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## **Original Research Article**

# A Study of Serum Calcium in Vitiligo Patients

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**Conflict of interest: Nil** 

#### Abstract

**Introduction:** Vitiligo is an acquired disorder of the skin and mucous membranes that is characterized by well circumscribed, depigmented macules, and patches, secondary to selective destruction of melanocytes.

Vitamin D is essential hormone that is synthesised in the skin that regulates calcium and bone metabolism, controls cell-proliferation and differentiation. Low levels of vitamin D and calcium have been observed in vitiligo patients.

**Aims & Objectives:** (1) To estimate serum calcium levels in vitiligo patients. (2) To compare serum calcium levels in patients and controls.

**Material & Methods:** 30 vitiligo cases and 30 apparently healthy controls who are age and sex matched are included in the study. Estimation of serum calcium was done by OCPC method using kit from ERBA Manheim on ERBA chem 5 plus V2 semiauto analyser.

**Results & Conclusions:** Our study shows low levels of serum Calcium in Vitiligo patients compared to controls (P<0.05). Adequate supplementation of Vitamin D3 and calcium is required to prevent melanocyte loss in vitiligo and stimulate melanin production.

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#### Introduction

Vitiligo is an acquired disorder of the skin and mucous membranes that is characterized by well circumscribed, depigmented macules and patches secondary to selective destruction of melanocytes Vitamin D is an essential hormone that is synthesized in the skin that regulates calcium & bone metabolism, controls cell proliferation and differentiation and exerts immunoregulatory activities. Low levels of vitamin D have been observed in vitiligo patients. With this background information, the present study is designed to assess the role of serum calcium in the pathogenesis of vitiligo patients.

Review of literature: Vitamin D might limit the melanocyte loss in vitiligo by counteracting the local immune process in the skin, oxidative stress, programmed cell death, and aberrant calcium fluxes. Vitamin D plays a role in calcium regulation by vitamin D, receptors on melanocytes or by regulation of defective calcium homeostasis.

Defective Calcium transport has been shown in keratinocytes and melanocytes obtained from vitiliginous skin samples. Decreased intracellular Ca leads to high levels of reduced thioredoxin, which inhibits tyrosinase activity and results in inhibition of melanin synthesis Therefore,

decreased calcium levels could be one of cause in melanocyte loss.

# Objectives of the study

- To estimate serum calcium levels in vitiligo patients.
- To compare serum calcium levels in vitiligo patients & healthy controls

## Materials and Methods

**Source of data:** All vitiligo cases of both sexes and apparently healthy controls who are age and sex matched will be included in the study. Cases will be those attending dermatology department in BRIMS. Bidar. The study groups were divided into 2:

- Group 1-30 healthy controls.
- Group 2-30 patients with vitiligo.

**Inclusion Criteria:** 15-50 Age group years. All cases of vitiligo irrespective of the type and duration. Apparently healthy controls.

#### **Exclusion Criteria:**

- Children<9years.
- Persons with chronic diseases like Hypertension, Tuberculosis, Renal impairment, Chronic liver disease, Parathyroid disease,

Osteomalacia, Rickets and Metabolic bone disease.

- Those who have undergone phototherapy within 1 month prior to recruitment.
- Persons taking oral calcium supplements, vitamin D analogues, calcium channel blockers or systemic steroids or any other medications which can alter the study parameter.

Method of collection of data (including sampling procedure): After taking informed consent from study participants, a complete history will be taken including age of onset, duration, family history of vitiligo and past history of common. systemic diseases associated with vitiligo like anaemia, alopecia areata, thyroid dysfunction and diabetes mellitus. A thorough dermatological examination will be done. Under all aseptic precautions about 3ml of venous blood will be collected in a plain bulb (for serum). Serum will be separated by centrifugation and stored at -4°C until analysis is done.

# Sample Size:

- 30 cases
- 30 controls.

The following estimations will be done with the samples:

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#### **Estimation**

a) Estimation of Serum calcium by OCPC method: Principle: O-Cresolphtalein complex zone reacts with calcium in alkaline medium to form a purple coloured complex. The intensity of purple colour is proportional to the calcium concentrate and is measured photometrically between 540nm and 600nm with maximum absorbance at 575nm.

**Type of study:** This will be a case control study.

**Statistical Analysis:** The statistical package for the social science (SPSS version 11.5 for Windows and MS-EXCEL) will be used for statistical analysis. Results will be expressed as mean  $\pm$  SD.

For comparison between cases and controls, Unpaired student 't' test and chi-square test will be used and P value less than 0.5 will be considered to be statistically significant.

#### Results

Table 1: Age wise distribution of	of vitiligo	<u>patients a</u>	nd controls
			~ -

Age in years	Vitiligo patients		Controls		Total	
	No.	<b>%</b>	No.	%	No.	%
≤ 20	5	16.7	4	13.3	9	15.0
21-30	9	30.0	9	30.0	18	30.0
31-40	7	23.3	8	26.7	15	25.0
41-50	4	13.3	5	16.7	9	15.0
51-60	5	16.7	4	13.3	9	15.0
Total	30	100.0	30	100.0	60	100.0
$Mean \pm SD$	$33.03 \pm 13.15$ $34.23 \pm 11.54$ $33.6$		33.61 ±	12.34		
t-test value; P-value & Significance	t = 0.376, $P = 0.709$ , NS					

NS= not significant, S=significant, HS=highly significant, VHS=very highly significant

Study observed that, Maximum number of cases 33(55.0%) belongs to the age group of 21-40 years. There was no statistical significance difference of age among vitiligo patients and controls (P>0.05).

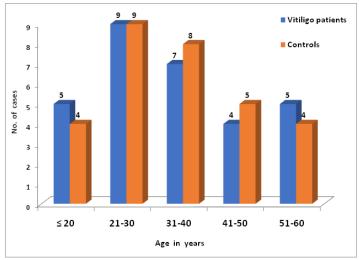


Figure 1: Multiple bar diagram represents age wise distribution of vitiligo patients and controls

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Table 2: Sex wise distribution of vitiligo patients and controls

Age in years	Vitilig	Vitiligo patients		Controls		Total	
	No.	%	No.	%	No.	%	
Males	12	40.0	14	46.7	26	43.3	
Females	18	60.0	16	53.3	34	56.7	
Total	30	100.0	30	100.0	60	100.0	
Chi-square test; P-value & sig.	2=0.27	1, P	>0.05,	NS			

NS= not significant, S=significant, HS=highly significant, VHS=very highly significant

Study reveals that, There was no statistical significance difference of sex among vitiligo patients and controls (P>0.05).

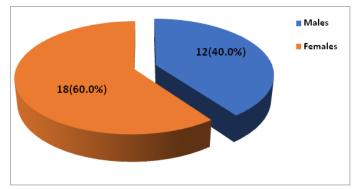


Figure 2: Pie diagram represents sex wise distribution of vitiligo patients

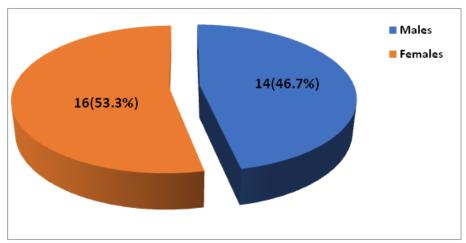


Figure 3: Pie diagram represents sex wise distribution of controls

Table 3: Comparison of serum calcium with vitiligo patients and controls

Age in years	Vitiligo patients	Controls	SE	CI	t-test value	
	Mean ± SD	Mean ± SD			P-value & Significance	
Serum Calcium	$9.38 \pm 0.23$	$9.87 \pm 0.25$	0.064	95%	t = 7.730,	
					P = 0.000, VHS	

NS= not significant, S=significant, HS=highly significant, VHS=very highly significant

Study reveals that, There was statistically very highly significance difference of serum calcium among vitiligo patients and controls (P < 0.001)

Mean serum calcium level was significantly low in vitiligo patients as compare to controls (Normal cases).

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Figure 4: Simple bar diagram represents Comparison of serum calcium with vitiligo patients and controls

Table 4: Comparison of age and sex within the vitiligo patients and controls

Age in years	Males	Females	SE	CI	t-test value
	Mean ± SD	Mean ± SD			P-value & Significance
Vitiligo patients	$38.72 \pm 15.67$	$29.27 \pm 9.96$	0.064	95%	t = 2.18,
					P = 0.048, S
Controls	$36.28 \pm 7.43$	$34.0 \pm 9.96$	3.29	95%	t = 0.404,
					P = 0.689, NS

NS= not significant, S=significant, HS=highly significant, VHS=very highly significant.

Study reveals that, There was statistical significance difference of age among males and females in vitiligo patients (P < 0.05)

Mean age of males was significantly higher in vitiligo patients as compare to females

There was no statistical significance difference of age among males and females in vitiligo patients (P>0.05).

Table 5: Types of vitiligo wise distribution of patients

Types of Vitiligo	No. of Cases	Percentage
Segmental vitiligo	2	6.7
Mixed vitiligo	5	16.7
Acrofacial vitiligo	6	20.0
Generalised vitiligo	17	56.6
Total	30	100.0

Study observed that, Maximum number of cases 17 (56.6%) had the generalized vitiligo. belongs to the age group of 21-40 years.

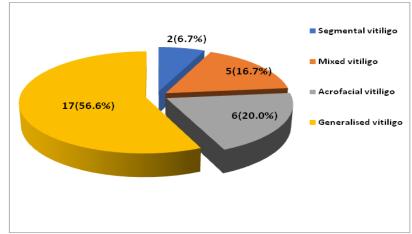


Figure 5: Pie diagram represents types of vitiligo wise distribution of patients

#### **Discussion**

Defective calcium transport has been shown in keratinocytes and melanocytes obtained from vitiliginous skin samples. Decreased intracellular calcium Leads to high levels of reduced thioredoxin, which inhibits Tyrosinase activity and results in the inhibition of melanin synthesis [9].

Low levels of calcium in turnindicate vitamin D3 deficiency in vitiligo cases leading to melanocyte loss by increasing the expression of cytokines such as IL-6, IL-8, TNF-alpha, TNF-gamma and also increase in the apoptotic activity in melanocytes [9].

Our study was in agreement with TAKCI et.al [10] showing low levels of both calcium and vitamin D

#### Conclusion

Adequate supplementation of Vutamjn D3 and calcium is required to prevent melanocyte loss in vitiligo ad stimulate melanin productionby proliferation and maturation of melanocytes and keratinocytes.

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