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Correlation of Serum Vitamin B12, Vitamin D and Iron Levels with NEET Score and Academic Performance among First-Year BHMS Students - A Cross-Sectional Analytical Study

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1. Background and Rationale

Micronutrients such as Vitamin B12, Vitamin D and Iron play a crucial role in neurocognitive development, neurotransmitter synthesis, my elination, oxygen transport and mood regulation. Deficiencies of these nutrients are highly prevalent among Indian adolescents and young adults, including medical students, due to dietary habits, limited sun exposure, stress and lifestyle factors.

Medical entrance examinations like NEET assess sustained cognitive ability, concentration, memory and stress tolerance, while university examination performance reflects ongoing learning capacity and academic efficiency. Despite their importance, limited Indian data exist correlating objective biochemical micronutrient status with objective academic performance indicators among medical students.

Understanding this relationship may help in

- early identification of at-risk students,
- formulation of preventive nutritional strategies, and
- Improvement of academic and mental well-being.

2. Review of Literature

The human brain, despite constituting approximately two percent of total body mass, accounts for nearly twenty percent of daily energy expenditure and is uniquely dependent on a continuous supply of specific micronutrients for its structural integrity and functional capacity [1,2]. Micronutrients serve as cofactors for enzymatic reactions, regulators of gene expression, mediators of neurotransmitter synthesis, and modulators of myelin formation — processes that collectively govern cognitive performance across the lifespan. Benton established that brain development continues well beyond the early postnatal period into adulthood, and that micronutrient status can influence neurological functioning at every stage, with deficiencies having long-term implications for cognitive development [1]. Bryan et al. further demonstrated that multiple nutrients — particularly iron, zinc, iodine, and B-vitamins — are critical for cognitive development in school-aged children and young adults, with simultaneous deficiencies producing compounding impairments in attention, processing speed and executive function [2]. Gómez-Pinilla, in a landmark review in Nature Reviews Neuroscience, highlighted the mechanisms by which dietary nutrients affect brain plasticity and cognitive function, emphasizing that the brain is especially vulnerable to micronutrient insufficiencies because its high metabolic demand renders it sensitive to even minor disruptions in the supply of essential cofactors [3]. Critically, subclinical deficiencies — those existing in the absence of overt clinical signs — can significantly impair attention, memory, executive function, fatigue tolerance and academic performance, representing a silent but pervasive burden among young adults in both developing and industrialized settings [3].

Vitamin B12 (cobalamin) is a water-soluble, cobalt-containing vitamin that functions as an indispensable cofactor in two key metabolic reactions: the conversion of methylmalonyl-CoA to succinyl-CoA, essential for neuronal energy metabolism, and the remethylation of homocysteine to methionine, which is required for one-carbon methylation reactions including DNA synthesis and epigenetic regulation [4]. Through these pathways, Vitamin B12 plays a vital role in myelin sheath formation, neuronal structural integrity, and the maintenance of neurotransmitter equilibrium. Smith and Refsum conducted a comprehensive review demonstrating that moderately elevated plasma total homocysteine — a direct consequence of Vitamin B12 deficiency — is a strong, independently modifiable risk factor for cognitive decline, white matter damage, brain atrophy, and dementia [4]. The neurotoxic effects of homocysteine include promotion of oxidative stress, induction of neuronal apoptosis, and impairment of N-methyl-D-aspartate (NMDA) receptor function, all of which disrupt the synaptic plasticity mechanisms essential for learning and memory consolidation [4].

Allen described the clinical spectrum of Vitamin B12 deficiency, noting that neuropsychiatric manifestations — including impaired concentration, memory deficits, fatigue, and mood disturbances — frequently precede the development of hematological abnormalities such as megaloblastic anemia [5]. This dissociation between neurological and hematological presentation means that reliance on anemia as a diagnostic threshold substantially underestimates the true burden of functionally significant deficiency. O'Leary and Samman, in a comprehensive review of the metabolism and health consequences of Vitamin B12, confirmed that sub-clinical deficiency is common and its long-term consequences encompass vascular disease, impaired cognitive health, adverse pregnancy outcomes, and reduced bone mineral density, with particular vulnerability observed in individuals adhering to plant-based dietary patterns [6]. Refsum and colleagues, investigating a population from Maharashtra, India, documented elevated hyperhomocysteinemia and methylmalonic acid levels as biochemical indicators of widespread cobalamin deficiency among Asian Indians, attributing this in part to the high prevalence of vegetarian dietary practices that restrict intake of the primary dietary sources of Vitamin B12, namely animal-derived foods [7]. Pawlak et al. conducted a systematic review of the literature and reported Vitamin B12 deficiency rates ranging from 11% to 90% among various vegetarian populations globally, with the highest rates observed in those following strictly plant-based diets since birth, findings of direct relevance to the predominantly vegetarian dietary culture of Indian medical students [8].

Vitamin D3 (cholecalciferol) operates not merely as a classical nutrient but as a neurosteroid hormone, with vitamin D receptors (VDR) and the key biosynthetic enzyme 1-alpha hydroxylase distributed throughout the central nervous system — including the hippocampus, prefrontal cortex, cerebellum, and limbic system [9]. Garcion and colleagues were among the first to comprehensively characterize the neurobiological roles of 1,25-dihydroxyvitamin D3 in the nervous system, demonstrating its involvement in regulation of nerve growth factor synthesis, modulation of neuroinflammatory pathways, induction of antioxidant defenses, and control of calcium-mediated neuronal signaling — mechanisms that collectively support neurodevelopment, neuroprotection, and synaptic plasticity [9]. The anatomical distribution of VDR expression in the hippocampus and prefrontal cortex — regions critically implicated in memory encoding, executive decision-making, and mood regulation — provides a neurobiological basis for the cognitive and neuropsychiatric consequences of Vitamin D deficiency that have been observed epidemiologically [9]. Holick, in a seminal review in the New England Journal of Medicine, brought international attention to the global prevalence of Vitamin D deficiency and its wide-ranging clinical consequences, which extend beyond classical musculoskeletal disease to encompass muscle weakness, chronic fatigue, impaired immune function, and neuropsychiatric manifestations including depression and cognitive difficulties [10]. Eyles, Burne, and McGrath subsequently provided detailed evidence of Vitamin D's essential roles in brain development, adult brain function, and the pathophysiological links between low Vitamin D levels and neuropsychiatric disorders, including schizophrenia, depression, and Alzheimer's disease, with preclinical data demonstrating that developmental Vitamin D deficiency disrupts dopamine ontogeny, axonal connectivity, and brain structure [11].

India, despite abundant sunshine, paradoxically reports some of the highest rates of Vitamin D deficiency in the world, with prevalence estimates ranging from 70% to 100% in the general population, attributable to socioreligious and cultural practices that restrict sunlight exposure, the high melanin content of darker skin that reduces dermal synthesis, widespread consumption of foods low in Vitamin D, and the absence of mandatory food fortification policies [10,11]. Among Indian medical students specifically, Gupta and colleagues documented a high prevalence of Vitamin D deficiency, highlighting particular vulnerability in this population due to predominantly indoor academic schedules, limited outdoor activity, and dietary habits that inadequately compensate for reduced cutaneous synthesis [12]. Kancherla and colleagues further reported significant associations between Vitamin D deficiency and heightened psychological stress in young adults, noting that low Vitamin D status was linked to increased anxiety, reduced stress tolerance, poor sleep quality, and diminished academic engagement — outcomes particularly consequential for students navigating the high-pressure environment of medical education [13]. These findings collectively suggest that widespread Vitamin D insufficiency among Indian medical undergraduates may constitute a modifiable biological determinant of cognitive performance and academic outcomes.

Iron is an essential trace element that is indispensable for multiple neurobiological processes, including cerebral oxygen delivery via hemoglobin and myoglobin, mitochondrial energy metabolism through its role in the electron transport chain, myelination of axons by oligodendrocytes, and the biosynthesis of catecholamine neurotransmitters — specifically dopamine, norepinephrine, and serotonin — through its function as a cofactor for tyrosine hydroxylase and tryptophan hydroxylase [14]. Lozoff and Georgieff provided a comprehensive review of the neurodevelopmental consequences of iron deficiency, demonstrating that even in the absence of frank anemia, iron deficiency impairs attention, working memory, speed of information processing, and long-term learning capacity, with effects mediated through disruption of dopaminergic neurotransmission, impaired myelination, and altered hippocampal neurochemistry [14]. These neurocognitive deficits are particularly pertinent for young adults engaged in intensive academic learning, where sustained attention and efficient memory consolidation are prerequisites for academic success. Beard demonstrated that iron deficiency alters brain energy metabolism and neurotransmitter function through mechanisms distinct from anemia, producing behavioral changes and cognitive performance decrements that can persist even following hematological correction, highlighting the importance of assessing iron status biochemically rather than relying solely on hemoglobin measurements [15]. The World Health Organization recognizes iron deficiency as the most prevalent nutritional disorder globally, disproportionately affecting women of reproductive age, adolescents, and young adults — groups that include the majority of undergraduate medical students [16]. McCann and Ames conducted a systematic review of causal evidence linking iron deficiency during development with deficits in cognitive and behavioral function, concluding that the evidence for causality is substantially supported by converging biological plausibility, epidemiological consistency, dose-response relationships, and reversibility in some domains following supplementation [17].

Medical students represent a distinctly vulnerable subpopulation for micronutrient deficiency, owing to a constellation of risk factors that are uniquely concentrated in the medical education environment. These include irregular and nutritionally inadequate dietary habits driven by time constraints and academic pressure, chronic psychological stress, sleep deprivation, severely restricted outdoor activity and sunlight exposure, and in the Indian setting, the additional risk conferred by predominantly vegetarian dietary practices [7,8,12]. Harinarayan and colleagues documented a high prevalence of low dietary calcium and Vitamin D deficiency in healthy South Indians, attributing this to traditional dietary patterns high in phytates and low in Vitamin D-rich foods, combined with limited sunlight exposure — factors directly applicable to the medical student population in Maharashtra [18]. Dyrbye, Thomas, and Shanafelt, in a landmark review of medical student distress, established that the medical training environment is inherently stressful, with studies documenting a progressive worsening of mental health following medical school enrollment, mediated through academic overload, exposure to patient suffering, sleep deprivation, and loss of personal time — conditions that are known to exacerbate nutritional depletion through increased metabolic demand, impaired dietary behavior, and disruption of absorptive function [19]. The cumulative biological and psychosocial burden experienced by medical students creates a setting in which micronutrient deficiencies are not only prevalent but also clinically consequential, with reported associations with physical fatigue, reduced cognitive performance, and impaired academic functioning [19].

The National Eligibility cum Entrance Test (NEET) is a nationally standardized, high-stakes competitive examination that assesses a comprehensive range of cognitive domains including sustained attention, rapid information processing, multi-step problem-solving, factual memory retrieval, and stress tolerance under timed conditions. University internal examination performance additionally reflects ongoing learning efficiency, memory consolidation, academic consistency, and the ability to apply knowledge over extended periods. The use of these objective, externally validated academic metrics as outcome variables in research designs minimizes the subjective reporting bias inherent in self-assessed academic performance measures and provides standardized, comparable indicators of cognitive function across study populations [20]. Kulkarni and Nimbalkar demonstrated that academic performance in medical entrance examinations is influenced by multiple cognitive predictors, and emphasized the value of incorporating objective biological variables when investigating determinants of academic performance in this population [20]. Despite this, limited research has systematically examined the biological and nutritional determinants of NEET performance among Indian medical students using directly measured biochemical markers. The existing literature, while establishing associations between micronutrient deficiencies and cognitive outcomes, has predominantly relied on dietary recall instruments rather than serum biochemical assays for exposure measurement, and on self-reported or subjective academic performance measures rather than standardized examination scores as outcome variables — methodological limitations that reduce the precision and generalizability of findings. Furthermore, the homoeopathic medical student population represents an entirely unstudied group whose nutritional profile, dietary practices, and academic stressors may differ meaningfully from those of students in conventional allopathic programs, warranting targeted investigation. The present study addresses this gap by correlating directly measured serum levels of Vitamin B12, Vitamin D and Iron with NEET score among first-year BHMS students. The identification of significant associations would provide a biochemically grounded basis for the implementation of routine nutritional screening protocols and targeted supplementation strategies within homoeopathic medical institutions, with the potential to improve both student health and academic outcomes.

3. Aim of the Study

To study the correlation between serum Vitamin B12, Vitamin D and Iron levels and NEET score as well as university academic performance among BHMS students.

4. Objectives

Primary Objectives

- To estimate serum levels of Vitamin B12, Vitamin D and Iron among BHMS students.
- To correlate serum Vitamin B12, Vitamin D and Iron levels with NEET Score/percentile.
- To correlate serum Vitamin B12, Vitamin D and Iron levels with first-year academic performance.

Secondary Objectives

- To compare academic performance between students with deficient vs sufficient micronutrient levels.
- To assess the influence of gender, diet pattern and BMl on micronutrient status and academic performance.

5. Research Question

- Is there significant association between serum Vitamin B12, Vitamin D and Iron levels and NEET Score / university academic performance in first-year BHMS students?

6. Hypothesis

Null hypothesis (H): There is no significant correlation between serum Vitamin B12, Vitamin D and Iron levels and NEET score or academic performance.
Alternate hypothesis (H,): Lower serum levels of Vitamin B12, Vitamin D and Iron are significantly associated with lower NEET Score and poorer academic performance.

7. Study Design

Type of study: Cross-sectional analytical study
Study setting: Private Homoeopathic Medical College
Study duration: 10 days

8. Study Population

BHMS students admitted through NEET. Sample Size
Total sample: 350 students (boys and girls)
(Sample size adequate for correlation and regression analysis.)

9. Inclusion Criteria

BHMS students

- Age 17-22 years
- Both genders
- Willing to give informed consent

10. Exclusion Criteria

- Known chronic illness (CKD, liver disease, malabsorption)
- Diagnosed psychiatric or neurological disorders Current vitamin/mineral supplementation (last 3 months)
- Known anemia under treatment

11. Study Variables

Independent Variables

- Serum Vitamin B12 (pg/mL) Serum Vitamin D [25(OH)D] (ng/mL)
- Serum Iron / Ferritin / Hemoglobin (as available) Dependent Variables
- NEET Score / percentile
- First-year internal assessment marks /university marks Confounding Variables
- Gender

BMI

- Diet pattern (vegetarian / mixed)
- Physical activity
- Sleep duration

12. Data Collection Method

Step 1: Baseline Data

- Demographic details
- Diet pattern
- Height, weight (BMI)

Step 2: Biochemical Assessment

Blood samples collected under aseptic precautions

- Analysis done in NABL-accredited laboratory

- Cut-off values (WHO/ICMR):

Vitamin B12: Deficient < 200 pg/mL
Vitamin D:
Insufficient 200-300 pg/mL
Normal > 300 pg/mL
Deficient< 20 ng/mL
Insufficient 20-30 ng/mL
Sufficient > 30 ng/mL
Iron parameters as per standard reference ranges

Step 3: Academic Performance Data

- NEET Score/percentile (verified from records) University marks

13. Statistical Analysis Plan

- Data entry using MS Excel
- Analysis using SPSs (version XX)
- Statistical Tests
- Descriptive statistics (mean, SD, percentage)
- Pearson/Spearman correlation between micronutrient levels and
- NEET/academic scores
- Independent t-test / ANOVA for group comparisons
- Multiple linear regression to identify independent predictors of academic performance
- Significance Level
- $p < 0.05$ considered statistically significant

14. Ethical Considerations

- Institutional Ethics Committee approval will be obtained
- Written informed consent from all participants
- Confidentiality and anonymity maintained
- Participants with deficiencies will be advised appropriate medical consultation

15. Expected Outcomes

High prevalence of Vitamin B12, Vitamin D and Iron deficiencies

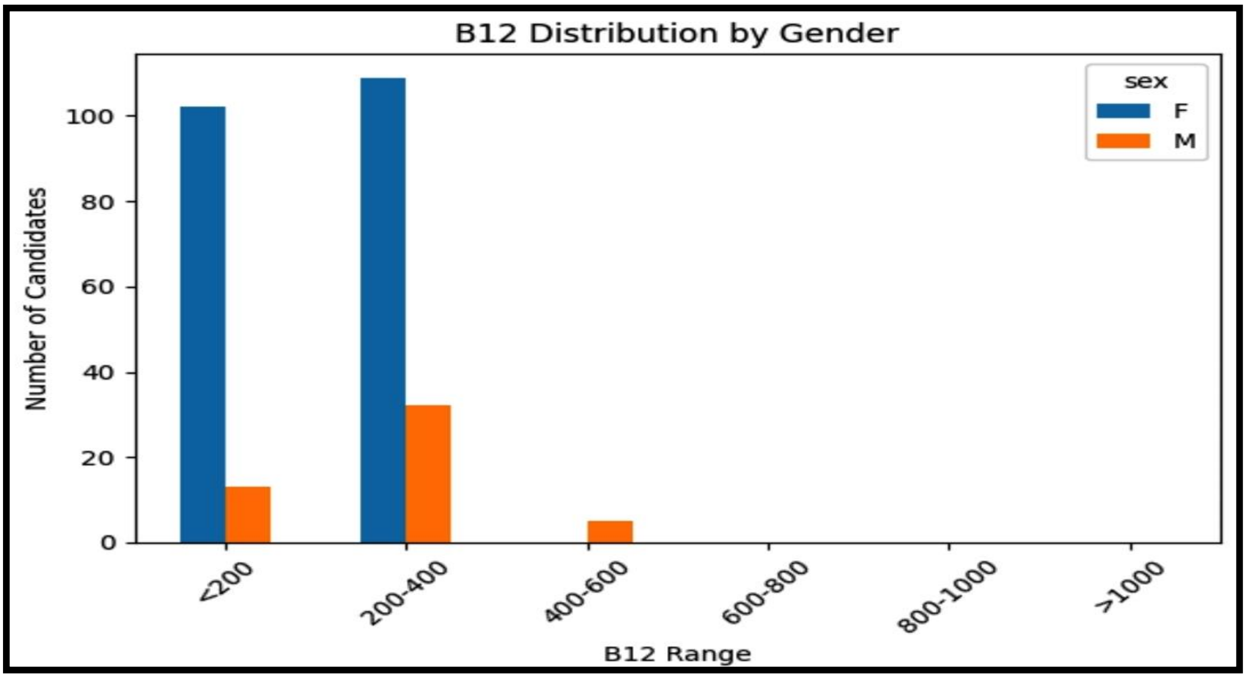
- Students with deficiencies likely to have:
- Lower NEET SCores
- Poorer academic performance
- Evidence supporting nutritional screening at entry level

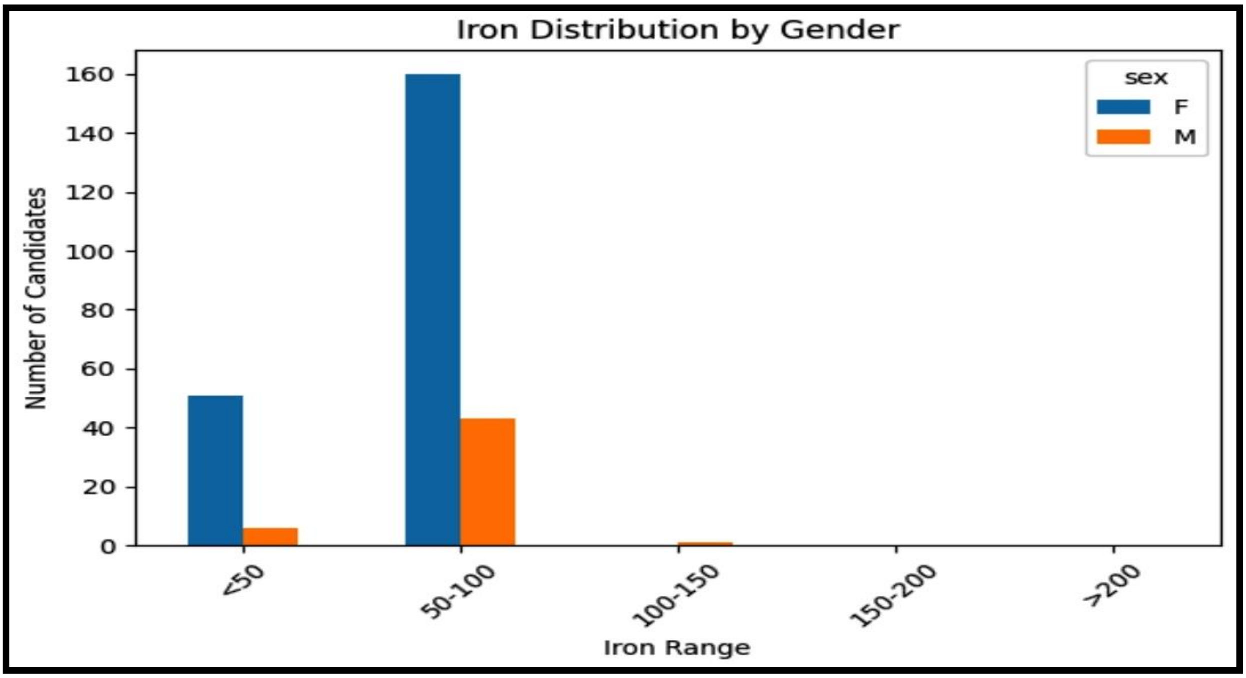
16. Limitations of the Study

- Cross-sectional design cannot establish causality
- Single-center study
- Academic performance influenced by multiple non-nutritional factors

17. Significance of the Study

- Early identification of nutritional risk in medical students Basis for preventive supplementation programs
- Improvement in academic and mental well-being
- Policy implications for medical education institutes





18. Result- **A.** For correlation of serum iron level and NEET score
Chi square value-14**.** 14
P value------------ 0.00085

Statistically significant association between serum iron status and NEET score categories.

B. For correlation of serum Vit D level and NEET score
Chi Square Value – 13**.** 88
P value ---------------0**.** 0077

Statistically significant association between serum Vit D status and NEET score categories.

C. For correlation of serum Vit B12 level and NEET score
Chi square value- 17**.** 36
P value--------------0**.** 00164

Statistically significant association between serum Vit B12 status and NEET score categories.

Interpretation-

There was a statistically significant association between vit B12 status, Vit D status, and serum iron status and NEET score categories, indicating that micro nutrients status is related to academic performance among study participants**.**

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