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Article

Assessment of Serum Iron and Ferritin Status Among Day and Night Shift Workers: A Cross Sectional Study

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Abstract: Shift work is defined as a work pattern where workers perform tasks outside regular working hours. It has become an increasingly common phenomenon in various industries, many of which have been linked to burnout and identified as possibly detrimental to heart health. This study was conducted to explore the potential influence of the shift work on serum iron and ferritin**.** The current study included (80) participants consisting of (40) healthy day shift workers. In addition, (40) were night shift workers, spin react spin 120 system, completely automated biochemical analyzer (Germany) was used to determine the serum iron concentrations which assayed by using a commercial kit (BIO Research, Germany). Ferritin concentrations kits provided from (Abbott laboratories, USA) measured by automated Abbott Architect Ci4100 analyzer. The results of the current study, showed that the mean of serum iron levels were significantly lower ($p \leq 0.05$) in night shift workers (51.72±27.71 µg/dL) than day shift workers (74.17±15.50 µg/dL), while the ferritin levels didn't show significant differences between day and night shift workers. Depending on the years of work. The serum iron exhibited a significant decrease ($p \leq 0.05$) among workers with more than 5 years of exposure (42.56±23.01 µg/dL) compared to those with less than 5 years (80.82±22.38 µg/dL), while serum ferritin didn't show significant differences. The night shift work was associated with reduced serum iron concentrations compared with day work. Although ferritin concentrations tended to be higher with prolonged night work no statistically significant differences were observed.

Keywords: Day Shift Work, Ferritin, Iron, Night Shift Work

Introduction

Shift work has become an integral element of modern industrial society, which is stipulated by the nature of the work performed in vital spheres of activity, including medicine, security, and industry. Nonetheless, this working structure has significant physiological and health burdens because it disrupts the natural circadian rhythm of the human body, and the effects of such disruption are multidimensional [1]. Epidemiological and clinical data proves that shift workers, especially evening and night shift workers, are at high risk of cardiovascular diseases, metabolic syndrome, and sleep disorders as compared to those working regular morning shifts [2]. These increased health risks are mostly caused by the disturbance between the inner biological clock and external environmental signals that negatively influence the release of such important hormones as melatonin and cortisol and, in turn affect the significant changes in metabolic and immune functions [3].

The changes in iron metabolism may also be among those physiological alterations that can be influenced heavily when circadian rhythm is disrupted due to its great importance in several biological functions such as oxygen transport, energy generation, and DNA synthesis and the protein ferritin is the main iron storing protein found in the body and can thus be used clinically to measure iron storage [4]. The research that has been carried out has validated the existence of a complicated and reciprocal interaction between the circadian clock and the iron homeostasis gene system and has proven that the levels of serum iron and transferrin saturation acquire a strong diurnal fluctuation that reaches its highest point at certain times of the day [5]. Although, when working in a normal physiological environment, the ferritin concentrations might not demonstrate any apparent daily variations, evening shifts employees, such as sleep deprivation, and alterations in the timing of eating, might indirectly influence iron metabolism and absorption [6].

The study of the impact of iron and ferritin presence in the body of shift workers is crucially relevant with regard to the complicated function of the hepatic hormone hepcidin in the regulation of iron metabolism in the body. The unique characteristic of hepcidin is associated with the existence of specific biorhythms of hepcidin concentration in the organism, which increases from the morning hours till the end of the night, leading to decreased iron absorption during these periods [7]. Therefore, people performing evening shifts and who need to change the times of meal intake will face problems with the efficient assimilation of the iron content in food items, and thus, become more susceptible to the development of iron deficiency anemia [8]. Furthermore, current studies have proven that sleep disorders associated with evening shifts may lead to increased inflammatory mediators and high levels of ferritin as an acute phase reactant [9]. This study aims to conducted to explore the potential influence of the shift work on serum iron and ferritin**.**

Methodology

Study Design

The current study included (80) of these male, participants classified into two groups based on their work schedule into (40) day shift workers their mean age (35.12±10.38) and (40) night shift workers their mean age (34.44±10.45) years. The night shift workers group was classified according to their years of work into two subgroups; those group1 (n=20) worked for fewer than five years and group 2 (n=20) those working for more than five years. This cross sectional study was conducted between October 2024 to April 2025. Information on age, gender, amount of sleep per day, experience with shift work, length of shift work, and weekly working hours was collected, workers must been in their jobs for at least one year. Subjects must be worked the same pattern of shifts (either day or night) for at least six months.

Exclusion Criteria

The participants were not allowed to participate in this study if they had any of the following problems: any history of anemia or any other problems associated with their iron metabolism. Those taking iron supplement, multivitamin containing iron, or drugs that can influence their iron status, suffer from any kind of chronic disease like kidney diseases, liver diseases, or cancer, and smoking. The patients having acute infections at the time of drawing blood samples from their body. All the subjects gave informed consent in written form before participation.

Samples Collection

The collection of blood samples was done by venipuncture using (80) subjects. Five ml of blood sample was collected using a syringe. This sample was put into a sterilized test tube and left to coagulate. The serum was then extracted after centrifuging the sample at 4000 rpm in 15 minutes. The serum sample obtained was kept at -20 ºC and used for the estimation of serum ferritin and serum iron concentration.

Measurements of Serum Iron and Ferritin Concentrations

Spin react spin 120 system, completely automated biochemical analyzer (Germany) was used to determine the serum iron concentrations which assayed by using a commercial kit (BIO Research, Germany). Ferritin concentrations kits provided from (Abbott laboratories, USA) measured by automated Abbott Architect Ci4100 analyzer.

Statistical Analysis

The descriptive statistics like mean, standard deviation were performed on all continuous variables. The categorical variables were represented by frequency and percentage by chi-square test. The normality test showed that ferritin concentrations were not normally distributed (Shapiro-Wilk test). Therefore, non–parametric analysis (Mann-Whitney U-test) was applied, while iron concentrations were normally distributed Comparisons between the day shift worker group and the combined night shift worker group were performed using independent samples t-tests. A two-tailed p-value of less than 0.05 was considered statistically significant. All statistical analyses were performed using SPSS version 25.0.

Results

The findings in (**Table 1**) exhibited no significant differences between night and day shift workers as well as based on mean of age. Moreover, all subjects in both group are males so there no significant differences. Regarding to sleep duration night shift workers (6.42±0.97 hours/day) were lower significantly ($p \leq 0.05$) than day shift workers (7.25±1.86 hours/day) while regarding mean of meal day shift workers (2.69±0.60) significantly ($p \leq 0.05$) lower than night shift workers (3.11±0.89).

Table 1. Demographic and life style characteristic between day and night shift workers

Variables	Day shift workers (n=40)	Night shift workers (n= 40)	*p*-value
Age (years) (mean±SD)	35.12 ± 10.38	34.44 ± 10.45	0.83
Gender (male %)	40 (100%)	40(100%)	
Sleep duration (hours/days) (mean±SD)	7.25 ± 1.86	6.42 ± 0.97	0.01*
Mean numbers of meals/day	2.69 ± 0.60	3.11 ± 0.89	0.02*

n= number of sample, *= Significant difference at ($p \leq 0.05$) , SD= standard deviation

The comparison of serum ferritin concentrations between day shift workers and night shift workers expressed as median (IQR) due to non-normal distribution using the Mann-Whitney U-test showed no statistical significant differences ferritin data as in (**Table 2**).

Table 2. Measurement of serum ferritin ng/ml concentrations between night shift workers and day shift workers

Parameters	Groups	N	Median (IQR: 25-75%)	*p*-value
Ferritin (ng/ml)	Night shift workers	40	72.79 (36.84-95.40)	0.789
	Day shift workers	40	65.96 (33.84- 103.18)	

n= number of sample, SD= standard deviation, IQR =Interquartile Range

The results of the current study in (**Table 3**), showed the mean of serum iron levels were significantly lower ($p \leq 0.05$) in night shift workers (51.72±27.71 µg/dL) than day shift workers (74.17±15.50 µg/dL).

Table 3. Serum iron concentrations µg/dL between night shift workers and day shift workers

Parameters	Groups	N	Mean ± SD	*p*-value
Iron (µg/dL)	Night shift workers	40	51.72 ± 27.71	0.01*
	Day shift workers	40	74.17 ± 15.50	

n= number of sample, *= Significant difference at ($p \leq 0.05$), SD= standard deviation

Depending on the years of work, the serum iron exhibited a significant decrease ($p \leq 0.05$) in serum iron levels among workers with more than 5 years of exposure (42.56±23.01 µg/dL) compared to those with less than 5 years (80.82±22.38 µg/dL) as shown in (**Table 4**), while the ferritin levels didn't show significant differences between groups as (**Table 5**).

Table 4 . The concentrations of serum ferritin (ng/ml) among the night shift workers depending on duration of work

Parameters	Groups	n	Median (IQR:25-75%)	*p*-value
Ferritin ng/ml	Less than 5 years	20	41.60 (32.28-58.26)	0.09
	More than 5 years	20	80.07 (73.51-94.98)	

n= number of sample, SD= standard deviation, IQR =Interquartile Range

Table 5. The concentrations of serum iron µg/dL among the night shift workers depending on duration of work

Parameters	Groups	n	Mean ± SD	*p*-value
Iron (µg/dL)	Less than 5 years	20	80.82 ± 22.38	**0.001***
	More than 5 years	20	42.56 ± 23.01	

*= Significant difference at ($p \leq 0.05$), n= number of sample, SD= standard deviation, IQR=Interquartile Range

Discussion

The demographic features of the two groups were similar. There was no significant difference between day and night shift workers so that the differences in biochemical markers seen later in this study are unlikely to be due to age-related physiological changes. The cross-sectional study was conducted on men only, and sex was chosen as the biological confounder as menstrual blood loss, hormonal status, and pregnancy status have a significant effect on iron status and ferritin in women [11]. This enhances within-study comparisons due to the lack of female shift workers. The average sleep duration in night shift workers was significantly less than ($p \leq 0.05$) the day shift workers. The difference in numbers is under an hour but the biological implications should not be ignored. The American academy of sleep medicine recommends that adults sleep no less than 7 hours, and sleep< 7 hours on a regular basis is correlated with elevated levels of systemic inflammation, cortisol hyperactivity, and the inability to recover from a metabolic challenge [12]. In various large-scale studies, night shift workers consistently report lower sleep efficiency and shorter sleep duration compared to their endogenous circadian clock, typically while sleeping during the day and being exposed to social noise and light during night time [13,14].

Night shift workers had a significantly higher ($p \leq 0.05$) mean meal frequency compared to the others, which is consistent with occupational cohorts previously reported. Shift workers especially nighttime workers are likely to eat during biologically inappropriate timeframes such as late evening and early morning when the circadian-regulated metabolic processes are at their lowest point [15]. It is not wise to read too much into the idea of eating more meals. Those meals are crucial due to their timing, quality, and composition; indeed, there is evidence that shift workers' food selections are toward energy-dense, micronutrient-poor foods, when performing their tasks at times of fatigue and limited food availability [16]. Hepcidin is the peptide hormone most involved in iron homeostasis, by binding and degrading the only known iron export protein ferroportin, which in turn regulates iron absorption from the duodenum and iron recycling from macrophages [17]. Hepcidin levels are known to have a diurnal pattern with a rise throughout the morning hours and a peak in the late afternoon and evening, a pattern that helps to regulate iron uptake during the "biological night" and early morning hours [18,19]. Consuming meals at the usual time of day, and working at the same time, this is a physiologically coordinated pattern, which allows his meal time to coincide with the low levels of hepcidin when iron can be absorbed. However, the shift to night-time feeding times in night shift workers results in a mismatch between the time of the main meal and the time when endogenous hepcidin levels are highest, leading to impaired iron absorption in a sustained manner [19].

The issue of sleep deprivation has two ways of getting worse. Chronic sleep loss first stimulates the hypothalamic-pituitary-adrenal (HPA) axis leading to increased cortisol, which in turn leads to decreased erythropoiesis and decreased signaling of iron requirement [20]. Second, low level chronic inflammation and elevation of circulating levels of interleukins, particularly IL-6, have been associated with restriction of sleep which also strongly induces hepcidin transcription via the JAK/STAT3 signaling pathway [21]. This causes to have a high hepcidin level, which also does not reflect the iron stores, thus inhibiting iron absorption even if consuming iron in his diet. This leads to a progressive decrease in the iron pool in circulation and the iron present in serum in this study appears to conform to this.

The results reported are similar to previous studies reporting changes in hematological parameters due to shift work. Sooriyaarachchi *et al*., investigated several hematological parameters in health care shift workers and identified that night shift workers had abnormalities in several red blood cell (RBC) related parameters, but in some cases, the main parameter of serum iron was not the primary focus [22]. Camaschella *et al.*, have reported diurnal variation in iron status indices such as serum iron and transferrin saturation, with a variation of up to 30% across a day, reflecting the importance of methodological considerations for research in this area [23]. A dose-response pattern was found with the analysis by duration of night shift exposure. The serum iron of workers with more than 5 years of night shift exposure was significantly lower ($p \leq 0.05$) than the workers with less than 5 years of night shift exposure. The gradient is undoubtedly the most interesting result from this study. It suggests that the physiological consequences of circadian misalignment do not reach a steady state but increase over time. The adverse effect of chronic exposure to night work on the iron status seems to be time-dependent, which has potentially important implications for workers who have long-term rotational or fixed-night work periods. This is in line with the wider shift work literature which has shown that the cardiometabolic and endocrine effects of night work increase with duration of night work [13,24].

There were no significant differences in the serum ferritin level when compared between day and night shift workers and between the night shift workers by duration of exposure. Such dissociation of serum iron and ferritin is not physiologically insignificant, and should not be ignored.

Serum ferritin indicates iron stores which are iron bound in the hepatocytes and reticuloendothelial macrophages for the most part. It is not very responsive to changes in iron status and operates on a weeks to months' time scale, which is different from serum iron, which reflects the circulating, transport available iron pool, and is highly variable on shorter time scales (less than a week) [19]. When comparing ferritin levels between the duration subgroups, a similar pattern is seen, with workers that had more than 5 years of night shift exposure having significantly higher ferritin than those with less than 5 years. At first glance this seems counterintuitive, how can you maintain or even increase the levels of iron stores when the levels of circulating iron are already low? The reason for this, almost surely is the dual nature of ferritin. Ferritin is classically thought to be the indicator of iron storage, but it is a well-established acute phase reactant, increasing in response to inflammation, infection, liver disease and oxidative stress, irrespective of iron stores [19,21].

Ongoing sleep restriction and circadian disruption are linked to persistently increased levels of pro-inflammatory cytokines, and long-term night shift workers could have a subclinical increased inflammatory tone, which would result in a higher ferritin level than would be expected based on the actual iron stores. This has been acknowledged as a limitation of ferritin as a sole marker of iron adequacy, and serves as a reminder that there are also other markers, such as soluble transferrin receptor, reticulocyte hemoglobin content, and perhaps direct measurement of hepcidin, that can help distinguish between inflammation-induced ferritin increase and true iron repletion. The present study did not measure these markers, but their addition to future studies would make the biological interpretations stronger [21,22].

There are a number of caveats to note. The cross-sectional design does not allow for causation conclusions and any discussion of mechanism should be interpreted as a possible explanation based on existing physiological literature, not as a conclusion based on this set of data. The study was limited to male workers, which makes the results less generalizable, and female shift workers have other hormonal and reproductive factors that can act independently and influence iron metabolism, and which constitute a critical gap in the occupational health literature. The dietary intake of iron was not measured, so there is an unmeasured confounding of food quality and iron bioavailability between the two groups. The mechanism mentioned in this discussion, based on the hormone hepcidin, was untouched by direct measurement, so the suggested pathway is still speculative. Lastly, the number of subjects was sufficient for the main comparisons, but too small for subgroup analyses and requires increased sample size for future studies.

Conclusion

The current results point out that night shift work is connected with the significant decrease in the level of serum iron in comparison with that in day shift work. In addition, it should be mentioned that this trend was even more evident for those employees who had more experience in night work, as their level of iron was lower than that of others working during the day time. Thus, it becomes evident that there exists a connection between long-term night work and changes in the physiological condition. It is possible to state that such connection exists because of the impact of several different factors (change of sleep-wake cycle, diet, etc.). All these elements could result in the depletion of iron and the occurrence of certain problems. However, the level of ferritin did not change considerably.

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