolates ($r = 1.000^{**}$), the p-value of the correlation (0.001), this correlation is highly significant because the p-value is less than 0.05.

Table (4): Correlation between vitamin D level and the frequency of Group A Streptococcal isolates (GAS).

	Vitamin D level	Frequency of GAS
Vitamin D level		
Pearson's Correlation	1	1.000(**)
Sig. (2 tailed)		0.001
N	3	3
Frequency Streptococcal pharyngitis		
Pearson's Correlation	1.000(**)	1
Sig. (2 tailed)	0.001	
N	3	3

**** Correlation is significant at the 0.01 level (2-tailed).**

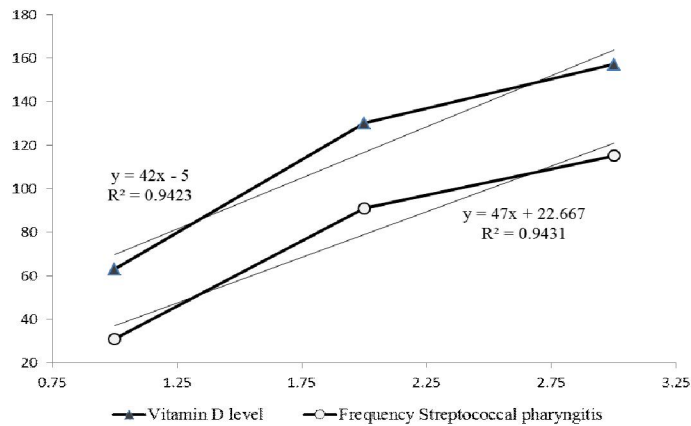


Fig (2): Correlation between vitamin D levels and the frequency of Group A Streptococcal (GAS) isolates ($r = 1.000^{}$, $p = 0.001$).**

Correlation between vitamin D levels and the frequency of Group B Streptococcal (GBS) isolates

Table 5 & Fig 3 indicated a higher significant correlation between vitamin D levels and the frequency of Group B Streptococcal (GBS) isolates ($r = 1.000^*$, $p = 0.016$). Pearson's correlation coefficient

between vitamin D levels and the frequency of Group B Streptococcal (GBS) isolates ($r = 1.000^*$), the p-value of the correlation (0.016), this correlation is highly significant because the p-value is less than 0.05.

Table (5): Correlation between vitamin D level and the frequency of Group B Streptococcal (GBS) isolates.

	Vitamin D level	Frequency of GBS
Vitamin D level		
Pearson's Correlation	1	1.000(*)
Sig. (2 tailed)		0.016
N	3	3
Frequency Streptococcal pharyngitis		
Pearson's Correlation	1.000(*)	1
Sig. (2 tailed)	0.016	
N	3	3

* Correlation is significant at the 0.05 level (2-tailed).

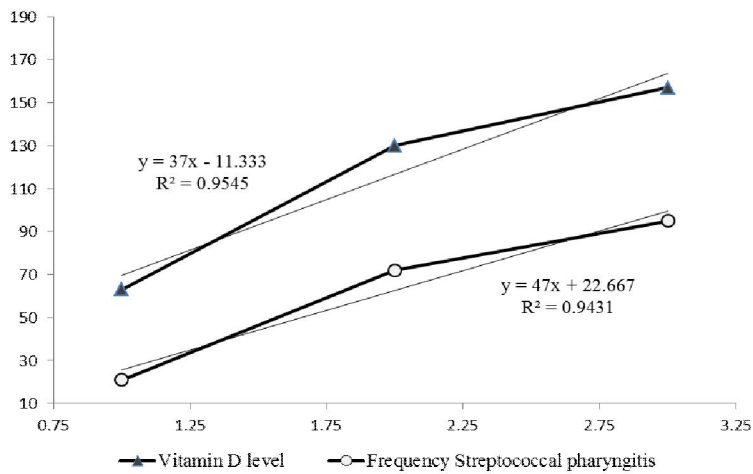


Fig (3): Correlation between vitamin D levels and the frequency of Group B Streptococcal (GBS) isolates ($r = 1.000^*$, $p = 0.016$).

Correlation between vitamin D levels and the frequency of Group C Streptococcal (GCS) isolates

Table 6 & Fig 4 recorded higher significant correlation between vitamin D levels and the frequency of Group C Streptococcal (GCS) isolates

($r = 1.000^*$, $p = 0.020$). Pearson's correlation coefficient between vitamin D levels and the frequency of Group C Streptococcal (GCS) isolates ($r = 1.000^*$), the p-value of the correlation (0.020), this correlation is highly significant because the p-value < 0.05.

Table (6): Correlation between vitamin D level and the frequency of Group C Streptococcal (GCS) isolates.

	Vitamin D level	Frequency of GCS
Vitamin D level		
Pearson's Correlation	1	1.000(*)
Sig. (2 tailed)		0.020
N	3	3
Frequency Streptococcal pharyngitis		
Pearson's Correlation	1.000(*)	1
Sig. (2 tailed)	0.020	
N	3	3

* Correlation is significant at the 0.05 level (2-tailed).

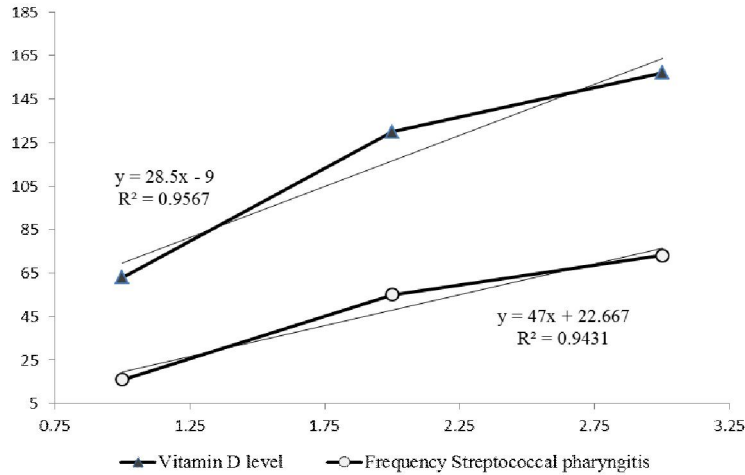


Fig (4): Correlation between vitamin D levels and the frequency of Group C Streptococcal (GCS) isolates (r = 1.000*, p = 0.020).

Correlation between vitamin D levels and the frequency of Group D Streptococcal (GDS) isolates

Table 7 & Fig 5 showed higher significant correlation between vitamin D levels and the frequency of Group D Streptococcal (GDS) isolates (r = 1.000*, p = 0.014). Pearson’s correlation coefficient

between vitamin D levels and the frequency of Group D Streptococcal (GDS) isolates (r = 1.000*), the p-value of the correlation (0.014), this correlation is highly significant because the p-value is less than 0.05.

Table (7): Correlation between vitamin D level and the frequency of Group D Streptococcal (GDS) isolates.

	Vitamin D level	Frequency of GDS
Vitamin D level		
Pearson’s Correlation	1	1.000(*)
Sig. (2 tailed)		0.014
N	3	3
Frequency Streptococcal pharyngitis		
Pearson’s Correlation	1.000(*)	1
Sig. (2 tailed)	0.014	
N	3	3

* Correlation is significant at the 0.05 level (2-tailed).

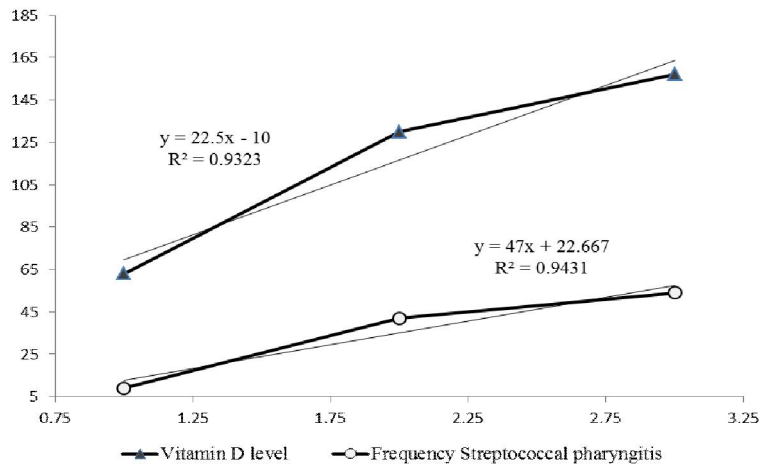


Fig (5): Correlation between vitamin D levels and the frequency of Group D Streptococcal (GDS) isolates (r = 1.000*, p = 0.014).

Correlation between vitamin D levels and the frequency of Group E Streptococcal (GES) isolates

Table 8 & Fig 6 recorded a higher significant correlation between vitamin D levels and the frequency of Group E Streptococcal (GES) isolates ($r = 1.000^{**}$, $p = 0.008$). Pearson's correlation

coefficient between vitamin D levels and the frequency of Group E Streptococcal (GES) isolates ($r = 1.000^{**}$), the p-value of the correlation (0.008), this correlation is highly significant because the p-value < 0.05.

Table (8): Correlation between vitamin D level and the frequency of Group E Streptococcal (GES) isolates.

	Vitamin D level	Frequency of GES
Vitamin D level		
Pearson's Correlation	1	1.000(**)
Sig. (2 tailed)		0.008
N	3	3
Frequency Streptococcal pharyngitis		
Pearson's Correlation	1.000(**)	1
Sig. (2 tailed)	0.008	
N	3	3

**** Correlation is significant at the 0.01 level (2-tailed).**

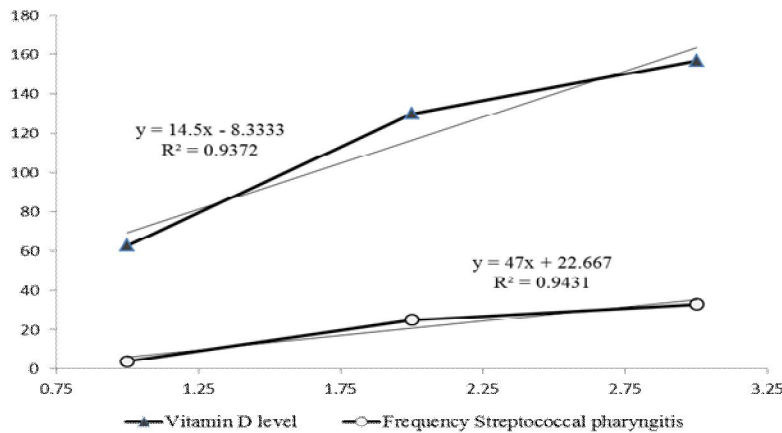


Fig (6): Correlation between vitamin D levels and the frequency of Group E Streptococcal (GES) isolates ($r = 1.000^{}$, $p = 0.008$).**

Correlation between vitamin D levels and the frequency of Group F Streptococcal (GFS) isolates

Table 9 & Fig 7 showed higher significant correlation between vitamin D levels and the frequency of Group F Streptococcal (GFS) isolates ($r = 1.000^*$, $p = 0.014$). Pearson's correlation coefficient

between vitamin D levels and the frequency of Group F Streptococcal (GFS) isolates ($r = 1.000^*$), the p-value of the correlation (0.014), this correlation is highly significant because the p-value is less than 0.05.

Table (9): Correlation between vitamin D level and the frequency of Group F Streptococcal (GFS) isolates.

	Vitamin D level	Frequency of GFS
Vitamin D level		
Pearson's Correlation	1	1.000(*)
Sig. (2 tailed)		0.014
N	3	3
Frequency Streptococcal pharyngitis		
Pearson's Correlation	1.000(*)	1
Sig. (2 tailed)	0.014	
N	3	3

*** Correlation is significant at the 0.05 level (2-tailed).**

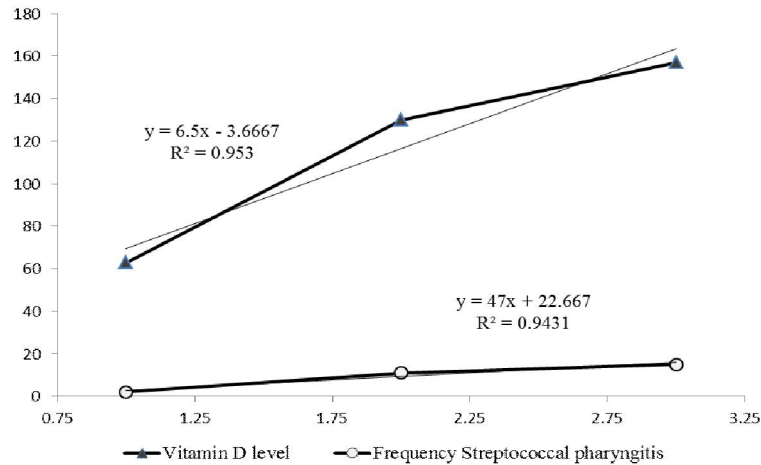


Fig (7): Correlation between vitamin D levels and the frequency of Group F Streptococcal (GFS) isolates ($r = 1.000^*$, $p = 0.014$).

4. Discussion

Vitamin D is crucial for bone health; it plays a role in calcium absorption, increased bone mineral density, and in preventing rickets, osteomalacia and fractures. Vitamin D deficiency has received significant media attention in recent years for its association with bone disorders, and for its possible association with other adverse health outcomes, including cancer, autoimmune diseases, infections, diabetes mellitus and cardiovascular conditions. Our study showed that, vitamin D deficiency within females was significantly higher than the prevalence in males. It recorded 122 (64.89%) and 66 (35.10%), respectively, p -value < 0.05 . This finding was in agree to the previous studies, where a lower level of 25-hydroxyvitamin D was commonly seen in those of younger age and again in females (Lee *et al.*, 2009; Higgins *et al.*, 2012; Braun *et al.*, 2012; Moromizato *et al.*, 2014; Anwar *et al.*, 2017; Elbistanl *et al.*, 2017; Bhurayanontachai *et al.*, 2018).

Nevertheless, lower sun exposure may be a leading cause of vitamin D deficiency within the female population (Islam *et al.*, 2008; Rajebi *et al.*, 2016). Similarly, our study recorded that, low daily sun exposure for at least 20 minutes, not physically active, drinking carbonated soft drink and not taking multivitamin supplements students had higher risk of vitamin D deficiency. Vitamin D deficiency recorded 195 (60.37%); 116 (62.03%); 151 (85.31%) and 107 (59.77%), respectively. Also, our study indicated that, school students, those with light brown skin color and normal weight appeared to be at higher risk of vitamin D deficiency. Vitamin D deficiency recorded 95 (53.37%) and 85 (46.44%), respectively. Contrary to our finding Reid *et al.*, (2011), found that low vitamin D levels were associated with dark skin, high body mass index, and large tonsil sizes.

Kaddam *et al.*, (2017) found that, Vitamin D deficiency was significantly higher with the increase in BMI, where the obese had prevalence of 60.2% as compared to 54.2% in the overweight and 47.4% in the normal weight students. However, there was no significant difference in the prevalence of vitamin D deficiency in school students by skin color and taking omega 3 supplements. On the other hand our work indicated that, vitamin D deficiency was significantly higher with the decrease in BMI, where the normal weight students had prevalence of 85 (46.44%) as compared to 68 (37.15%) in the overweight and 30 (16.39%) in obese students, those with light brown skin color, and those not taking omega 3 supplements and multivitamins appeared to be at higher risk of vitamin D deficiency, they recorded 95 (53.37%); 152 (85.39%) and 107 (59.77%), respectively.

Group A Streptococcus (GAS) is the most common bacterial cause of acute pharyngitis, responsible for 20-30% of sore throat in children (Shulman *et al.*, 2012). Many recent studies have confirmed the positive correlation between low levels of vitamin D and increased incidence of upper respiratory tract infections (URTIs) (Cannell *et al.*, 2006; Avenell *et al.*, 2007; Holick, 2007; Laaksi *et al.*, 2007; Nnoaham and Clarke, 2008; Ginde *et al.*, 2009; Adams and Hewison, 2010; Sabetta *et al.*, 2010; Aydin *et al.*, 2011; Berry *et al.*, 2011; Nseir *et al.*, 2012; Yildiz *et al.*, 2012; Science *et al.*, 2013; Collak *et al.*, 2014; Alladi and Gopal, 2017; Jian *et al.*, 2018).

Similarly our finding showed that, the frequency of Group A Streptococcal isolate recorded the most higher percentage between enrolled students which have deficient vitamin D level in compared to sufficient vitamin D level. It recorded 115 (73.24%) and 31 (49.20%), respectively. Followed by Group B, C and D Streptococci which recorded also a higher

percentage between deficient vitamin D levels in enrolled students in compared to sufficient vitamin D level. It recorded 95 (60.50%), 21 (33.33%); 73 (46.49%), 16 (25.39%) and 54 (34.39%), 9 (14.28%), respectively. Statistically, there is a higher significant different between vitamin D deficiency and the frequency of Group A, B, C, D, G and F Streptococcal isolates, p-value < 0.05.

Also, our data recorded a higher significant correlation between vitamin D deficiency and frequency of Group A, B, C, D, E and F Streptococcal isolates. Pearson's correlation coefficient between vitamin D levels and the frequency of Streptococcal isolates, $r = 1.000$, this correlation is highly significant because the p-value < 0.05.

5. Conclusion

Group A Streptococcus (GAS) is the most common bacterial cause of acute pharyngitis. Also, there is a significant correlation between vitamin D deficiency and the frequency of Streptococcal Pharyngitis.

Corresponding Author:

Dr. Mohamed Abdelhameid Fareid Alekhtaby
Botany and Microbiology Department
Faculty of Science, Al-Azhar University, Nasr City,
Cairo 11884, Egypt.
Present address: Basic Science Dept., Health Colleges,
H'ail University, Saudi Kingdom.
E-mail: mohamedfareid73@yahoo.com

References

- Adams JS, Hewison M (2010): Update in vitamin D. *J Clin Endocrinol Metab.*, 95: 471-478.
- Al-Ghamdi AH, Fureeh AA, Alghamdi JA, Alkuraimi WA, Alomar FF, Alzahrani FA, Alzahrani RA, Alzahrani AA, Alzahrani SA, Alghamdi AM (2017): High prevalence of vitamin D deficiency among Saudi children and adolescents with type 1 diabetes in Albaha Region, Saudi Arabia. *IOSR J. Pharm. Biol. Sci.*, 12, 5-10.
- Alladi YR, Gopal J (2017): Vitamin-D Deficiency and Relation to Recurrent Respiratory Tract Infection in children less than 5 years. *Journal of Dental and Medical Sciences (IOSR-JDMS)*. 16 (8): 88-96.
- Anwar E, Hamdy G, Taher E, Fawzy E, Abdulattif S, Attia MH (2017): Burden and outcome of vitamin D deficiency among critically ill patients: a prospective study. *Nutr Clin Pract.*, 32(3):378- 384.
- Avenell A, Cook JA, MacLennan GS, Macpherson GC (2007): Vitamin D supplementation to prevent infections: a sub-study of a randomised placebo-controlled trial in older people (RECORD trial, ISRCTN 51647438). *Age Ageing*; 36:574-577.
- Aydin S, Aslan I, Yildiz I (2011): Vitamin D levels in children with recurrent tonsillitis. *Int J Pediatr Otorhinolaryngol.*, 75(3): 364-367.
- Aydin S, Aslan I, Yildiz I, Agachan B, Toptas B, Toprak S (2011): Vitamin D levels in children with recurrent tonsillitis. *Int J Pediatr Otorhinolaryngol.*, 75:364-367.
- Ball SL, Siou GP, Wilson JA, Howard A, Hirst BH, Hall J (2007): Expression and immunolocalisation of antimicrobial peptides within human palatine tonsils. *J Laryngol Otol.*, 121(10): 973-978.
- Berry DJ, Hesketh K, Power C, Hypponen E (2011): Vitamin D status has a linear association with seasonal infections and lung function in British adults. *Br J Nutr.*, 106(9): 1433-1440.
- Bhurayanontachai R, Maipang K, Leelawattana R (2018): Correlation of admission serum 25-hydroxyvitamin D levels and clinical outcomes in critically ill medical patients. *Clinical Nutrition Experimental*; 20: 30-40.
- Bisno AL, Stevens DL (2000): *Streptococcus pyogenes*. P. 2101- 2117. In Mandell, G. L., Bennett, J. E. and Dolin, R. (ed.) *Mandell, Douglas and Bennett's Principals and Practice of Infectious Diseases*. 5th ed. Churchill Livingstone, New York.
- Brahmadathan K, Gladstone P (2006): Microbiological diagnosis of Streptococcal pharyngitis. *Indian J. Med. Microbiol.*, 24 (2): 92-96.
- Braun AB, Gibbons FK, Litonjua AA, Giovannucci E, Christopher KB (2012): Low serum 25-hydroxyvitamin D at critical care initiation is associated with increased mortality. *Crit Care Med.*, 40(1):63-72.
- Brogden KA (2005): Antimicrobial peptides: Pore inhibitors or metabolic inhibitors in bacteria? *Nat Rev Microbiol.*, 3: 238-250.
- Cannell JJ, Vieth R, Umhau JC, Holick MF, Grant WB, Madronich S (2006): Epidemic influenza and vitamin D. *Epidemiol Infect.*, 134:1129-1140.
- Carroll K, Reimer L (1996): Microbiology and Laboratory Diagnosis of Upper Respiratory tract Infection. *Clin. Infect. Dis.*, 23: 442-448.
- Cheesbrough M (2002): *District Laboratory Practice in Tropical Countries*. Part 2, Cambridge University Press UK. Pp.: 253-266.
- Collak A, Bozaykut A, Demirel B, Sezer R G, Seren LP, Dogru M (2014): Serum vitamin D levels in children with recurrent tonsillopharyngitis. *North Clin Istanbul – NCI*; 1(1):13-18.
- DuBose KC (2002): Group A Streptococcal pharyngitis. *Prim. Care Update Ob Gyns.*, 9: 222-225.
- Dyson A, Pizzutto SJ, MacLennan C, Stone M, Chang AB (2014): The prevalence of vitamin D deficiency in children in the Northern Territory. *J. Paediatr. Child.* 50, 47-50.
- Elbistanl MS, Güneş S, Yegin Y, Çelik M, Koçak HE, Evren C, Kayhan FT (2017): Relationship Between Serum Vitamin D Levels and Childhood Recurrent Tonsillitis. *Otolaryngol Open J.*, 3(1): 16-21.
- Esteitie R, Naclerio RM, Baroody FM (2010): Vitamin D levels in children undergoing adenotonsillectomies. *Int J Pediatr Otorhinolaryngol.*, 74(9): 1075-1077.

23. Facklam RR (1980): Manual of Clinical Microbiology^o, 3rd ed., Amer. Soc. For Microbiology, Washington, D. C., pp. 88-110.
24. Ginde AA, Mansbach JM, Camargo CA (2009): Association between serum 25 hydroxyvitamin D level and upper respiratory tract infection in the third National Health and Nutrition Examination survey. *Arch Int Med.*, 169(4):384-390.
25. Gordon CM, Feldman HA, Sinclair L, Williams AL, Kleinman PK, Perez-Rossello J, Cox JE (2008): Prevalence of vitamin D deficiency among healthy infants and toddlers. *Arch. Paediatr. Adolesc. Med.*, 162, 505-512.
26. Higgins DM, Wischmeyer PE, Queensland KM, Sillau SH, Sufit AJ, Heyland DK (2012): Relationship of vitamin D deficiency to clinical outcomes in critically ill patients. *J Parenter Enter Nutr.*, 36(6):713-720.
27. Holick MF (2004): Sunlight and vitamin D for bone health and prevention of autoimmune diseases, cancers, and cardiovascular disease. *Am J Clin Nutr.*, 80 Suppl 6:1678-1688.
28. Holick MF (2007): Vitamin D deficiency. *N Engl J Med.*, 357:266-281.
29. Islam MZ, Shamim AA, Kemi V, Nevanlinna A, Akhtaruzzaman M, Laaksonen M (2008): Vitamin D deficiency and low bone status in adult female garment factory workers in Bangladesh. *Br J Nutr.*, 99(6):1322-1329.
30. Jian Z, Juan D, Leting H, Youcheng W, Yimei S, Hailong L (2018): Preventive Effects of Vitamin D on Seasonal Influenza A in Infants: A Multicenter, Randomized, Open, Controlled Clinical Trial. *The Pediatric Infectious Disease Journal.* 37 (8): 749-754.
31. Kaddam IM, Al-Shaikh AM, Abaalkhail BA, Khalid S, Asseri KS, Al-Saleh, YM, Al-Qarni AA, Al-Shuaibi AM, Tamimi, WG, Mukhtar, AM. (2017): Prevalence of vitamin D deficiency and its associated factors in three regions of Saudi Arabia. *Saudi Med J.*, 38 (4): 381-390.
32. Laaksi I, Ruohola JP, Touhimaa P, Auvinen A, Haataja R, Pihlajamaki H (2007): An association of serum vitamin D concentration < 40 nmol/L with acute respiratory tract infection in young Finnish men. *Am J Clin Nutr.*, 86:714-717.
33. Lee P, Eisman JA, Center JR (2009): Vitamin D deficiency in critically ill patients. *N Engl J Med.*, 360 (18):1912- 1914.
34. Mansour MHK, Alhadidi KM (2012): Vitamin D deficiency in children living in Jeddah, Saudi Arabia. *Indian J Endocrinol. Metab.*, 16, 263-269.
35. McDonald M, Tower R, Fagan P (2006): Recovering streptococci from the throat, a practical alternative to direct plating in remote tropical communities. *J. Clin. Microbiol.*, 44 (2): 547-552.
36. Moromizato T, Litonjua AA, Braun AB, Gibbons FK, Giovannucci E, Christopher KB (2014): Association of low serum 25- hydroxyvitamin D levels and sepsis in the critically ill. *Crit Care Med.*, 42(1):97-107.
37. Nnoaham KE, Clarke A (2008): Low serum vitamin D levels and tuberculosis: a systemic review and meta-analysis. *Int J. Epidemiol.*, 37:113-119.
38. Norman AW, Bouillon R, Whiting SJ, Vieth R, Lips P (2007): 13th Workshop consensus for vitamin D nutritional guidelines. *J Steroid Biochem Mol Biol.*, 103:204-205.
39. Nseir W, Mograbi J, Abu-Rahmeh Z, Mahamid M, Abu-Elheja O, Shalata A (2012): The association between vitamin D levels and recurrent group A streptococcal tonsillopharyngitis in adults. *Int J Infect Dis.*, 16(10): 735-738.
40. Rajebi H, Khodadad A, Fahimi G, Abolhassani H (2016): Vitamin D deficiency among female nurses of Children's medical center hospital and its related factors. *Acta Med Iran;* 54(2):146- 150.
41. Reid D, Morton R, Salkeld L, Bartley J (2011): Vitamin D and tonsil disease--preliminary observations. *Int J Pediatr Otorhinolaryngol.*, 75(2): 261-264.
42. Rosen CJ, Abrams SA, Aloia JF, Brannon PM, Clinton SK, Durazo-Arvizu RA (2012): IOM committee members respond to Endocrine Society vitamin D guideline. *J Clin Endocrinol Metab.*, 97: 1146-1152.
43. Ross AC, Taylor CL, Yaktine AL, Del Valle HB (2011): Dietary reference intakes for calcium and vitamin D. Washington (DC): National Academies Press (US).
44. Sabetta JR, DePetrillo P, Cipriani RJ, Smardin J, Burns LA, Landry ML (2010): Serum 25-hydroxyvitamin D and the incidence of acute viral respiratory tract infections in healthy adults. *PLoS One.*, 5(6): e11088.
45. Science M, Maguire JL, Russell ML, Smieja M, Walter SD, Loeb M (2013): Low serum 25-hydroxyvitamin D level and risk of upper respiratory tract infection in children and adolescents. *Clin Infect Dis.*, 57: 392-397.
46. Shulman ST, Bisno AL, Clegg HW (2012): Clinical practice guideline for the diagnosis and management of group A streptococcal pharyngitis: 2012 update by the infectious diseases society of America. *Clin Infect Dis.*, 55(10): 86-102.
47. Taheri Z, Ghafari M, Hajivandi A, Amiri M (2014): Vitamin D deficiency in children and adolescents; an international challenge. *J. Parathy. Dis.*, 2, 27-31.
48. Yildiz I, Unuvar E, Zeybek U, Toptas B, Cacina C, Toprak S (2012): The role of vitamin D in children with recurrent tonsillopharyngitis. *Ital J Pediatr.*, 38(25): 1-6.