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Original research article: Study of serum levels of vitamin D3 In females of hypothyroidism

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Abstract
Background: Thyroid is an endocrine gland which is present in the neck below the Adam's apple and secretes thyroid hormones i.e. T3, T4, TSH and calcitonin. Vitamin D is fat soluble and also known as sunshine vitamin. It is of 2 types D3 and D2. Vitamin D3 (cholecalciferol) is found in animal products and Vitamin D2 (ergocalciferol) is found in plants. Both are converted into active form of Vitamin D by a series of pathway in the body and the active form i.e. calcitriol is circulated in the blood and combines with the Vitamin D receptor (VDR). The VDR function with the help of calcitonin which is a hormone secreted by thyroid gland. The receptors of thyroid and vitamin D are steroid hormone receptors.
Objectives: The Objectives of the study is to determine the level of Vitamin D3 in female patients of hypothyroidism.
Methodology: In the hypothyroid patients, the level of T3 and T4 is low and level of TSH is high. The level of vitamin D and Serum calcium decreases in hypothyroidism. The detection of T3, T4, TSH and Vitamin D is done by an automated machine known as siemens Advia centaur XP which works on the principle of chemiluminescence. The serum calcium is analyzed by Beckman coulter which works on the principle of calorimeter. 2 groups were formed, 50 subjects in study group which were hypothyroid and vitamin D deficient and 50 subjects in control group which were normal.
Results: On the basis of 3 age ranges i.e. 1-20, 20-40 and above 40 were showing hypothyroidism in vitamin D deficiency. Another criteria on the basis of diet was studied which shows that non-vegetarians have low absorption of vitamin D with hypothyroidism because the meat contains fat which causes hindrance in the absorption.
Conclusion: From our study I would like to conclude that the levels of FT3 and FT4 were low and TSH was high, serum Calcium was low and vitamin D was low as compared to the normal healthy subjects. There is significantly high level of TSH in case group as compared to control group. The above study also concludes that vitamin d supplements should be given to hypothyroid patients and vegetarians are healthier than non-vegetarians.

Keywords: hypothyroidism, Vit D, T3, T4, TSH

Introduction
Vitamin D is a fat soluble hormone which increases absorption of calcium and magnesium in intestine. The most important compounds in this group are vitamin D3 (also known as cholecalciferol) and vitamin D2 (ergocalciferol). Cholecalciferol (vitamin D3) is found mainly in animal and its products like milk, cheese and some fish. Ergocalciferol (vitamin D2) is found mainly in plants like alfalfa and fungi like mushroom. Both of these hormones are collectively referred to as vitamin D, and they can either be obtained in two ways. One is by exposure of the skin to the ultraviolet (UV) rays of sunlight or also from dietary intake. Vitamin D is found naturally in fish (such as salmon and sardines) and fish oils, eggs and cod liver oil [1].
The thyroid gland is an endocrine gland in the neck, consisting of two lobes connected by an isthmus. It is found at the front of the neck, below the Adam's apple. The thyroid gland secretes thyroid hormones, which primarily influence the metabolic rate and protein synthesis. The thyroid hormones triiodothyronine (T3) and thyroxine (T4) are created from iodine and tyrosine. The thyroid also produces the hormone calcitonin, which plays a role in calcium homeostasis [2, 3].
Both vitamin D and thyroid hormone bind to similar receptors called steroid hormone receptors.
McDonnell *et al*. found a strong homology between the molecular structure of vitamin D3 receptor and the receptor for thyroid hormone, which was due to two regions that they have in common: the first is a 70 amino acid, cysteine-rich sequence and the second region is a 62 amino acid one located towards the carboxyl terminus of the proteins [4].

Material and Method
The present study was carried out on confirmed hypothyroid and vitamin D deficient patients, attending outdoor Biochemical Laboratory, Department of Biochemistry, SMS Medical College and Hospital,
Jaipur from jan 2017-june 2017 for periodic check up of their thyroid hormone levels and Vitamin D levels in the blood. The basis of study was absolutely biochemical in nature.
The subjects of study was divided into the following groups:-

- **Control groups**: This comprise of 50 euthyroid and healthy subjects of different age groups, who were having their thyroid hormone levels and Vitamin D levels within normal range.

- **Study groups:** The study group was further divided into following subgroups
  1. Hypothyroid and vitamin D deficient patients:- This group comprised of 50 hypothyroid patients having vitamin D deficiency
  2. Physical examination:- included age, sex, diet, habit like smoking, alcohol and tobacco chewing and menstrual history.
- The above thyroid hormones and vitamin D levels were analysed by seimens advia centaur XP.
- The serum calcium was analysed by beckman coulter.

Results

Table 1: showing Mean, S.D., T value, P-value in normal and hypothyroid and vitamin D deficient patients

	FT3(pg/ml)	FT4(ng/dl)	TSH(uIU/ml)	Serum CalciuM(mg/dl)	Vitamin D
Patient (n=50) Mean±S.D.	2.4484±0.530349567	0.70868±0.230776368	8.2722±11.23002196	8.1188±0.910980447	12.7196±6.36902171
Control (n=50) Mean±S.D	2.7068±0.31942193	1.1516±0.1683941	2.9046±1.412767165	8.9628±0.53962209	32.299±11.79883
T-Value	0.003014897	3.76977E-14	0.000565676	4.11188E-07	8.245E-14
P-Value	0.498809	0.00022	<0.00001	0.000075	<0.00001

Table 2: Showing Mean, SD, T- Value, P- Value of thyroid hormone, serum calcium and vitamin D in both deficient and normal subjects with age range of 1-20 yr.

	FT3(pg/ml)	FT4(ng/dl)	TSH(uIU/ml)	Serum Calcium(mg/dl)	Vitamin D
Patient (n=1) Mean±S.D.	3.38	0.8	6.69	8.95	5.95
Control (n=7) Mean±S.D	2.7757±0.3613	1.158±0.123	3.324±1.392	8.92±0.40743	37.295±20.5926
T-Value	2.353	2.353	2.353	2.353	2.353
P-Value	0.050016	0.050016	0.050016	0.050016	0.050016

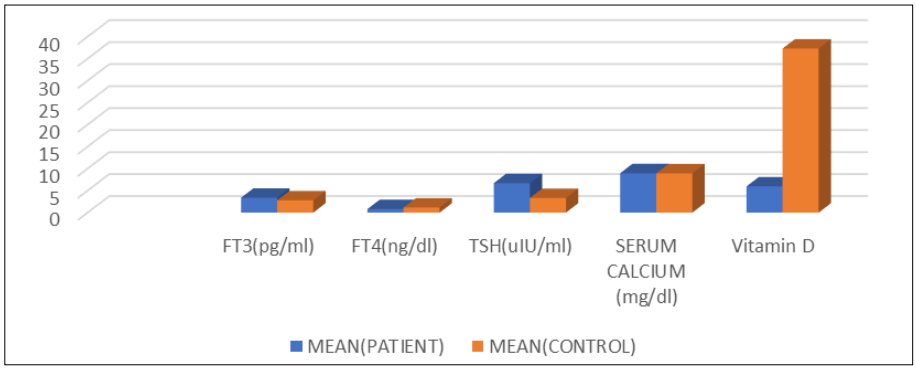

Fig 1: Graphical presentation of thyroid hormone, serum calcium and vitamin D in both deficient and normal subjects with age range of 1-20 yr.

Table 3: showing Mean, SD, T- Value, P- Value of thyroid hormone, serum calcium and vitamin D in both deficient and normal subjects with age range of 20-40 yr

	FT3(pg/ml)	FT4(ng/dl)	TSH(uIU/ml)	Serum Calcium(mg/dl)	Vitamin D
Patient (n=25) Mean±S.D.	2.4176±0.401	0.68256±0.27061	6.9852±4.43244	7.966±1.05961	13.1216±4.29541
Control (n=15) Mean±S.D	2.668± 0.28456	1.131333333±0.2014897	2.868666667±1.480665	9.178666667±0.766969	29.24466667±7.320301386
T-Value	0.020516722	1.164E-06	0.000661186	0.00021412	5.09865E-11
P-Value	0.491907	0.129416	.499764	.499921	0.000032

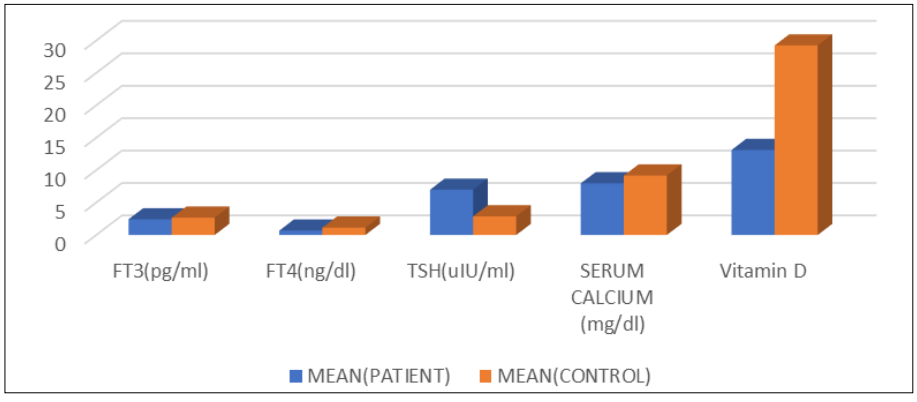

Fig 2: Graphical presentation of thyroid hormone, serum calcium and vitamin D in both deficient and normal subjects with age range of 20-40 yr.

Table 4: showing Mean, S.D, T- Value, P- Value of thyroid hormone, serum calcium and vitamin D in both deficient and normal subjects with

age range above 40 yr.

	FT3(pg/ml)	FT4(ng/dl)	TSH(uIU/ml)	Serum Calcium(mg/dl)	Vitamin D
Patient (n=24) Mean±S.D.	2.441666667±0.61710	0.732083333±1.88079	9.67875±15.45289	8.24333±0.708941	12.58291667±7.957024
Control (n=28) Mean±S.D	2.710357143±0.334791	1.160714286±0.1618	2.818928571±1.4145	8.857857143±0.38571	32.68607143±10.9001
T-Value	0.026063106	4.36859E-12	0.011618153	0.000119604	5.29158E-10
P-Value	0.489732	0.000096	0.495418	.499961	<. 00001

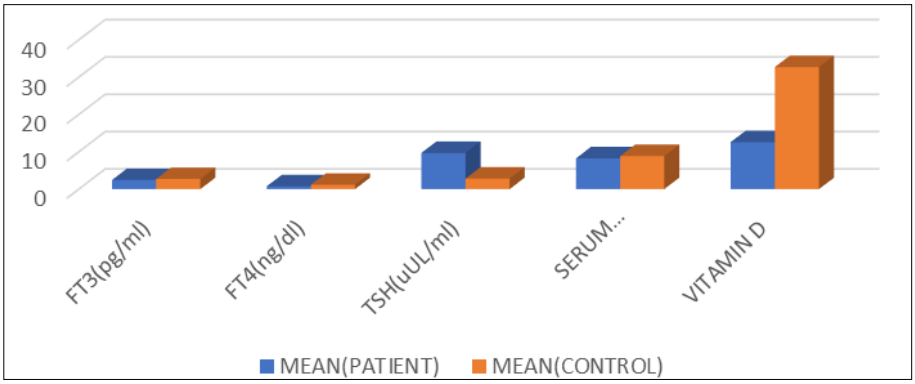

Fig 3: Graphical presentation of thyroid hormone, serum calcium and vitamin D in both deficient and normal subjects with age range above 40 yr.

Table 5: Comparison between normal subjects on the basis of vegetarian and non- vegetarian diet.

	DIET	FT3(pg/ml)	FT4(ng/dl)	TSH(uIU/ml)	Serum Calcium (mg/dl)	Vitamin D
mean±S.D. n= 10	NON VEG	2.84909±0.3656	1.26±0.2112	2.7863±1.3112	8.99±0.3446	29.72±6.3534
mean±S.D. n = 40	VEG	2.66 ±0.2987	1.121±1.1389	2.93±1.4547	8.95±0.5865	33.024±12.901
T value		0.047318789	0.007019061	0.378430195	0.425092865	0.209341722
P value		0.481333	0.497236	0.35423	0.337311	0.41799

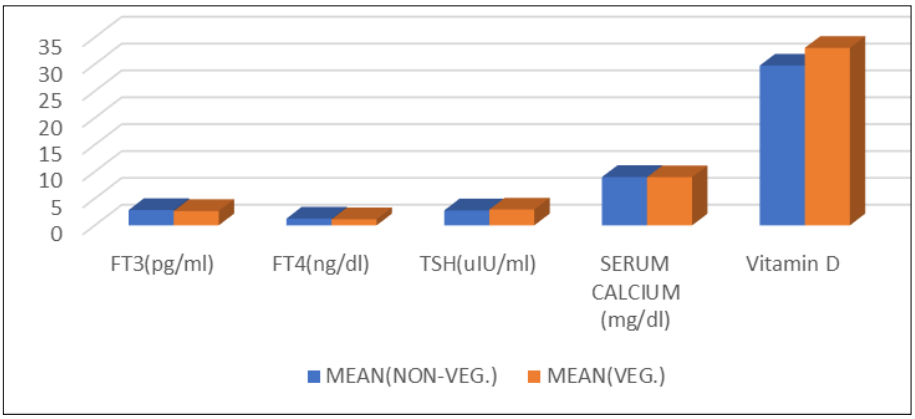

Fig 4: Graphical presentation of Comparison between normal subjects on the basis of vegetarian and non- vegetarian diet.

Table 6: Comparison between deficient patient on the basis of vegetarian and non- vegetarian diet.

	DIET	FT3(pg/ml)	FT4(ng/dl)	TSH(uIU/ml)	Serum Calcium (mg/dl)	Vitamin D
Mean ±S.D (n=18)	NON VEG	2.5272±0.480171	0.7224±0.35641	6.7144 ±2.8349	8.1394±0.7756	11.2472±11.3473
Mean ±S.D (n=32)	VEG	2.404062±0.536541	0.7009375 ±0.220781	9.1484375±13.777	8.1071875±0.9742	13.5478125±5.7316
T Value		0.215825665	0.377672476	0.231536865	0.452601398	0.109602812
P Value		.415484	0.354523	0.409444	0.327451	0.456819

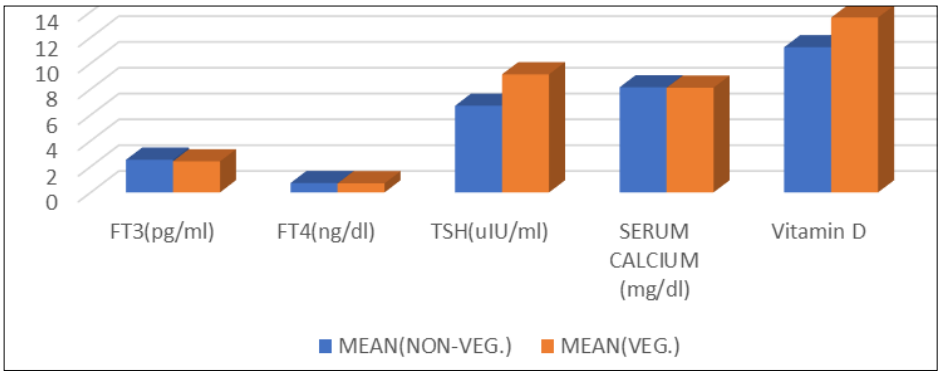

Fig 5: Comparison between deficient patient on the basis of vegetarian and non- vegetarian diet.

Discussion

Present study is carried out on 50 control and 50 hypothyroid patients with vitamin D deficiency attending outdoor central laboratory of Department of Biochemistry, SMS Medical College and Hospital, Jaipur for periodic checkup of their thyroid hormone, serum calcium and vitamin D levels. It was observed that none of the hypothyroid subjects in table no.1 and the respective graphs had sufficient vitamin D levels as compared to the subjects showing normal thyroid levels, serum calcium and vitamin D. Table no.2, 3, 4 and their respective graphs showing different age group also have hypothyroidism with vitamin D deficiency which shows that all age groups are affected by thyroid hormone dysfunction. Vitamin D deficiency negatively correlates with TSH and the severity of vitamin D deficiency corresponds to the severity of thyroid disease.

There was a significant decrease in the levels of Vitamin D amongst hypothyroid patients. In a study conducted by Shilpa *et al* in 2014, there were 56% of the hypothyroid subjects in whom Vitamin D levels were below 20.

There were only 10% subjects who had sufficient levels of Vitamin D. Deficiency of Vitamin D can lead to Grave's disease and various other autoimmune thyroid disorders [2].

Table no.5 are showing normal subjects on the basis of their non-vegetarian and vegetarian diet respectively.

Similarly, Table no.6 are showing hypothyroid and vitamin D Deficient patients on the basis of their non-vegetarian and vegetarian diet respectively.

Though vitamin D is fat soluble but fat or meat in the diet creates hindrance in its absorption. Red meat is a valuable source of vitamin D, and the vitamin is in absorbable form, but according to Mawer and Davies in 2001, meat causes mal-absorption of vitamin D by some mechanism so far unknown [5, 6]. The above study relates with our results which shows that hypothyroid and vitamin D deficient patients who eat non-vegetarian food have less vitamin D level and who eat vegetarian food have more vitamin D level. In 2014, another study by McDonnell *et al*. found a strong homology between the molecular structure of vitamin D3 receptor and the receptor for thyroid hormone, which was due to two regions that they have in common: the first is a 70 amino acid, cysteine-rich sequence and the second region is a 62 amino acid one located towards the carboxyl terminus of the proteins. In particular, low vitamin D concentrations, certain vitamin D receptor (VDR) gene polymorphisms and pathologies of vitamin D-binding proteins and of their gene may favor the development of Hashimoto's thyroiditis (HT) [7, 8]. It may be suggested that vitamin D be supplemented to all hypothyroid patients (Nirensingh Koch *et al*, International Journal of Clinical Biochemistry and Research 2016).Thus, all the above observations of the present study are in conformity with those of Nirensingh Koch *et al* (2016).

Conclusion

FROM our study I would like to conclude that the levels of FT3 and FT4 were low and TSH was high, serum calcium was low and vitamin D was low as compared to the normal healthy subjects. There is significantly high level of TSH in case group as compared to control group. The above study also concludes that vitamin d supplements should be given to hypothyroid patients and vegetarians are more healthy than non-vegetarians.

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