



ASSESSMENT OF SERUM VITAMIN D3, THYROID-STIMULATING HORMONE, AND CALCIUM LEVELS IN DIFFUSE HAIR LOSS AMONG PRE AND POST MENOPAUSAL FEMALES

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ABSTRACT

Introduction: Diffuse hair loss is a common concern among women and may be influenced by hormonal, nutritional, and metabolic factors. Vitamin D3, thyroid hormones, and calcium play critical roles in hair follicle cycling and growth.

Aims: To evaluate the serum levels of vitamin D3, thyroid-stimulating hormone (TSH), and calcium in pre- and post-menopausal females presenting with diffuse hair loss and assess their potential association with the severity of hair loss.

Materials and Methods: This cross-sectional observational study was conducted at the Department of Biochemistry, Index Medical College, Indore, from August 2022- October 2025. It included 200 pre- and post-menopausal women presenting with diffuse hair loss, excluding those with scarring alopecia, systemic illnesses, or medications affecting hair or mineral metabolism.

Results: In our study of 200 women with diffuse hair loss, post-menopausal women had a higher prevalence of vitamin D3 deficiency (82% vs. 65%, $p = 0.008$) and abnormal TSH levels (35% vs. 22%, $p = 0.03$) compared to pre-menopausal women. Sufficient vitamin D3 levels were less common in post-menopausal women (3% vs. 10%, $p = 0.04$), while low calcium levels showed no significant difference between the groups (12% vs. 8%, $p = 0.34$). Mean serum levels mirrored these findings, with lower vitamin D3 (14.30 ± 5.70 ng/mL vs. 18.50 ± 6.20 ng/mL), higher TSH (3.60 ± 1.40 μ IU/mL vs. 2.80 ± 1.10 μ IU/mL), and slightly lower calcium (8.90 ± 0.60 mg/dL vs. 9.10 ± 0.50 mg/dL) in post-menopausal women. **Conclusion:** Vitamin D3 deficiency and thyroid dysfunction are common among women with diffuse hair loss and may contribute to its pathogenesis. Routine screening for these biochemical parameters in pre- and post-menopausal females presenting with hair loss may aid in early diagnosis and targeted management.

Keywords: Diffuse hair loss, Vitamin D3, Thyroid-stimulating hormone, Calcium, Pre-menopausal, Post-menopausal

INTRODUCTION

Diffuse hair loss is a prevalent dermatological condition that affects a substantial proportion of women, with significant psychological and social implications. It often manifests as generalized thinning of scalp hair without scarring, and may be influenced by multiple factors including hormonal changes, nutritional deficiencies, metabolic disorders, and autoimmune conditions [1]. Among these, deficiencies of vitamin D3, thyroid dysfunction, and alterations in calcium homeostasis have been increasingly recognized as important contributors to the pathogenesis of diffuse hair loss [2, 3].

Vitamin D3 (cholecalciferol) plays a crucial role not only in calcium and bone metabolism but also in hair follicle biology. The vitamin D receptor (VDR) is expressed in keratinocytes and dermal papilla cells, which regulate hair follicle cycling and growth [4]. Studies have demonstrated that low serum vitamin D3 levels are commonly observed in women with diffuse hair loss, suggesting that deficiency may impair hair follicle proliferation and differentiation [5]. For example, a case-control study in South India reported that 81.8% of women with diffuse hair fall were vitamin D deficient, compared to only 45.5% of healthy controls, indicating a significant association between vitamin D status and hair shedding [6]. Other studies have confirmed these findings, emphasizing the potential role of vitamin D supplementation in the management of hair loss [7].

Thyroid hormones are critical regulators of metabolism and growth, and their dysregulation can significantly affect hair follicle function. Thyroid-stimulating hormone (TSH) indirectly influences hair follicle cycling through modulation of thyroid hormone levels, which regulate the anagen (growth) and telogen (resting) phases of the hair cycle [8]. Clinical evidence suggests that both hypothyroidism and hyperthyroidism may manifest with diffuse hair thinning, often presenting as telogen effluvium rather than patterned baldness [9]. Subclinical thyroid dysfunction, particularly in women, may remain undetected but still contribute to hair shedding, highlighting the importance of routine TSH evaluation in women presenting with diffuse hair loss [10].

Calcium is essential for cellular signaling and keratinocyte differentiation, and although direct evidence linking serum calcium levels to hair loss in humans is limited, experimental models have shown that disturbances in calcium metabolism can disrupt hair follicle cycling and result in alopecia. Calcium homeostasis is closely regulated by endocrine pathways, including parathyroid and thyroid hormones, which may influence hair growth indirectly. In combination with vitamin D3 deficiency, low calcium levels may exacerbate hair follicle dysfunction and contribute to diffuse hair shedding.

Menopause is a critical period associated with declining estrogen levels, which affects hair growth by shortening the anagen phase and increasing telogen hair shedding. Post-menopausal women are also at increased risk of vitamin D insufficiency and metabolic bone disease, factors that may further exacerbate hair loss. Nutritional deficiencies and endocrine abnormalities often coexist with advancing age and hormonal changes, making it essential to evaluate biochemical parameters systematically in both pre- and post-menopausal women presenting with hair loss.

Several observational studies reinforce the multifactorial nature of diffuse hair loss in women. A quantitative analysis of circulating biomarkers demonstrated that women with hair loss frequently exhibit lower levels of hemoglobin, iron, zinc, vitamin D3, calcium, and abnormal TSH values compared to healthy controls. Similarly, retrospective studies of telogen effluvium populations indicate that micronutrient deficiencies, including vitamin D3 and calcium, are prevalent and justify routine laboratory evaluation to guide management.

Despite increasing recognition of the role of vitamin D3 deficiency and thyroid dysfunction in diffuse hair loss, comprehensive assessment in relation to menopausal status remains limited. Evaluating serum vitamin D3, TSH, and calcium in pre- and post-menopausal females can provide insights into the interplay between nutritional status, hormonal changes, and hair follicle health, and may guide evidence-based diagnostic and therapeutic strategies.

The primary aim of this study is to evaluate the serum levels of vitamin D3, thyroid-stimulating hormone (TSH), and calcium in pre- and post-menopausal females presenting with diffuse hair loss,

and to determine their potential association with the severity of hair shedding. The objectives include assessing the prevalence of vitamin D3 deficiency, thyroid dysfunction, and calcium imbalance in this population, comparing these biochemical parameters between pre- and post-menopausal women, and identifying correlations between these markers and clinical patterns of diffuse hair loss, thereby facilitating evidence-based diagnostic and therapeutic approaches.

MATERIALS AND METHODS

This is a cross-sectional study conducted at Department of Biochemistry, Index Medical college Hospital & Research centre, Indore, M. P. The duration of study is from August 2022- October 2025 year. The total 200 numbers of subjects will be included having age group pre-menopausal females 18–50 years post-menopausal females 50 years and above.

Inclusion criteria:

- Pre- and post-menopausal females aged.
- Patients presenting with diffuse hair loss of any duration.
- Willingness to provide informed consent and participate in the study.

Exclusion Criteria:

- Females with scarring alopecia or patterned hair loss (androgenetic alopecia).
- History of systemic diseases affecting hair growth, such as chronic kidney disease, liver disease, or autoimmune disorders.
- Patients currently on medications that may influence hair growth or serum levels of vitamin D3, calcium, or thyroid hormones (e.g., corticosteroids, vitamin D/calcium supplements, thyroid medications).
- Pregnant or lactating women.
- Patients with malnutrition or other conditions leading to secondary hair loss unrelated to endocrine or micronutrient factors.

Parameters:

Vitamin D by Chemiluminescence immunoassay (CLIA) Method, Thyroid Stimulating hormone by Chemiluminescence immunoassay (CLIA) Method and Calcium by Colorimetric Method. Serum Vitamin D3, TSH, and Calcium Levels between Pre-menopausal and Post-menopausal Women

Statistical analysis :

Data from the study were analyzed using SPSS software, with continuous variables (e.g., age, liver enzyme levels) expressed as mean $\pm$ SD and compared using t-tests or Mann–Whitney U tests. Categorical variables (e.g., gender, CBD stones, and complications) were presented as frequencies and percentages, and compared using Chi-square or Fisher's exact tests. Diagnostic accuracy (sensitivity, specificity, PPV, NPV, and accuracy) was calculated for MRCP-first and EUS-first strategies, using ERCP/intraoperative findings as the reference. Kaplan-Meier analysis may be used for time-to-intervention comparisons. A p-value < 0.05 was considered significant.

RESULT

Table 1: Association of Vitamin D3 Status with Menopausal Status

Vitamin D3 Status	Pre-menopausal (n=100)	Post-menopausal (n=100)	p-value
Deficient (<20 ng/mL)	65 (65%)	82 (82%)	0.008
Insufficient (20–30 ng/mL)	25 (25%)	15 (15%)	0.07
Sufficient (>30 ng/mL)	10 (10%)	3 (3%)	0.04

Table 2: Association of TSH Status with Menopausal Status

TSH Status	Pre-menopausal (n=100)	Post-menopausal (n=100)	p-value
Normal (0.4–4.0 μ IU/mL)	78 (78%)	65 (65%)	0.03
Abnormal (<0.4 or >4.0 μ IU/mL)	22 (22%)	35 (35%)	0.03

Table 3: Association of Calcium Status with Menopausal Status

Calcium Status	Pre-menopausal (n=100)	Post-menopausal (n=100)	p-value
Normal (8.5–10.5 mg/dL)	92 (92%)	88 (88%)	0.34
Low (<8.5 mg/dL)	8 (8%)	12 (12%)	0.34

Table 4: Comparison of Serum Vitamin D3, TSH, and Calcium Levels between Pre-menopausal and Post-menopausal Women

Parameter	Group	Mean	SD	p-value
Vitamin D3 (ng/mL)	Pre-menopausal (n=100)	18.5	6.2	<0.001
	Post-menopausal (n=100)	14.3	5.7	
TSH (μ IU/mL)	Pre-menopausal (n=100)	2.8	1.1	0.002
	Post-menopausal (n=100)	3.6	1.4	
Calcium (mg/dL)	Pre-menopausal (n=100)	9.1	0.5	0.045
	Post-menopausal (n=100)	8.9	0.6	

Figure 1: Association of Vitamin D3 Status with Menopausal Status

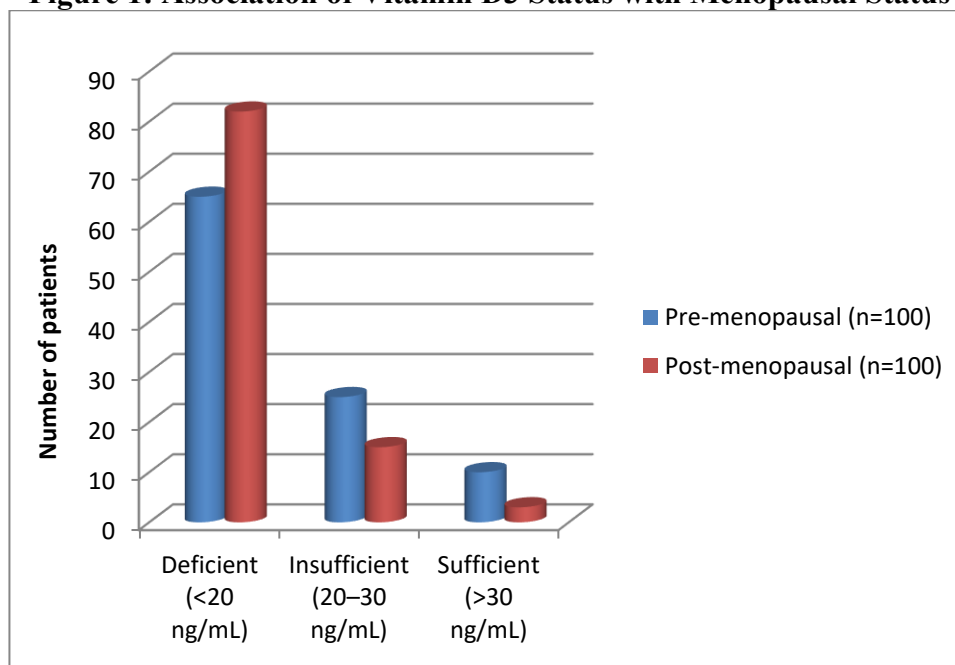
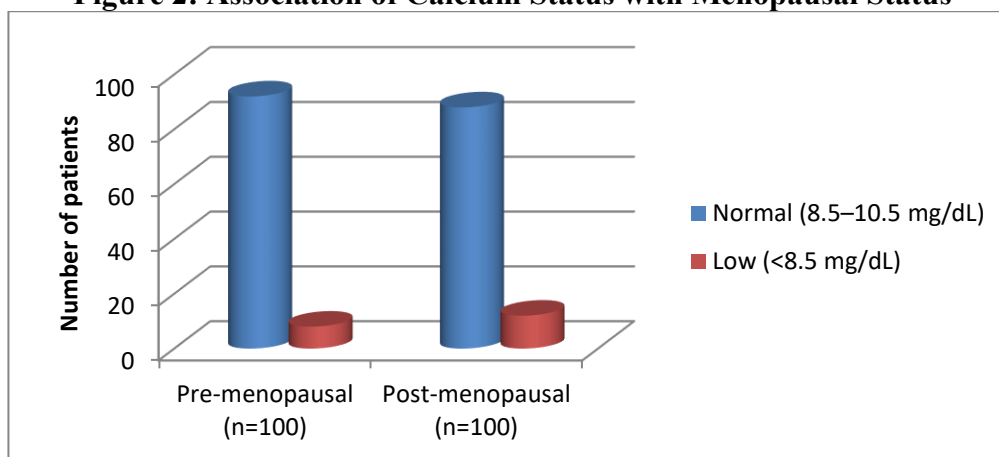


Figure 2: Association of Calcium Status with Menopausal Status



In this study of 200 women with diffuse hair loss, 65 pre-menopausal women out of 100 (65%) and 82 post-menopausal women out of 100 (82%) were found to have vitamin D3 deficiency (<20 ng/mL), and this difference was statistically significant ($p = 0.008$). Among those with insufficient vitamin D3 levels (20–30 ng/mL), 25 pre-menopausal women (25%) and 15 post-menopausal women (15%) were affected, which was not statistically significant ($p = 0.07$). In the sufficient category (>30 ng/mL), 10 pre-menopausal women (10%) and 3 post-menopausal women (3%) had adequate vitamin D3 levels, a difference that was statistically significant ($p = 0.04$).

In this study, 78 pre-menopausal women out of 100 (78%) and 65 post-menopausal women out of 100 (65%) had normal TSH levels (0.4–4.0 $\mu\text{IU/mL}$), and this difference was statistically significant ($p = 0.03$). Conversely, 22 pre-menopausal women (22%) and 35 post-menopausal women (35%) had abnormal TSH levels (<0.4 or >4.0 $\mu\text{IU/mL}$), which was also statistically significant ($p = 0.03$).

In this study, 92 pre-menopausal women out of 100 (92%) and 88 post-menopausal women out of 100 (88%) had normal serum calcium levels (8.5–10.5 mg/dL), while 8 pre-menopausal women (8%) and 12 post-menopausal women (12%) had low calcium levels (<8.5 mg/dL). The differences between pre- and post-menopausal groups were not statistically significant ($p = 0.34$).

In this study, the mean serum vitamin D3 level in pre-menopausal women ($n=100$) was 18.500 ± 6.200 ng/mL, whereas in post-menopausal women ($n=100$) it was 14.300 ± 5.700 ng/mL, and this difference was statistically significant ($p < 0.001$). The mean TSH level in pre-menopausal women was 2.800 ± 1.100 $\mu\text{IU/mL}$ compared to 3.600 ± 1.400 $\mu\text{IU/mL}$ in post-menopausal women, showing a significant difference between the groups ($p = 0.002$). For serum calcium, the mean level in pre-menopausal women was 9.100 ± 0.500 mg/dL, while post-menopausal women had a mean of 8.900 ± 0.600 mg/dL, which was also statistically significant ($p = 0.045$).

DISCUSSION

In the present study of 200 women with diffuse hair loss, we found that post-menopausal women had a significantly higher prevalence of vitamin D3 deficiency compared with pre-menopausal women (82% vs. 65%, $p = 0.008$), and mean serum vitamin D3 levels were significantly lower in post-menopausal women (14.300 ± 5.700 ng/mL) than in pre-menopausal women (18.500 ± 6.200 ng/mL, $p < 0.001$). This finding corresponds with the biological role of the vitamin D receptor (VDR) in hair follicle homeostasis, where VDR deficiency arrests hair cycling and results in alopecia in experimental models (Joko et al.) [11,12]. Clinical studies also support the link between low vitamin D and female hair loss: Sadoma et al. reported significantly decreased serum vitamin D levels in women with female pattern hair loss compared with controls [13], and Siddiqui et al. documented a high frequency of vitamin D3 deficiency among patients with diffuse female hair loss [14]. Nayak et al. further demonstrated significantly lower vitamin D3 levels in individuals with

diffuse hair fall than in healthy controls, reinforcing the association between vitamin D status and hair loss [15].

Beyond vitamin D, our study showed that abnormal thyroid-stimulating hormone (TSH) levels were significantly more common in post-menopausal women compared with pre-menopausal women (35% vs. 22%, $p = 0.03$), and mean TSH levels were higher in the post-menopausal group ($3.600 \pm 1.400 \mu\text{IU/mL}$ vs. $2.800 \pm 1.100 \mu\text{IU/mL}$, $p = 0.002$). Thyroid dysfunction is a well-recognized contributor to diffuse hair loss due to its effect on hair follicle growth and metabolism. Hussein et al. have reviewed that thyroid hormone imbalances—including both hypothyroidism and hyperthyroidism—are associated with telogen effluvium and other forms of hair shedding [16]. Popa et al. reviewed thyroid profiles in patients with various alopecia types and emphasized that thyroid hormone abnormalities may influence hair growth cycles [17]. Kebbab et al. found significant differences in TSH concentrations among women with diffuse hair loss compared with controls, supporting the notion that thyroid function tests are important in the evaluation of diffuse hair loss [18].

In respect to calcium, while the proportion of women with low serum calcium did not significantly differ between pre- and post-menopausal groups in our study (12% vs. 8%, $p = 0.34$), the mean calcium level was slightly lower in post-menopausal women ($8.900 \pm 0.600 \text{ mg/dL}$ vs. $9.100 \pm 0.500 \text{ mg/dL}$, $p = 0.045$). Although direct human clinical evidence linking calcium to hair loss is limited, the interplay between calcium homeostasis and VDR-mediated pathways supports the idea that calcium metabolism may indirectly influence hair follicle function. Moreover, VDR's role in keratinocyte differentiation and hair cycling further suggests a mechanistic link between vitamin D, calcium regulation, and hair follicle biology that merits additional investigation.

Our findings are also consistent with broader biomarker studies. Al-Fawaeir and Al-Odat showed that women with hair loss had significantly reduced levels of vitamin D and thyroid hormones compared with controls, suggesting a multifactorial biochemical profile in these patients [19]. Additionally, meta-analytic evidence indicates that vitamin D deficiency is often present across multiple alopecia types, including telogen effluvium and female pattern hair loss [20]. Collectively, these findings support the importance of assessing serum vitamin D3 and thyroid function in women with diffuse hair loss, particularly in post-menopausal populations.

CONCLUSION

The study demonstrates that diffuse hair loss in women is significantly associated with lower vitamin D3 levels and altered thyroid function, particularly in post-menopausal women. While calcium levels showed minor variations, they did not appear to play a major role. These findings highlight the importance of assessing vitamin D status and thyroid function in women presenting with diffuse hair loss, and suggest that correcting these deficiencies and hormonal imbalances may contribute to improved hair health.

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