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## Assessment of Vitamin D Levels Among a Sample of Medical Laboratory Visitors in Misurata City and their Association with Seasonal Variations

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### ABSTRACT

Vitamin D deficiency is one of the most prevalent health issues in contemporary societies, affecting individuals of both sexes across various age groups. This study aimed to assess the concentration levels of vitamin D among visitors to medical laboratories in Misurata City from September 2019 to August 2023. Data collection involved two approaches: blood samples were obtained from visitors between January and August 2023, while vitamin D level records from September 2019 to December 2022 were retrieved from laboratory archives.

The study's findings revealed a widespread deficiency in vitamin D levels among the participants throughout the study period. Notably, the deficiency was significantly higher in females (80.42%) than males (19.57%). In terms of age groups, individuals aged 35–45 years were identified as the most susceptible, with a deficiency rate of 34.10%, showing statistically significant differences.

Regarding the relationship between vitamin D levels and seasonal variations, the study found that winter and spring had the highest prevalence of vitamin D deficiency, with statistically significant differences compared to other seasons. Conversely, vitamin D levels were observed to increase during autumn and summer.

The findings further demonstrated that vitamin D levels among medical laboratory visitors were influenced by factors such as gender, age group, and seasonal variation. These results underscore the importance of monitoring vitamin D concentrations to mitigate the risk of developing conditions associated with its deficiency, such as hair loss and chronic diseases like kidney failure.

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### Introduction

Vitamins are essential nutrients required by the human body in small quantities to support numerous vital physiological functions. Since most vitamins cannot be synthesized by the body, they must be obtained from dietary sources or nutritional supplements. Vitamins are classified into two main categories: water-soluble vitamins, such as vitamins B and C, and fat-soluble vitamins, including vitamins A, D, E, and K [1].

This study focuses on vitamin D, which is considered one of the most critical vitamins due to its significant role in regulating various functional processes within the body. Vitamin D is unique in that it can be synthesized endogenously in the skin through exposure to ultraviolet (UV) rays or absorbed through the

intestines from dietary sources. Its primary function is to facilitate the absorption of calcium and phosphorus from the intestines, ensuring their balance and maintaining healthy bones.

Vitamin D deficiency is a widespread issue, particularly among infants and children. A deficiency in this vitamin is the leading cause of rickets, a condition characterized by weakened and deformed bones as well as impaired growth in children. Additionally, insufficient levels of vitamin D are associated with an increased risk of fractures and the development of osteoporosis. Among pregnant women, vitamin D deficiency has been linked to symptoms such as hair loss and depression [2].

The COVID-19 pandemic has further highlighted the importance of vitamin D, as it has brought about significant lifestyle changes, including reduced outdoor activity and limited exposure to sunlight. These changes may have had an impact on vitamin D

status at a population level [3]. Addressing vitamin D deficiency is therefore critical to improving overall health and reducing the risk of associated health complications.

Numerous studies have investigated vitamin D levels in Libya, including the research conducted by Damona et al., which aimed to assess vitamin D levels and their relationship with various biochemical indicators in hemodialysis patients at the Hemodialysis Treatment and Dialysis Center in Misurata [4]. The findings indicated that individuals with chronic kidney failure exhibited low vitamin D levels [4]. Similarly, the study by Al-Habib and Al-Hadhiri focused on the prevalence of vitamin D deficiency in Sabha and its associations with factors such as gender, age, geographical location, lifestyle, and medical conditions [5]. Their results revealed a 26.4% prevalence of vitamin D deficiency among both sexes during the study period from May to November 2019.

Additionally, Abu Mahdi conducted a study in Sabratha, Libya, to evaluate vitamin D levels among residents in that region [6]. This study, which included 240 women over nine months from June 1, 2019, to February 28, 2020, found a high prevalence of vitamin D deficiency, particularly among women aged 30 to 44 [6]. Furthermore, Bayazid examined vitamin D levels and associated diseases in Latakia, Syria, during 2012-2013, involving 127 randomly selected cases [7].

Lastly, Mahmoud et al. conducted a study at Samarra University in Iraq to evaluate vitamin D levels concerning liver and kidney function [8]. This research involved 90 blood serum samples (52 males and 38 females) aged 20 to 30 years, revealing a significant reduction in vitamin D levels. These studies underscore the importance of identifying vitamin D levels and the various health issues linked to its deficiency.

The objective of this study was to assess vitamin D levels among individuals visiting medical laboratories from 2019 to 2023. Additionally, the study aimed to identify the age group most at risk for vitamin D deficiency in both genders and to explore the relationship between vitamin D levels and seasonal variations.

## Materials and Methods

### Study Area

Misurata is situated in northwestern Libya, along the Mediterranean coast, approximately 200 km east of Tripoli and 257 km west of Sirte. The region is characterized by a Mediterranean climate, featuring hot, dry summers and warm, rainy winters, with an average annual temperature of 20°C.

### Sample Collection

This study involved the collection of random blood samples from individuals visiting various medical laboratories, including Tabarak Laboratory, Al-Suwaihli Laboratory, Al-Safwa Laboratory, Al-Majd Laboratory, Lamis Laboratory, and Aqzir Laboratory, during the period from January to August 2023. Additionally, vitamin D level results were gathered from the same laboratories' records covering the period from September 2019 to December 2022.

### Sample Examination

Blood samples were drawn by trained personnel and processed by centrifugation at 4000 rpm for 10 minutes to obtain serum. A vitamin D chip was then placed into a reader to determine vitamin D levels. A volume of 50 µl of serum was extracted using a pipette and transferred to the mixing tube of the test kit. Following this, 50 µl of releasing buffer was added, and the mixture was gently

shaken for 10 seconds. The tube was incubated in an iron holder at 35°C for 5 minutes. After incubation, 100 µl of detection buffer was added, and the mixture was shaken gently for another 10 seconds. It was then incubated at 35°C for 15 minutes, with shaking every 1.5 minutes while remaining in the iron holder. After 15 minutes, 75 µl of the mixture was placed on the test card and incubated for 8 seconds before being inserted into the reader for analysis.

### Statistical Analysis

Statistical analyses were performed to identify significant differences in vitamin D levels and their relationship to gender, age, and seasonal variations using the t-test. Additionally, a multi-directional variance test was employed to explore relationships between vitamin D levels, age, and gender during the years 2020-2022, utilizing the SPSS software, version 2022.

### Results

Assessment of Vitamin D levels and their association with Sex Table 1 presents the average vitamin D levels by gender. The percentage of undecided individuals was greater among females (80.42%) than males (19.57%). Additionally, the average vitamin D levels were higher in males, ranging from 1.01 to 101, while females had levels ranging from 0.72 to 109. The differences between genders were highly significant ( $P \leq 0.01$ ).

**Table 1: Average Vitamin D Levels and their Relationship to Gender.**

Gender	Mean $\pm$ S. E	Percentage
Males	**.947 $\pm$ 29.82	19.57%
Females	**.494.0 $\pm$ 26.12	%80.42

\*\* High significant differences ( $P \leq 0.01$ ).

### Evaluation of Vitamin D Levels and Their Relationship to Different Age Groups

Table 2 illustrates that the age group between 30 and 45 years had the highest number of individuals undergoing vitamin D testing, followed closely by those aged 15 to 30. In contrast, individuals over 60 years old were the least likely to take a vitamin D test. Interestingly, the average vitamin D level was highest in the age group under 15 years, with a mean level of 31.24 ng/mL, ranging from 29.2 to 33.2 ng/mL. This was followed by the over-60 age group, which had an average level of 30.78 ng/mL, within a range of 27.3 to 34.2 ng/mL. The average vitamin D levels for individuals aged 15 to 60 years were relatively close; however, statistical analysis revealed highly significant differences ( $P \leq 0.01$ ) in vitamin D levels across the different age groups.

**Table 2: Average Vitamin D Levels and Their Relationship to Age Groups.**

Age group	Mean $\pm$ S. E	Percentage
Less than 15	**1.03 $\pm$ 31.24	%15.24
30-15	**0.758 $\pm$ 24.73	%31.66
45-30	**0.856 $\pm$ 26.69	%34.10
60-45	**1.14 $\pm$ 24.94	%12.80
Over 60	**1.77 $\pm$ 30.78	%6.18

\*\* High significant differences ( $P \leq 0.01$ ).

### Evaluation of Vitamin D Levels Based on Seasonal Variations

When evaluating vitamin D levels based on seasonal variations, it was noted that summer exhibited the highest average vitamin D level at 34, followed by autumn at 32.3. Spring demonstrated a decrease to 29.5, while winter recorded the lowest level at 25, as detailed in Table 3. The results of the statistical analysis using the variance test also indicated significant differences ( $P \leq 0.01$ ).

**Table 3: Average Vitamin D Levels and their Relationship to the Seasons of the Year.**

seasons	Mean $\pm$ S. E	P-value
Autumn	**3.52 $\pm$ 32.3	000
Winter	**2.05 $\pm$ 25	000
Spring	**2.77 $\pm$ 29.5	000
Summer	**6.2 $\pm$ 34	

\*\* High significant differences ( $P \leq 0.01$ ).

In comparing vitamin D levels with seasonal variations for males, it was found that the average vitamin D level was similar during the fall and summer seasons, at 32.6. The average dropped to approximately 27.9 in spring, while winter recorded the lowest average at about 25.2, as detailed in Table 4. Statistical analysis using the t-test showed significant differences ( $P \leq 0.01$ ).

For females, the autumn and summer seasons displayed similar vitamin D levels, averaging 24.4 and 24.7, respectively. Spring recorded an average of 22.5, while winter saw a decrease to an average of 22.3, as shown in Table 4.

**Table 4: Average Vitamin D Levels for Both Sexes and their Relationship to the Seasons of the Year**

Seasons	Mean $\pm$ S. E		P-value
	Females	Males	
Autumn	**0.47 $\pm$ 24.4	**1.1 $\pm$ 32.6	000
Winter	**0.44 $\pm$ 22.3	**0.93 $\pm$ 25.1	000
Spring	**0.51 $\pm$ 22.5	**1.14 $\pm$ 27.9	000
Summer	**0.49 $\pm$ 24.7	**1.28 $\pm$ 32.6	

\*\* High significant differences ( $P \leq 0.01$ ).

Regarding the relationship between assessing vitamin D levels across different age groups according to seasonal variations, the findings are as follows: For those under 15, vitamin D levels were comparable during autumn and summer, with averages of 32.5 and 32.1, respectively. Spring and winter displayed lower averages of 29.6 and 28.3, as indicated in Table 5.

In the 15 to 30 years age group, autumn recorded the highest level at an average of 24.2, followed by summer and winter at 23.6 and 22.6, respectively. Spring saw a decline to 20.6. Among individuals, aged 30 to 45, vitamin D levels were similar during summer and autumn, averaging 24.9 and 24.7, respectively. Winter and spring recorded averages of 23.2 and 22.3.

In the 45 to 60 years age group, the average vitamin D level in autumn was about 28.2, followed by summer at 25.5, and spring and winter at 22.4 and 21.6, respectively.

Finally, for those over 60, the average vitamin D level was approximately 32.4, with spring next at 31.2, and autumn and winter averaging 27.5 and 26.5, respectively.

Overall, the results suggest that summer and autumn exhibit the highest levels of vitamin D across various age groups, with statistical analysis revealing significant differences ( $P \leq 0.01$ ), as detailed in Table 5.

**Table 5: Average Vitamin D Levels by Age Groups and their Relationship to the Seasons of the Year.**

seasons	Mean $\pm$ S. E					P-value
	Less than 15	30-15	45-30	60-45	Over 60	
Autumn	**1.3 $\pm$ 32.6	**0.77 $\pm$ 24.2	**0.69 $\pm$ 24.7	**1.35 $\pm$ 28.2	**1.85 $\pm$ 27.5	000
Winter	**1.3 $\pm$ 28.4	**1.46 $\pm$ 22.6	**1.78 $\pm$ 23.2	**0.80 $\pm$ 21.6	**1.46 $\pm$ 26.5	000
Spring	**1.4 $\pm$ 29.7	**0.68 $\pm$ 20.6	**0.76 $\pm$ 22.3	**1.01 $\pm$ 22.4	**2.53 $\pm$ 31.2	000
Summer	**1.4 $\pm$ 32.1	**0.81 $\pm$ 23.5	**0.78 $\pm$ 24.9	**1.04 $\pm$ 25.5	**2.17 $\pm$ 32.4	

\*\* High significant differences ( $P \leq 0.01$ ).

### Evaluation of vitamin D levels during the Period from 2020-2022

Through the results of the vitamin D obtained from previous records of previous laboratories, it was found that the average level of the vitamin was close during the years 2020 and 2021 (28.7, 30.4) respectively, followed by the year 2022 with an average estimated at (26.7), as in the table (6). The results of the statistical analysis using analysis of variance showed significant differences during the years 2021 and 2022 ( $P \leq 0.05$ ). While there were no significant differences during the year 2020 ( $P > 0.05$ ), as for the gender, it was found that the average level of the vitamin was high in males during the years 2021 and 2022 (32.5, 30.3) respectively. While during the year 2020, the average vitamin D level was higher in females than males (29.4), as in the table (7). Furthermore, it was also shown through statistical analysis using analysis of variance that there are no significant differences based on gender. Based on the analysis of vitamin D records from previous laboratories, it was found that the average levels of this vitamin were relatively close in the years 2020 and 2021, with averages of 28.7 and 30.4, respectively. However, in 2022, the average decreased to 26.7, as shown in Table 6. Statistical analysis using analysis of variance (ANOVA) indicated significant differences between the years 2021 and 2022 ( $P \leq 0.05$ ), while no significant differences were observed in 2020 ( $P > 0.05$ ).

Regarding gender, the average vitamin D levels were higher in males during 2021 and 2022, with values of 32.5 and 30.3, respectively. In contrast, in 2020, females had a higher average vitamin D level (29.4) than males. This data is summarized in Table 7. Furthermore, ANOVA results showed no significant differences in vitamin D levels based on gender from 2020 to 2022.

**Table 6: Average Vitamin D Levels during the Years 2020-2022.**

The years	Mean $\pm$ S. E	P-value
2020	1.10 $\pm$ 28.7	NS
2021	**1.03 $\pm$ 30.4	000
2022	**0.63 $\pm$ 26.7	000

\*\* High significant differences ( $P \leq 0.05$ ).

**Table 7: Average Vitamin D Levels for Both Genders during the Years 2020-2022.**

The years	Mean $\pm$ S. E		P-value
	Females	Males	
2020	1.005 $\pm$ 29.3	1.9 $\pm$ 27.9	NS
	2021	0.99 $\pm$ 28.4	0.93 $\pm$ 32.5
2022	0.55 $\pm$ 23.1	1.13 $\pm$ 30.3	NS

NS≡ Non-Significant Differences

Regarding the age groups from 2020 to 2022, it was found that individuals under 15 years of age had the highest participation levels compared to other age groups, with estimates of 32.02% in 2020, 35.3% in 2021, and 30.05% in 2022. The next highest participation level was observed in the 30-45 age group, with an estimate of 31.8% in 2020. Following this, the 45-60 age group had an estimated participation level of approximately 31.2%. In 2022, the remaining age groups showed similar participation levels, as indicated in Table 8. Additionally, statistical analysis revealed no significant differences between the age groups during the years 2020 to 2022.

**Table 8: Average Vitamin D Levels According to Age Groups during the Years 2020-2022. NS≡ non-significant differences**

The years	Mean $\pm$ S.E					P-value
	Less than 15	30-15	45-30	60-45	Over 60	
2020	2.02 $\pm$ 32.02	1.6 $\pm$ 26.2	1.7 $\pm$ 31.8	2.3 $\pm$ 23.6	3.23 $\pm$ 34.7	NS
2021	1.7 $\pm$ 35.3	1.5 $\pm$ 24.76	1.7 $\pm$ 27.68	2.6 $\pm$ 30.4	3.24 $\pm$ 31.2	NS
2022	1.4 $\pm$ 30.1	0.8 $\pm$ 22.38	0.7 $\pm$ 23.66	1.4 $\pm$ 25.1	2.16 $\pm$ 27.8	NS

## Discussion

This study aimed to assess vitamin D levels from January to August 2023, along with analyzing data from previous records covering September 2019 to December 2022. The research took into account factors such as gender, age, and seasonal variations, and involved data collection from 1,002 questionnaires randomly distributed among various groups in and around the city of Misurata.

The findings revealed a widespread deficiency in vitamin D levels, with females showing the lowest levels at approximately 80.42%. In contrast, males had a deficiency rate of about 19.57%. These results suggest a significant association between vitamin D deficiency and gender. This conclusion aligns with a study conducted in Tripoli by Alnagar and Abdalmula, which reported that 65% of females were vitamin D deficient compared to 35% of males [9]. Similarly, research from Sabha by Habib and Al-Hadhiri found that 89% of females were deficient, while only 11% of males were affected [5].

Moreover, a study in Benghazi by Sherif and Amnese (2018) indicated that 80% of females were vitamin D deficient, compared to 20% of males. This trend was also observed in research by Khalife et al. in Lebanon, which reported that 62% of females were deficient, versus 34% of males [10].

The results of this study indicate that the age group of 35-45 years is the most affected by vitamin D deficiency, with a prevalence rate of 34.1% across both sexes, and females being more affected than males. This increased vulnerability may be linked to factors such as a higher number of births, poor dietary habits, and a lack of vitamin D supplementation.

These findings are consistent with research conducted by Alnagar and Abdalmula in Tripoli, which showed that individuals aged 35 to less than 50 years are more likely to experience low vitamin D levels [9]. Similarly, a study by Habib and Al-Hudayri in Sabha found that the 25-45 age group had the highest rate of vitamin deficiency, at 61% Habib [5]. Additionally, research by Khalife et al. in Lebanon reported a deficiency rate in the 35-40-year age group, further supporting these findings [10].

Research indicates a connection between low vitamin D levels and seasonal variations. The findings show that vitamin D deficiency is most pronounced in winter, followed by spring. In contrast, autumn and summer generally exhibit higher average levels of vitamin D. This trend can be attributed to reduced sunlight exposure during the winter months, as people tend to stay indoors due to colder temperatures and heavier clothing.

Additionally, the study revealed that average vitamin D levels in females are lower than in males across all seasons. This discrepancy is likely due to women's tendency to wear more modest clothing that covers their bodies and their reduced outdoor activity compared to men, often related to work commitments.

These results are consistent with a study conducted in Ukraine by Grygorieva et al., which also highlighted the correlation between seasonal variations and vitamin D levels in the population, finding that winter and spring typically show lower vitamin D levels compared to autumn and summer [11].

## Conclusion

The results of this study indicate a decrease in vitamin D levels, particularly among females compared to males. The most

significant declines are observed in the 35-45 age group when compared to other age groups. Additionally, seasonal variations appear to influence vitamin D levels, with levels generally lower in autumn and summer than in winter and spring. This suggests that seasonal changes play a major role in the fluctuations of vitamin D levels.

Furthermore, several diseases have been linked to low vitamin D levels, including kidney disease, hair loss, diabetes, and heart and thyroid disorders. However, the study found no correlation between blood calcium concentration and low vitamin D levels.

### Availability of Data and Materials

No datasets were generated or analyzed during the current study.

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### Contributions

Ash and Mam: Data collection, initial analysis, and first draft of the manuscript. Lay, Kha, Khd, Fat and Sar: Supervision of the research project, completion of data analysis, proof reading and editing the manuscript. MAB, ROW and HAN: Proof reading and editing the manuscript.

### Ethics Approval and Consent to Participate

The Misurata University Board's approval was acquired from the Science College's Zoology Department.

### Consent for Publication

Yes.

### Competing Interests

The authors declare no competing interests.

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