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Original Article

A Retrospective Study of Serum Calcium Status in Tehran, Iran (105,128 Samples, from 2009-2018)

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Abstract

Background: Calcium is a necessary mineral for life to keep the body and bones healthy. Various factors including hormones, diet, age, and gender affect serum calcium status. The aim of this sturdy was to assess the serum calcium level (SCL) of Tehran population, which has about 10 million multi-Ethnic populations and represents from the whole country.

Methods: In this retrospective study, the measured SCL of 105,128 individuals referred to different laboratories of Tehran, Iran were evaluated and its relationship with the age, gender, seasons, and different years during 2009-2018, were analyzed.

Results: After excluding outliers, 91,257samples remained, which 61162 (58.64%) and 30,095 (41.36%) were female and male, respectively. The mean SCL was 9.36 (9.35, 9.37) mg/dl (95%CI). The highest and lowest SCLs were 3.1 and 18.2mg/dl, respectively. From the total study population, 74127 (81.23%) had normal SCLs, 14110 (15.46%) had hypocalcemia, and 3020 (3.31%) had hypercalcemia. SCLs were normal in 83.6% of men and 79.66% of women. Women had a significantly higher frequency of hypocalcemia compared to men (17.2% vs. 12.83%, $p<0.0001$).

Conclusion: Normal and abnormal SCLs were significantly different in age groups and in both genders. It means that gender and age affect SCLs. Every year of increasing age, reduces the chance of hypercalcemia by 40%, significantly. Age seems to affect hypercalcemia more than hypocalcemia. Age in men increases the risk of hypocalcemia, and reduces the risk of hypocalcemia in women. Therefore, it is recommended to encourage dietary calcium intake among premenopausal women and older men.

Keywords: Calcium; Hypocalcemia; Hypercalcemia; Iran

Introduction

Calcium is one of the essential mineral elements in the body (1-3). The existence of calcium in blood is in three distinct forms: about half is bound to proteins such as albumin and the rest is free or ionized (4). Ionized calcium is tightly regulated by the actions of parathyroid hormone (PTH) and 1,25 dihydroxy vitD (1,25[OH]2D) on the kidney, bone, and gastrointestinal tract (5).

VitD and parathyroid hormone (PTH), calcitonin, and estrogen hormones play an important role in the absorption of calcium, and therefore general health (6, 7).

Excessive calcium intake in the body, especially in the elderly, can cause calcification which is associated with a high risk of cardiovascular disease, brain vascular disease, and end-stage renal disease (ESRD)(8-10). If calcium is consumed more than the body requirement, it binds to bile acid in the gut and increases its secretion(11).

Normal serum calcium level (SCL) is maintained at approximately 8.5 to 10.5 mg/dl range (12). Calcium balance disorders are hypocalcemia (<8.2 mg/dl) and hypercalcemia (>10.4 mg/dl) (13, 14).

The function of the PTH is to increase the SCLs by reducing renal excretion, increasing intestinal absorption of calcium and osteoclasts (15). PTH also indirectly increases intestinal absorption of calcium through converting vitD to calcitriol (16). PTH reduces phosphate reabsorption in the proximal tubule of kidneys while increasing calcium reabsorption in the kidney (17) (Fig.1). By binding to receptors on osteoblasts and osteocytes which promote deposition of bone, PTH promotes the absorption of calcium from the bone (18).

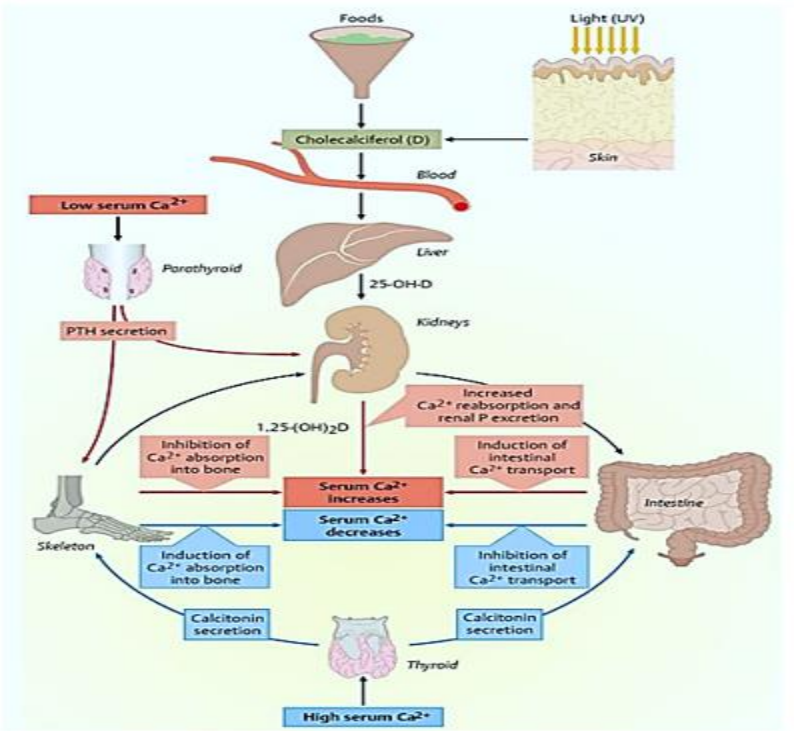

Fig. 1: Three main hormones involved in Calcium homeostasis, PTH (parathyroid), calcitonin (parafollicular cells, thyroid), calcitriol (mainly produced in the kidneys). In low serum Ca2+, the actions of PTH and calcitriol predominate, leading to increased Ca2+ uptake from the gut and bone, and decreased renal Ca2+ excretion. In high serum Ca2+, the action of calcitonin predominates, causing decreased Ca2+ uptake from the gut, increased renal excretion, and storage of excess Ca2+ in bone (38)

Calcitonin, secreted by thyroid parafollicular cells reduces serum calcium through three mechanisms: osteoclasts inhibition, increased renal calcium secretion in the urine, and increased osteoblasts (19, 20).

Estrogen increases bone mineral density by steroid receptors in bone cells and intestinal calcium absorption, blocks the osteoblast's synthesis of interleukin 6, decreases SCL. This hormone has also a protective manner against PHT bone resorption effects (21, 22).

Since calcium deficiency leads to problems related to bone health, risk of fractures, and diabetes in some populations(23), monitoring of calcium intake is essential. Most comprehensive studies in the world have focused on calcium intake and less on SCL.

The purpose of this study was to assess the serum calcium status in Tehran Province, Iran, as a mixed population representing the Iranian population, with a glance at the age and sex over 10 years.

Materials and Methods

Location

Tehran Province with an area of 18,909 square km (7,301m^2) is the capital of Iran and located in the north of the central plateau of the country. It is the most industrialized and the richest province of Iran which contributes approximately 29% of the country's Gross domestic products. By 2021, the population of Tehran was estimated at 9,259,009 (24). Furthermore, the climate of Tehran Province is warm, semi-arid, steppe in the south but cold, semi-humid, and mountainous in the north (24, 25).

Sampling

In this retrospective descriptive study, 105,128 samples were collected during 10 years from 2009-2018. Data were obtained from different laboratories in Tehran collected by the Noor Central Laboratory, Tehran, Iran. We classified data from Tehran Province by the year, age, and gender.

The sera calcium levels were measured according to the CPC colorimetric method using autoanalyzer, Cobas integra 400/700/800 (Roche Diagnostics).

Statistical Analysis

Data analysis was performed using STATA version14 software. Data were described by mean, standard deviation, and 95%CI. Continuous variables were compared between groups using independent T-test or Mann Whitney U-test. Categorical variables were compared between groups using the Chi-square test. Univariate and multivariate multinomial logistic regression models were applied to investigate the associations between the age, gender, and serum calcium.

Ethical Issues/Statement

Informed consent was obtained from all individual participants involved in the study. All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki Declaration and its later amendments or comparable ethical standards.

Results

Data of serum calcium level (SCL) of 105,128 individuals was collected from Tehran Province. Due to the large sample size, the data distribution is considered as normal distribution. After detecting and excluding outliers, we analyzed the remaining 91,257samples, that 61162 (58.64%) were female, 30,095 (41.36%) were male. The mean age of the study subjects was 41.11(±20.71) yr. The mean of SCL was 9.36(9.35, 9.37) mg/dl (95%CI) that were 9.41(9.40, 9.42) mg/dl and 9.33(9.32, 9.33) mg/dl for male and female, respectively (P<0.0001). The highest and lowest SCLs among the study subjects were 3.1 and 18.2mg/dl, respectively. Of the total number of 91257 samples, 74127 (81.23%) had normal SCLs, 14110 (15.46%) had hypocalcemia, and 3020 (3.31%) also had hypercalcemia. In addi-

tion, women had a significantly higher frequency of hypocalcemia compared to men (p<0.0001) (Table1).

Relationship between mean SCLs from 2009 to 2018 years by gender is shown in Fig.2. The mean SCLs in men was significantly higher than women.

Table 1: Normal and abnormal SCL in studied population, by gender

Variable			***Gender***		**P-*value***
			Male	Female	
Calcium status	Normal	Number	30,355	43,772	
		%	83.6	79.66	
	Hypocalcemia	Number	4,658	9,452	
		%	12.83	17.2	<0.0001
	Hypercalcemia	Number	1,296	1,724	
		%	3.57	3.14	
	Total	Number	36,309	54,948	
		%	100	100	

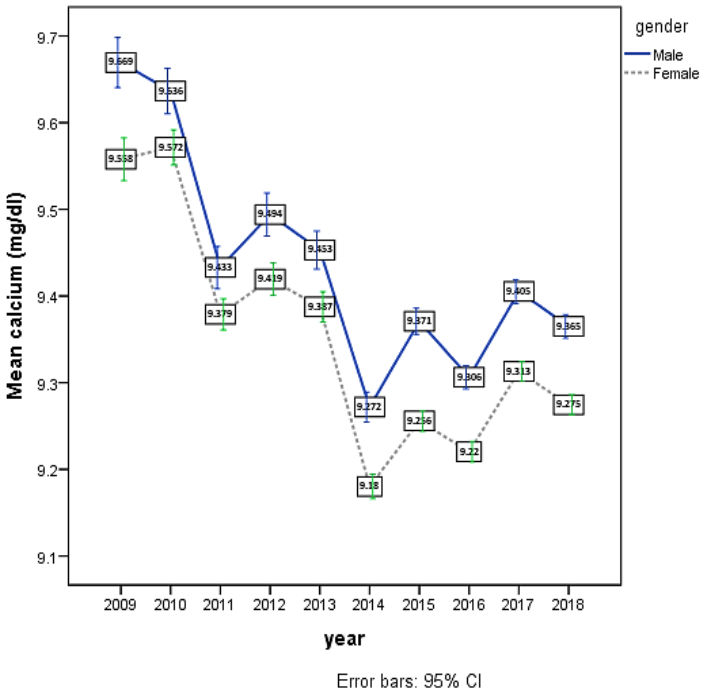

Fig. 2: Serum calcium level of the studied population during 2009-2018, by gender

The difference between the mean SCLs in male and female was significant. In both genders, the highest SCLs were in the age group of 0-10 years. In female, the lowest SCL was in the age group of 31-40 years and has increased since then. In male, the gradual decrease in SCLs continued from birth to old age and its lowest level was in the age group>80 year. Prior to menopause, SCLs in women are significantly lower than in men. But after menopause, SCLs in women increase significantly more than men (Fig. 3).

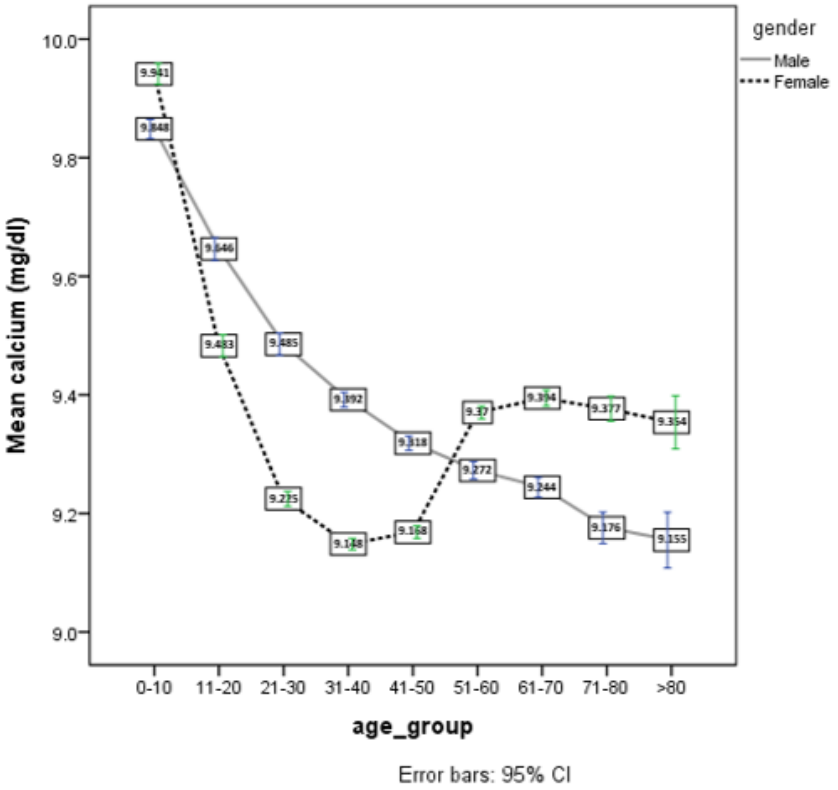

Fig. 3: Mean serum calcium level s in different age groups by gender (from 2009-2018)

In all age groups, there was a statistically difference between men and women regarding SCLs (Table 2). The highest and lowest frequencies of hypercalcemia and hypocalcemia according to the gender were reported in Fig. 4. Figure 5 shows the highest and lowest frequencies of hypercalcemia and hypocalcemia in different years.

Table 2: Serum calcium level s of both genders among different age groups

Age	***Men***			***Women***			
group	Number	Mean	SD*	Number	Mean	SD	P value
<10	4,855	9.85	0.57	4,374	9.94	0.59	0.0001
11-20	2,653	9.65	0.50	2,912	9.48	0.50	0.0001
21-30	3,569	9.48	0.56	6,698	9.22	0.52	0.0001
31-40	6082	9.39	0.51	10366	9.15	0.50	0.0001
41- 50	6,734	9.32	0.51	10,014	9.17	0.53	0.0001
51-60	5,279	9.27	0.54	9,979	9.37	0.55	0.0001
61- 70	4,057	9.24	0.55	6,490	9.39	0.55	0.0001
71-80	2,332	9.17	0.65	3,151	9.38	0.59	0.0001
>80	742	9.15	0.65	957	9.35	0.71	0.0001

*Standard deviation

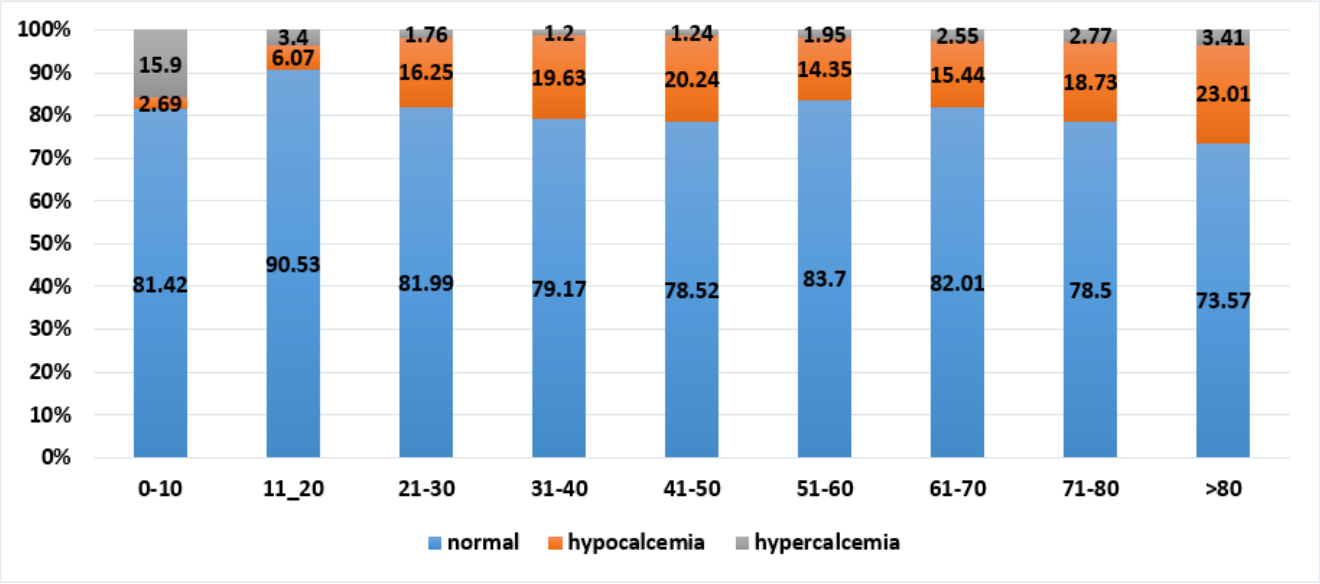

Fig. 4: Frequency of normal and abnormal serum calcium level based on age group

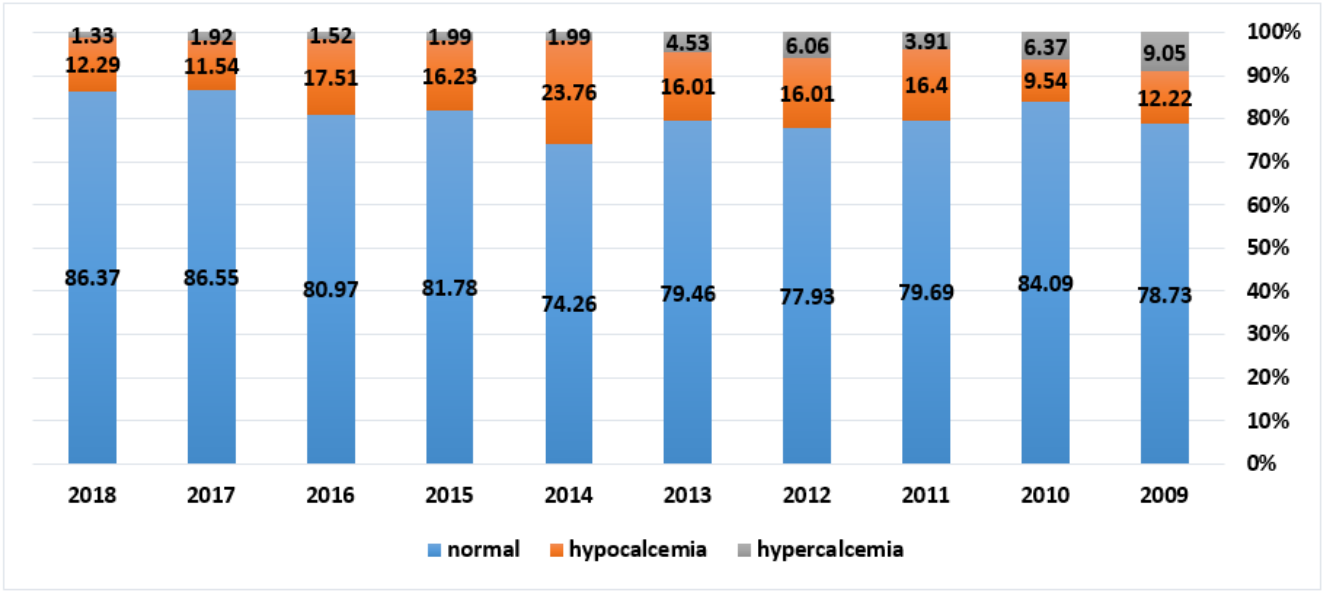

Fig. 5: Frequency of normal and abnormal serum calcium based on different years

During this 10-year period, in all months of the year, the mean SCL in male was significantly higher than female. This means that gender affects the amount of calcium in different months of the year (Fig. 6).

There was a significant relationship between the mean SCL and the mean age of individuals in 2018 and 2016 as well as 2009-2013 years (Fig.7).

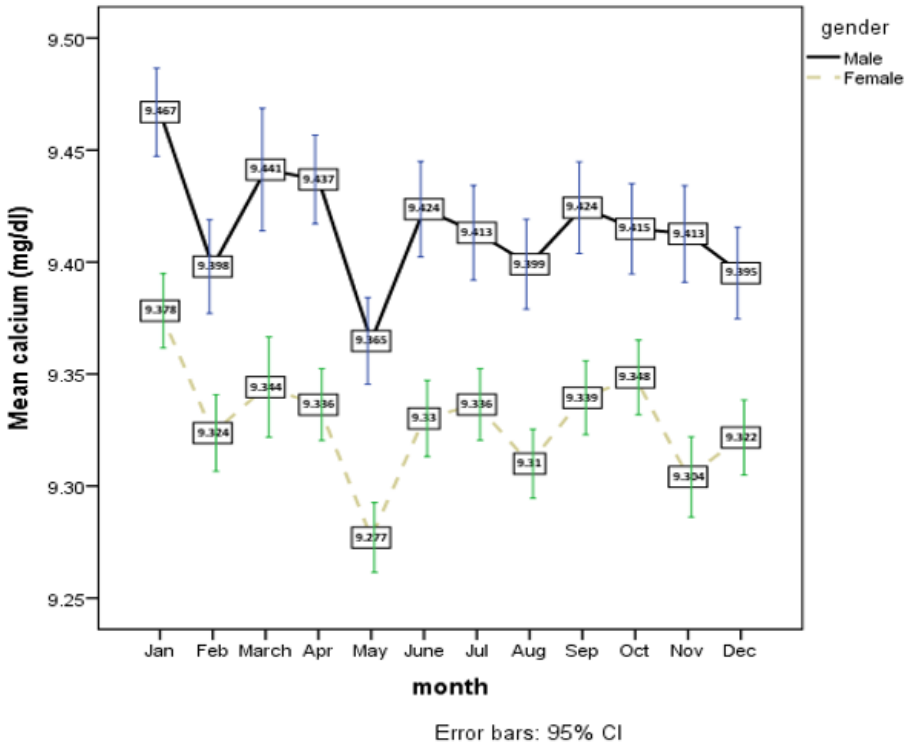

Fig. 6: Mean serum calcium level s in different months (from 2009-2018) in Male and Female

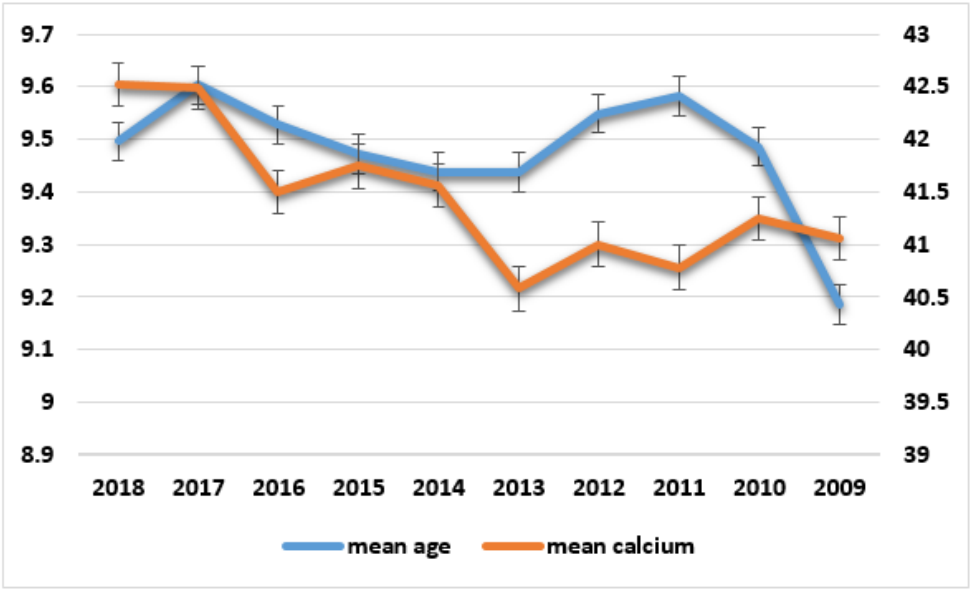

Fig. 7: Relationship between mean serum calcium level and mean age of individuals over a 10-year period (2009-2018)

Hypercalcemia, hypocalcemia, and normal SCLs in different age groups by gender (2009-2018) are shown in Table 3. Normal and abnormal SCLs were significantly different in age groups as well as in both genders. This means that gender and age affect SCLs (Table 3).

Table 3: Frequency of normal and abnormal serum calcium levels in different age groups by gender

SCLs		*Age groups*									*P value*	*Genders*		*P value*
		0-10	11-20	21-30	31-40	41-50	51-60	61-70	71-80	>80		Men	Women	
Normal	no	7,514	5,038	8,418	13,022	13,151	12,771	8,650	4,304	1,250	<0.0001	30,355	43,772	<0.0001
	%	81.42	90.53	81.99	79.17	78.52	83.7	82.01	78.5	73.57		83.6	79.66	
Hypocalcemia	no	248	338	1,668	3,229	3,389	2,190	1,628	1,027	391		4,658	9,452	
	%	2.69	6.07	16.25	19.63	20.24	14.35	15.44	18.73	23.01		12.83	17.2	
Hypercalcemia	no	1,467	189	181	197	208	297	269	152	58		1,296	1,724	
	%	15.9	3.4	1.76	1.2	1.24	1.95	2.55	2.77	3.41		3.57	3.14	
Total	no	9,229	5,565	10,267	16,448	16,748	15,258	10,547	5,483	1,699		36,309	54,948	
	%	100	100	100	100	100	100	100	100	100		100	100	

Regression analysis showed that the female gender increased the risk of hypocalcemia by 1.41 times (41%), significantly. The effect of female gender on hypercalcemia was OR=0.92. In other words, females reduce significantly the risk of hypercalcemia by 8%. Every year of age increases the chance of hypocalcemia by 3% (OR=1.03, p=0.104), while being over 50 years old caused a 40% reduction in the risk of hypercalcemia (OR=0.60, *P*<0.0001) (Table 4).

Table 4: Association of demographic factors with abnormal serum calcium

Demographic factors	*Abnormal calcium*	*Odds ratio*	*P value*	*95% confidence interval*
Effect of female gender	Hypocalcemia	1.41	<0.0001	1.35, 1.46
	Hypercalcemia	0.92	0.032	0.86, 0.99
Effect of age	Hypocalcemia	1.03	0.104	0.99, 1.07
	Hypercalcemia	0.60	<0.0001	0.56, 0.66
Effect of age among men	Hypocalcemia	2.51	<0.0001	2.36, 2.67
	Hypercalcemia	0.37	<0.0001	0.32, 0.44
Effect of age among women	Hypocalcemia	0.62	<0.0001	0.59, 0.65
	Hypercalcemia	0.78	<0.0001	0.71, 0.86

Every year of increasing age reduces the chance of hypercalcemia by 40%, which is also statistically significant. Age seems to affect hypercalcemia more than hypocalcemia. The effect of age in men on hypocalcemia has OR=2.51 (*P*<0.0001). In other words, age in men increases the risk of hypocalcemia by 2.51 times. Thus, aging is a risk factor for men hypocalcemia (Table 4).

Aging reduces the risk of hypocalcemia in women, which can be related to menopause and taking calcium and vitD supplements, and No pregnancy and breastfeeding, etc. The odds ratios for

hypocalcemia were estimated as 2.51 (P<0.0001) in men, and 0.62 (P<0.0001) in women. Besides, the odds ratios for hypercalcemia among men and women were estimated as 0.37 (P<0.0001) and 0.78 (P<0.0001) respectively (Table 5).

Discussion

Calcium ion is essential for many biochemical processes such as blood coagulation, nerve transmission, cell membrane integrity, intracellular signaling, muscle function, vascular contraction and vasodilatation, and many cellular enzymatic activities (25, 26).

Almost all studies on calcium are about calcium intake (27-29). The prevalence of calcium deficiency varies in different parts of the world. According to a study in 2009, only 32% of American adults in 1999–2004 had adequate calcium intake (1). But later in another study, 53% of the US population was reported to consume each of dietary supplement, about 43% consume dietary supplements containing calcium, and also intake of males was significantly higher than females in 2003–2006 (30).

A systematic review in 2017 investigated the average national dietary calcium intake ranges (175-1233 mg/d) data across the 74 countries. Accordingly, Northern European countries had the most calcium intake (>1000 mg/d) while most Asian countries had <500 mg/d of dietary calcium intake, and African countries had calcium intake between 400-700 mg/d (27).

Also, given the importance of calcium intake in children, adequate calcium in their diet requires an appropriate strategy. A study in Iran also assessed calcium intake in 501 school-aged children, which found that dairy products provide 69.3% of the daily calcium intake of children and only 17.5% of them consumed adequate calcium (31).

Our study sought to investigate the SCL, as an important mineral in the body, and it correlates with age and gender during10-years in Tehran. Furthermore, no study with such a population has been reported previously in our country. This study showed that the mean SCL was 9.36(9.35, 9.37) mg/dl, which according to the reference range is the normal, the mean serum calcium in men was higher than in women (Table 5).

Table 5: Comparison of average blood calcium level of Tehran population, by gender and year (2009-2018)

Year	***Total***			***Men***			***Women***			**P *value***
	Number	Mean	SD	Number	Mean	SD	Number	Mean	SD	
2009	5,279	9.60	0.69	2,200	9.67	0.69	3,079	9.55	0.69	<0.0001
2010	6,197	9.60	0.64	2,473	9.64	0.66	3,724	9.57	0.63	0.0001
2011	7,490	9.40	0.64	2,981	9.43	0.68	4,509	9.37	0.62	0.0004
2012	8,155	9.45	0.69	3,195	9.49	0.71	4,960	9.41	0.67	<0.0001
2013	8,519	9.41	0.64	3,308	9.45	0.64	5,211	9.39	0.64	<0.0001
2014	10,018	9.22	0.56	3,933	9.27	0.55	6,085	9.18	0.56	<0.0001
2015	11,308	9.30	0.51	4,366	9.37	0.50	6,942	9.25	0.50	<0.0001
2016	12,196	9.25	0.50	5,095	9.31	0.49	7,101	9.22	0.51	<0.0001
2017	12,050	9.35	0.50	4,726	9.40	0.48	7,324	9.31	0.50	<0.0001
2018	10,045	9.31	0.45	4,032	9.36	0.44	6,013	9.27	0.45	<0.0001
Total	91,257	9.36	0.58	36,309	9.41	0.58	54,948	9.33	0.58	<0.0001

- Standard deviation

The total calcium during 10 year-period indicates a steady decline in SCLs in male and female. With the Intensification of sanctions in years 2010 and 2011, the first decrease in calcium occurred, and in 2014 it reached its lowest level in ten years. It also resulted in significantly decreased people's

purchasing power food and drug products. On the other hand, economic problems due to the sanction have crippled Iran's domestic pharmaceutical, drug production, import of medicines, and essential raw materials. As a result of the drug shortage, the consumption of medicines and dietary supplements has decreased.

Women in age group 31-40 year showed the lowest SCL but still in the normal range. With the onset of menopause among women, the lowest amount of calcium is occurred (32). But after the age of 50-60 years it can be seen that the graph is upward sloping and then almost reaches a stable plateau. Among the reasons for lower SCLs in pre-menopausal women than men the following can be mentioned: 1. Pregnancy and lactation, 2. Inadequate consumption of dairy products and supplements such as calcium and vitD, 3. VitD deficiency, 4. The type of clothing in women that can lead to less synthesis of vitD from sunlight, 5. Women's workplace, which is mainly indoors and away from sunlight.

In the United States, about 50% of women over 50 use calcium supplements (33). The diversity of lifestyle, dietary habits and the amount of dairy consumption in the capital, as an example of the whole community, have an impact on the amount of essential minerals in the body.

The reasons for a significant increase in SCLs in postmenopausal women compared to men are the following: 1. No Pregnancy and lactation 2. Take supplements such as calcium and vitD, 3. Pay more attention to improving the quality of lifestyle such as increasing physical activity and exercise, 4. Changing the type of clothes, 5. More exposure to sunlight (retirement ...), etc.

Estrogen decreases SCLs and increases calcium storage in bones through enhancing intestinal calcium absorption (34, 35). Besides, usually the age of pregnancy and lactation can be one of the causes of a decrease in serum calcium.

The most subjects with hypocalcemia in this study were aged more than 80, as well as 41-50 years. In addition, the highest rates of hypercalcemia were observed in age group 0-10 year. The reasons can be related to different nutrition in different periods of life, exposure to sunlight, supplement therapy in different age groups, physical activity, other diseases, etc.

Comparing the trends of mean age and calcium levels during the years of the study period shows a slightly negative correlation during the time. It seems that the trend of the calcium levels from 2009 to 2018 can be partially explained by the changes in the age of the study participants. However, the role of the other factors such as gender, climate change, co-morbidities, and the cause of subjects referring to the study laboratory should be taken into consideration, since they are not involved in this survey.

To increase the consumption of calcium dairy products, as the best source of calcium (36), and non-dairy sources, we need nutritional training programs and strategies. The best non-dairy sources of calcium can be salmon, tuna and sardines, orange juice, dark, leafy greens such as kale, spinach, cabbage, broccoli are also high in calcium, nuts (Out of all the nuts, almonds are up there with the highest amount of calcium), green beans, which should be added to people's diet (37).

Regression analyses showed that aging in men is a risk factor for hypocalcemia, while reduced the risk of hypocalcemia among women. The reason can be menopause, and taking calcium and vitD supplements, no pregnancy and lactation, etc. in women.

Due to the large sample size of the current study, many estimates were observed statistically significant. However, it is necessary to consider the clinical importance of the results instead of statistical significance. In addition, our study sample was selected from one of the main laboratory clinics in the Capital of Iran. We assumed that these samples can be representative of the whole country.

Conclusion

The mean level of serum calcium among the population in the Capital of Iran was in the normal range, less than 20% of them had abnormal levels of serum calcium. We also provided some

evidence that the aging can be a risk factor for hypocalcemia among men and a protective factor in women.

It is recommended that the government provides easy access to dietary supplements especially calcium-vitD. Encouraging people to exercise, especially public exercise, can also help regulate blood calcium and keep bones healthy.

For lifelong bone health, strengthening bones, preventing osteoporosis and other calcium-related diseases, it is important to take into account the amount of calcium. More extensive studies on serum calcium and the factors affecting it are recommended, especially in other provinces.

Journalism Ethics considerations

Ethical issues (Including plagiarism, informed consent, misconduct, data fabrication and/or falsification, double publication and/or submission, redundancy, etc.) have been completely observed by the authors.

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Conflict of interest

The authors declare that they have no conflict of interest

References

1. Bailey RL, Dodd KW, Goldman JA, Gahche JJ, Dwyer JT, Moshfegh AJ, Sempos CT, Picciano MF (2010). Estimation of total usual calcium and vitamin D intakes in the United States. *J Nut,* 140:817-822.
2. Health UDo, Services H (2004). Bone health and osteoporosis: a report of the Surgeon General. *Rockville, MD: US Department of Health and Human Services, Office of the Surgeon General,* 87.
3. Ross AC, Taylor CL, Yaktine AL, Del Valle HB (2011). *Institute of Medicine (US) Committee to Review Dietary Reference Intakes for Vitamin D and Calcium. Dietary reference intakes for calcium and vitamin D*. Washington, DC: National Academies Press.
4. Siyam FF, Klachko DM (2013). What is hypercalcemia? The importance of fasting samples. *Cardiorenal Med,* 3:232-238.
5. Shaker JL, Deftos L (2018). Calcium and phosphate homeostasis. *Endotext [Internet]*. South Dartmouth (MA): MDText.com, Inc.; 2000.
6. DeLuca HF (2008). Evolution of our understanding of vitamin D. *Nutr Rev,* 66:S73-S87.
7. Farhud DD, Mehrabi A, Sarafnejad A, et al (2019). A comprehensive, epidemiological and ecological descriptive study on vitamin d status in Iran (308005 People, from 2009–2018). *Iran J Public Health,* 48:644.
8. Chen NC, Hsu CY, Chen CL (2017). The strategy to prevent and regress the vascular calcification in dialysis patients. *BioMed Res Int,* 2017.
9. Kraus MA, Kalra PA, Hunter J, Menoyo J, Stankus N (2015). The prevalence of vascular calcification in patients with end-stage renal disease on hemodialysis: a cross-sectional observational study. *Ther Adv Chronic Dis,* 6:84-96.
10. Bos D, Ikram MA, Elias-Smale SE, et al (2011). Calcification in major vessel beds relates to vascular brain disease. *Arterioscler Thromb Vasc Biol,* 31:2331-2337.
11. Bostick RM, Kushi LH, Wu Y, Meyer KA, Sellers TA, Folsom AR (1999). Relation of calcium, vitamin D, and dairy food intake to ischemic heart disease mortality among postmenopausal women. *Am J Epidemiol,* 149:151-161.
12. Walker HK, Hall WD, Hurst JW (1990). *Clinical methods: the history, physical, and laboratory examinations*. Boston: Butterworths; 1990. ISBN-10: 0-409-90077-X.
13. Cooper MS, Gittoes NJ (2008). Diagnosis and management of hypocalcaemia. *BMJ,* 336:1298-1302.
14. Frølich A (1998). Prevalence of hypercalcaemia in normal and in hospital populations. *Dan Med Bull,* 45:436-439.

15. Moe SM (2008). Disorders involving calcium, phosphorus, and magnesium. *Prim Care,* 35:215-237.
16. Del Valle HB, Yaktine AL, Taylor CL, Ross AC (2011). *Dietary reference intakes for calcium and vitamin D.* Washington (DC): National Academies Press (US); 2011.
17. Prasad N, Bhadauria D (2013). Renal phosphate handling: Physiology. *Indian J Endocrinol Metab,* 17:620.
18. Bellido T, Saini V, Pajevic PD (2013). Effects of PTH on osteocyte function. *Bone,* 54:250-257.
19. Copp DH, Cheney B (1962). Calcitonin—a hormone from the parathyroid which lowers the calcium-level of the blood. *Nature,* 193:381-382.
20. Felsenfeld AJ, Levine BS (2015). Calcitonin, the forgotten hormone: does it deserve to be forgotten? *Clin Kidney J,* 8:180-187.
21. Bansal N, Katz R, de Boer IH, et al (2013). Influence of estrogen therapy on calcium, phosphorus, and other regulatory hormones in postmenopausal women: the MESA study. *J Clin Endocrinol Metab,* 98:4890-4898.
22. Kassem M, Harris SA, Spelsberg TC, Riggs BL (1996). Estrogen inhibits interleukin-6 production and gene expression in a human osteoblastic cell line with high levels of estrogen receptors. *J Bone Miner Res,* 11:193-199.
23. Beto JA (2015). The role of calcium in human aging. *Clinical Nutrition Research,* 4:1-8.
24. Anonymus (2021) URL. Available from: http://www.citypopulation.de/en/world/agglomerations/
25. Starki S, Loebis B, Husada SM, Mahdinasari NN, Efffendy E (2021). Calcium Levels Differences in Men with Schizophrenia Treated with Olanzapine and Risperidone in Prof. Dr. M Ildrem Psychiatric Hospital Medan. *Open Access Macedonian Journal of Medical Sciences,* 9:274-279.
26. Thongprayoon C, Cheungpasitporn W, Chewcharat A, Mao MA, Thirunavukkarasu S, Kashani KB (2020). Hospital mortality and long-term mortality among hospitalized patients with various admission serum ionized calcium levels. *Postgrad Med,* 132:385-390.
27. Balk E, Adam G, Langberg V, et al (2017). Global dietary calcium intake among adults: a systematic review. *Osteoporos Int,* 28:3315-3324.
28. Nicklas TA, O'Neil CE, Fulgoni III VL (2009). The role of dairy in meeting the recommendations for shortfall nutrients in the American diet. *J Am Coll Nutr,* 28:73S-81S.
29. Novotny R, Boushey C, Bock MA, et al (2003). Calcium intake of Asian, Hispanic and white youth. *J Am Coll Nutr,* 22:64-70.
30. Bailey RL, Gahche JJ, Lentino CV, et al (2011). Dietary supplement use in the United States, 2003–2006. *J Nutr,* 141:261-266.
31. Omidvar N, Neyestani T-R, Hajifaraji M, Eshraghian M-R, Rezazadeh A, Armin S, Haidari H, Zowghi T (2015). Calcium intake, major dietary sources and bone health indicators in Iranian primary school children. *Iran J Pediatr,* 25.
32. Bone HG, Bolognese MA, Yuen CK, Kendler DL, Wang H, Liu Y, San Martin J (2008). Effects of denosumab on bone mineral density and bone turnover in postmenopausal women. *J Clin Endocrinol Metab,* 93:2149-2157.
33. Covell LL, Ganti AK (2015). Treatment of advanced thyroid cancer: role of molecularly targeted therapies. *Target Oncol,* 10:311-24.
34. Pravina P, Sayaji D, Avinash M (2013). Calcium and its role in human body. *International Journal of Research in Pharmaceutical and Biomedical Sciences,* 4:659-668.
35. Van Cromphaut S, Rummens K, Stockmans I, et al (2003). Intestinal calcium transporter genes are upregulated by estrogens and the reproductive cycle through vitamin D receptor-independent mechanisms. *J Bone Miner Res,* 18:1725-1736.
36. Rafferty K, Walters G, Heaney R (2007). Calcium fortificants: overview and strategies for improving calcium nutriture of the US population. *J Food Sci,* 72:R152-R158.
37. Mäkinen OE, Wanhalinna V, Zannini E, Arendt EK (2016). Foods for special dietary needs: Non-dairy plant-based milk substitutes and fermented dairy-type products. *Crit Rev Food Sci Nutr,* 56:339-349.
38. Maxwell JE, Sherman SK, O'Dorisio TM, Howe JR (2014). Medical management of metastatic medullary thyroid cancer. *Cancer,* 120:3287-301.