

Measurement of Calcium and Vitamin D3 Levels in the Blood Serum of Smokers and Compared to Healthy Controls

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Annotation: This study aims to determine the evolution of calcium and vitamin D3 levels in the blood serum of smokers, and how smoking duration affects each level compared to healthy individuals. The study was conducted in the city of Baqubah (at the specialized Al-Shams laboratory), where 40 blood samples were taken – 20 from smokers and 20 from non-smokers in good health aged 20 to 70 years old. The results of the current study showed a significant decrease in the level of probability ($P < 0.001$) in the values of average calcium and vitamin D3 in the blood serum of smokers when compared to healthy non-smokers. The results also indicated that the duration of smoking does not have a significant impact on the levels of calcium and vitamin D3 in the blood serum, although there is an inverse relationship between the duration of smoking and the levels of calcium and vitamin D3, as their levels decreased with the increase in the duration of smoking, but they were not statistically significant. The results of this study show that smoking cigarettes is linked to bad health effects in the body. Also, smoking

cigarettes may mess up the gut system's calcium absorption systems, making it harder for the body to absorb calcium and vitamin D3.

Keywords: Vitamin D3 Levels; Smokers; Calcium

Introduction

Cigarette smoking is the biggest threat to public health worldwide, killing more than seven million people annually [1]. More than 7,000 harmful chemicals are found in cigarette smoke. When a smoker breathes in this smoke, they are exposed to these toxins, which have negative effects on their body right away [2,3]. In just a few fractions of a second, cigarette smoke can reach the heart, brain, and other parts of the body. It is breathed in straight into the alveoli and then spreads into the pulmonary vein [4]. Almost every function in the body is affected by smoking. Smokers get many diseases that lower their health and quality of life [5]. A change or difference in the levels of electrolytes (including calcium) and vitamins (vitamin D3) is one of a set of changes that occur to the body as a result of or in response to smoking. The change occurs at the systemic, cellular and molecular level [5]. Vitamin D3 and calcium have many biological effects on the body's organs and systems. Calcium is an essential element or mineral that plays a very important role in the physiological and biochemical processes of living things and cells. The calcium ion is an electrolyte that is important for keeping the digestive system, blood flow, and muscles working properly. It is also important for bone growth and formation and for making blood cells [6,7]. Vitamin D3 is an essential molecule for bone formation and growth. Vitamin D3 plays an important physiological role for cellular metabolism and immune and respiratory system function for both sexes and at all ages [8,9]. Studies have shown that smoking cigarettes makes it harder for the body to absorb vitamin D3, which is needed to absorb calcium from food. Not having enough vitamin D3 in the body makes it harder for the body to absorb calcium from food in the intestine, which leads to osteoporosis and weak bones that are more likely to break [10]. This is due to several factors, including the difference in lifestyle in smokers compared to others, smokers are less physically active and have malnutrition, as well as toxins resulting from smoking cigarettes, all of these factors have a toxic and direct effect on bone cells [11,12]. However, lifestyles that may affect the concentration of vitamin D3 in blood serum have not been clearly identified. In particular, smoking was associated with serum vitamin D3 levels, which was not clear. Most recent studies have indicated lower levels of vitamin D3 in blood serum in smokers compared to non-smokers [13].

Methodology:

The study relied on a non-random simple selection method for collecting samples, based on several variables including: name, age, gender, smoking duration. A total of 20 blood samples were collected from smokers and compared with 20 samples from healthy non-smokers, ranging in age from 20 to 70 years. For both the healthy group and the smokers, 10 milliliters of venous blood were drawn and placed in dry, clean gel tubes. The tubes were left at 37°C for 30 minutes, then the serum was separated using a centrifuge (3000 rpm) for 10 minutes. Afterward, the levels of calcium and vitamin D3 in the serum were examined.

Measurement Of Serum Calcium Concentration:

The level of calcium in the blood serum was measured by chromatography using the diagnostic kit prepared by Spinact. The colorimetric method doesn't need the protein to be precipitated. Instead, it

uses 8-hydroxyquinoline to dissolve the Cresolphthalein complex O. This mixes with calcium ions in a base medium to make a coloured complex whose intensity can be measured by a spectrometer. It is related to the amount of calcium in the sample that the colour turns is very strong [14].

Measurement Of Serum Vitamin D3 Concentration:

Vitamin D3 levels were measured in the blood serum using the MAGLUMI 1000 device, which was equipped by the Chinese company Shenzhen New Industries.

Principle of action

Using Vitamin 25(OH) D3 purified antigen for ABEI education, and using single-origin antibody to feed the sample FITC, titration, reagent, FITC and coated magnetic granules were mixed with FITC antibodies and then incubated at 37°C to form an antibody complex. After deposition in a magnetic field, the clear is eliminated, then washed off and once. Hence the addition of the ABEI teacher, after incubation and washing for the second time, the sample antigen and the ABEI antigen were competed for the purpose of combining with FITC - antibodies, and forming antibody-antigen compounds. After that, the detectors were added, and the light signals were measured through an amplifier as RLU within 3 seconds, as the intensity of the light signal is proportional to the concentration of vitamin 25(OH)D3 present in the samples [15].

The test was performed automatically through chemiluminescence immunoassay (CLIA) analyzer maglumi, and each parameter variable was identified by the RFID card on the detector.

Statistical analysis:

Statistical analysis is often used to analyze quantitative data, and provides methods for data description, simple inference for continuous and categorical data. The procedure involves the collection of data leading to test of the relationship between two statistical data sets. In this study all data are presented as frequency and percentage. Some of the tests we used were the dependent t-test (two-tailed) and the independent t-test (two-tailed) for factors that were normally distributed. For variables that did not have a normally distributed distribution, we used the Mann-Whitney U test, the Wilcoxon test, and the Chi-square test. $M < 0.05$ was seen as statistically significant.

Ethical approval:

The study was approved by the human ethics committee of the specialized Al-Shams laboratory, Everyone who took part in the study was told about it and asked to sign a consent form. The patient was also guaranteed that his information would be kept private.

Results and Discussion

Table 1 indicates a severe decrease in the values of average calcium and vitamin D3 levels in the blood serum of people who smoke compared to non-smokers (healthy). Their respective levels showed a significant decline at $p < 0.001$ probability.

Table 1: Values of Mean Calcium Ca and Vitamin D3 in the blood serum of smokers compared to healthy people.

Standard	Group of healthy individuals Mean \pm standard error	Group of smokers Mean \pm standard error	Mood level
Number	20	20	
Calcium	9.36 \pm 0.14	7.15 \pm 0.26	$P \leq 0.001$
Vitamin D3	46.18 \pm 2.09	15.79 \pm 1.38	$P \leq 0.001$

The low level of calcium is due to several reasons, including that cigarette smoke leads to a failure in the process of regulating electrolyte levels (including calcium) inside and outside the cell, as a result of the change in the cell's response due to smoking, which causes the loss of large amounts of calcium in the urine [16,17]. Or it may be because smoking has an effect on the functioning of the parathyroid gland that regulates the level of calcium in the blood. Smoking is linked to weak bones,

poor calcium intake, and low vitamin D3 levels [18,19]. The researchers also pointed out that smoking cigarettes has wide-ranging effects on the body as it works to reduce the absorption of calcium in the intestine, which indirectly affects the metabolism, growth and formation of bones. As well as low bone density (osteoporosis) [20].

There is less vitamin D3 in the blood of smokers than in the blood of non-smokers, which is good. Its amounts went down significantly at the $p < 0.001$ level of significance. For one thing, smoking generally goes hand-in-hand with worsening health and a sedentary lifestyle (less exercise, drinking, and bad eating habits), which means less time in the sun. This led to less vitamin D3 being made [21]. Recent studies have indicated that smoking plays a key role in vitamin D3 deficiency, as metabolic derivatives of nithalan (a substance in cigarette smoke) such as tetralones can inhibit the activity of the gene (CYP27A1) responsible for regulating the level of vitamin D3 in the body [22]. Furthermore directly influencing vitamin D3 metabolism and function could be the compounds in cigarette smoke. Consequently, smokers have a poor vitamin D3 intake; moreover, there is evidence that smoking may change the expression of some genes involved in the metabolic routes of vitamin D3 in the body. [23–26] Studies have shown that smoking reduces the synthesis of steroid hormones, therefore compromising vitamin D3 [27]. The results in Table (2) also indicated that the duration of smoking does not have a significant effect on the levels of calcium and vitamin D3 in the blood serum, although calcium levels decreased with the increase in the duration of smoking, but it was not statistically significant. This backs up what another study (Grace Umahi-Ottah et al., 2022) found: the harmful chemicals in cigarette smoke may mess up the processes that smokers' guts use to absorb calcium, which can lead to problems with absorption and low calcium levels. The amount of calcium in smokers' blood did not change based on how long, how often, or how strongly they smoked. The study's results showed that there were no statistically significant changes in calcium levels between smokers based on how long they had been smoking. Serum calcium levels aren't usually affected by how long someone smokes [28].

Table (2) Effect of smoking duration on calcium levels Ca and vitamin D3 in smokers.

Duration of smoking	Number	Ca Smoker	D3 Smoker
10-12 years	10	0.28± 7.81	1.38±13.91
13-16 years	5	0.68±6.68	3.57±20.09
17-23 years	5	0.30±6.32	2.92±15.26

That being said, the exact reason for this low calcium and vitamin D3 levels still needs to be figured out. Because smokers may not be getting enough calcium or vitamin D3, their bodies may not be absorbing calcium as well, they may be reabsorbing calcium from their bones more quickly, or they may be losing too much calcium ion in their urine. All of these things can cause an imbalance of calcium ions in the blood, which usually causes the level of parathyroid hormone (PTH) to rise [29]. Low extracellular calcium for a few weeks or months causes the parathyroid gland to grow too big, which is a sign of PTH [30].

Conclusion: It can be concluded that cigarette smoking is associated with harmful health consequences on the vital activities of the human body. It's important to know that smoking cigarettes may mess up the functions that absorb calcium in the digestive system. This can make it harder for the body to receive calcium and vitamin D3.

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