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Association between shift work and levels of thyroid hormones and interleukin-37

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Objective. Symptoms of thyroid defects involve sleep disorders, gain or loss of weight, tremors of hand, constipation, dry skin, bradycardia, diarrhea, irregular menses, and hot or cold tolerance. Workers in different professions face different situations that can lead to stress. This study aimed to examine the association between shift work (office or irregular) and thyroid function indicators, interleukin-37 (IL-37) levels, and development of chronic diseases (cardiovascular, diabetes).

Methods. The current study comprised three groups: 1) office staff, 2) irregular staff (comprising different jobs), and 3) controls (not working subjects). Eighty-five subjects, aged 30–55 years, were included in the study. Body mass index (BMI) was calculated for each participant. Venous blood was collected and serum levels of triiodothyronine (T3), thyroxine (T4), thyroid-stimulating hormone (TSH), and thyroid peroxidase (TPO) were determined by electrochemiluminescence (ECL) analysis using an automatic immunochemical analyzer Cobas E 411 (Roche Diagnostics, Germany). Serum IL-37 levels were measured using ELISA kit.

Results. Serum T3 ($p<0.01$) and T4 ($p<0.001$) levels were significantly decreased in both office and irregular shift work groups compared to the controls. The mean T3 levels were higher in subjects with irregular shift work (1.162±0.11 ng/mL) compared to subjects with office shift work (1.14±0.12 ng/mL). Significantly increased TSH and TPO serum levels ($p<0.01$) were found in both the irregular shift work and office shift work groups compared to the control group. Similarly, IL37 levels were significantly increased in office shift work (294.8±21.05 ng/mL) and irregular shift work subjects (278.0±16.22 ng/mL) ($p<0.001$) compared to controls (56.5±0.28 ng/mL). Cardiac disease ($p<0.01$), hypertension ($p<0.001$), and diabetes ($p<0.001$) showed significant differences between subjects with office shift work, irregular shift work, and the control group.

Conclusion. Elevated levels of IL-37 and TSH in shift workers may serve as biomarkers of the impact of shift work and the workplace on the immunological and hormonal status of employees.

Keywords: triiodothyronine (T3), thyroxine (T4), thyroid-stimulating hormone (TSH), interleukin-37

The thyroid gland is one of the largest endocrine glands in the human body and is located in the lower front part of the neck. It weighs 25 g and is involved in regulating body temperature and metabolic rate. Common reasons for thyroid defects include iodine deficiency, autoimmune illness, race, and geographical factors (Jaafer et al. 2023). Thyroid defect symptoms involve sleep disorders, gain or loss of weight, tremors of hand, constipation, skin dry, bradycardia, diarrhea, irregular menses, and hot or cold tolerance (Achanta and Khasbage 2022). Workers in different professions face different situations that can lead to stress. A person can tolerate stress due to social relationships in which they interact with people present in the workplace. Thus, stress has both its dark and bright sides (Ugur et al. 2007). Stress at

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work and stress factors in the working environment create conditions that make life difficult and warrant the term negative health effects of workplace stress (Mark 2008). Stress can hurt the productivity. It acts as a barrier and leads to negative outcomes. It causes blocks that prevent people from achieving their goals. In an office, stress leads to behavioral changes that generally reduce cooperation among the office employees (AbuAlRub and Al-Zara 2008).

Job stress occurs when the demands of a job are too much for an employee's skills. If stress causes physical or mental harm, it represents a loss for that person. Stress can harm productivity and affect office results. This damage will ultimately impact yield and outcomes (Blaug et al. 2007). Stress associated with an imbalance between demands imposed by external sources and the ability to meet these demands usually leads to physiological and physical consequences (Kaur 2023). Jeong et al. (2023) observed that nurses had undesirable performance scores in psychosocial relationships with patients because the nursing staff studied had high stress and psychological incapacity, which made it impossible for them to provide psychological assistance to patients. The shift work is defined as "a method of organization of working time, in which workers succeed one another at the workplace so that the establishment can operate longer than the hours of workers" (Silva et al. 2020).

According to NIOSH (National Institute of Occupational Safety and Health), researchers define shift work as any work type other than regular work hours (7:00 am to 6:00 pm) (Woo et al. 2022). Shift work causes health issues and conflict with circadian bio-rhythms by exposing workers to more light at night and leading to the prevention of the production of melatonin (Melendez-Fernandez et al. 2023). The lower quality of sleep duration induces an imbalance in hormones appetite that elevated feelings of hungry and metabolism alteration, which lead to insulin resistance, obesity, and decreased lipid tolerance (Mesarwi et al. 2013). The circadian rhythm disruption can lead to adverse outcomes, including the progression and worsening of various gastrointestinal problems and diseases (Voigt et al. 2019). In 2019, IARC (International Agency for Research on Cancer) has deducted that "night shift work" may be carcinogenic in humans (Group 2A carcinogen) (Hansen and Pedersen 2024).

Thyroid gland produces hormones including triiodothyronine (T3) and thyroxine (T4) that are necessary for the body metabolism, production of heat, appropriate development, and cell differentiation, and growth (Shahid et al. 2025). The hypothalamic-pituitary-thyroid axis monitors thyroid function, including the production of the hormones T3 and T4, which are secreted in response to thyroid-stimulating hormone (TSH) by the pituitary gland (Ortiga-Carvalho et al. 2016). During normal sleep, plasma TSH levels begin to increase in the late afternoon or early evening before bedtime and reach peak levels in the early part of the night (Mariotti et al. 2000). After the nocturnal peak, TSH levels decrease in the rest of sleeping period up to meet the low diurnal level. Sleeping has been proved to have an inhibitory effect on TSH production as evidenced by increased hormone secretion due to sleep deprivation (Leso et al. 2020). Many people, especially night-shift workers, have caused circadian disruption by desynchronizing their sleep/wake rhythm from their biological time system (Boivin et al. 2022).

Several studies suggest that sleep disorders are associated with an increased risk of thyroid cancer (Luo et al. 2013; Cho 2021). A link between night work, benign thyroid disease, and thyroid hormone disturbances (Leso et al. 2020; Coppeta et al. 2020) including higher TSH levels (Moon et al. 2016), a marker linked to thyroid cancer risk (Haymart et al. 2008), has been demonstrated. The association of rotating night shift work, sleep duration, and sleep difficulty with thyroid cancer risk has been evaluated in the Nurses' Health Study 2 (NHS2) by Papantoniou et al. (2023). Over 26 years of follow-up, 588 incident cases were identified among 114,534 women in the NHS2 cohort. The authors found modest evidence for an increased risk of thyroid cancer in relation to sleep difficulty, which was more pronounced among night shift workers. Difficulty falling or staying asleep was suggestively associated with a higher incidence of thyroid cancer when reported sometimes (hazard ratios, HR 1.26, 95% CI 0.95, 1.66) and all or most of the time (HR 1.35, 95% CI 1.00, 1.81). Night shift workers (>10 years) with sleep difficulty all or most of the time (HR 1.47; 0.58-3.73) or with >7 h of sleep duration (HR 2.17; 95% CI, 1.21-3.92) had a higher risk of thyroid cancer (Papantoniou et al. 2023).

In the context, both environmental and occupational exposure have been demonstrated to affect biochemical and physiological factors. For example, Ali et al. (2022) found significant changes in biomarkers of liver and kidney functions in workers in Baghdad due to exposure to exhaust emissions from diesel generators. Although this study did not evaluate thyroid function directly, it illustrates the capability to be affected by occupational and environmental stressors, which weaken homeostatic mechanisms, of which thyroid regulation is one. This is reflected in the premise of this research, whereby shift work as an occupational stress may affect the balance of thyroid hormones and the immune marker IL-37 as well.

This study aimed to examine the association between office shift work or irregular shift work and thyroid function indicators (T3, T4, TSH levels), interleukin-37 (IL-37) levels, and development of chronic diseases (cardiovascular, diabetes).

Materials and Methods

Personal and job demographics. We obtained the following data from all study participants: age, gender, whether they were smokers, marital status, education, work history, and number of children. Exclusion criteria were infections, such as influenza-like infection, and chronic diseases.

Ethical approval and participants' consent. Consent from all participants in this study was obtained for sample collection. The ethical approval is in accordance with the Helsinki declaration from the committee of the Scientific Research Commission/ Research and Technology Center of Environment/ Water and Renewable Energy to conduct the study (SRC/115/0001).

Study population. In the current study, 255 samples were collected and divided into three study groups: 1) office staff, 2) irregular staff (representing various job positions), and 3) controls (without work). Eighty-five subjects aged 30–55 years were included in each study group. Of these, in the age range of 30–39 years, the number of office employees was 38 (44.7%), irregular employees 30 (35.3%), and there were 32 (37.6%) subjects in the control group. In the age range of 40–55 years, the number of office workers was 47 (55.3%), irregular workers 55 (64.7%), and controls 53 (62.5%).

Samples collection. Venous blood samples (5 mL) were collected from individual volunteers (workers and controls) into gel tubes, sera were separated by centrifugation at 5000 x g for 5 minutes, then placed in Eppendorf tubes and stored at -20 °C until analysis.

Body mass index. We measured the weight of all study participants using a portable scale and their height using a tape measure. All measurements were taken twice and averaged. Body mass index (BMI) was calculated using the formula BMI=weight (kg)/ height^2 (m^2) (Nuttall 2015).

Assessment of thyroid functions. T3, T4, TSH, and thyroid peroxidase (TPO) serum levels were determined by electrochemiluminescence (ECL) analysis using an automatic immunochemical analyzer Cobas E 411 (Roche Diagnostics, Germany).

Interleukin-37 determination. Serum IL-37 levels were measured using Human Interleukin 37, IL-37 ELISA kit (BT LAB Bioassay Technology Laboratory, China). The sensitivity of the kit was 4.56 ng/L and the detection range 7–400 ng/L. The intra-assay ranged from 3.1 to 3.6 (CV%).

Statistical data analysis. The data are presented as mean±SEM. The median age (interquartile range) was calculated online. One-way analysis of variance (ANOVA) with Tukey post-hoc test was used to analyze statistical differences among groups. The probability of a difference from the control group was statistically significant if $p<0.05$. Pearson's Chi-square analysis was used for categorical variables. ROC curves were created to assess the area under the curve (AUC). The 95% confidence interval (CI) was examined. Cutoff points, sensitivity, and specificity were also analyzed. Sample size power was calculated by the G Power software.

Results

Sample size power. The sample size of controls (n=85) and workers (n=85) was calculated by G power software. The actual sample size power, determined at a two-tailed alpha error probability value of 0.05 and an effect size of 0.5, was 0.872. The acceptable sample size power was 0.80.

Baseline characteristics. The mean age showed nonsignificant differences between the office shift, irregular shift, and control group. Workers and controls were divided into two age groups (<40 and ≥40 years) and there was no significant variation between these age groups (p=0.43) (Table 1). Smoking showed a significant difference between the office shift, irregular shift, and control group (p=0.0004). No significant difference was found between the office shift, irregular shift, and controls (p=0.06) for BMI, while cardiac disease, hypertension, and diabetes showed significant differences between the office shift, irregular shift and control groups (p=0.007, p=0.0005, p=0.0001, respectively) (Table 1).

Workers (40–55 years) had a decreased prevalence of short sleeping duration, impaired sleeping-associated ADL (activities of daily living), and insomnia compared with those aged 30–39 years.

Laboratory results. Statistically significant differences were found between the experimental groups)including office shift, irregular shift) and the control group for serum levels of T3 (p=0.0001), T4 (p<0.002), and TSH (p=0.01), as shown in Table 2 and Figure 1. The mean serum levels of T3 were significantly higher in the irregular shift group (1.162±0.11 ng/mL)

Table 1
Baseline characteristics of study participants (office staff, irregular staff, and controls)

Parameter	Type of shift work		Controls (N=85)	p-value
	Office staff (N=85)	Irregular staff (N=85)		
Age				
30–39 years	38 (44.7%)	30 (35.3%)	32 (37.6%)	
40–45 years	47 (55.3%)	55 (64.7%)	53 (62.5%)	0.430
Median (IR)	37.8 (31.5–42)	40.2 (35–45)	39.24 (34–44.5)	
Smoking				
Yes	32 (37.6%)	40 (47.1%)	16 (18.8%)	0.0004
No	53 (62.5%)	45 (52.9%)	69 (81.2%)	
BMI (kg/m^2)	27.60±0.22	28.35±0.64	28.35±0.63	0.600
Cardiac disease				
Yes	25 (29.4%)	22 (25.9%)	9 (10.6%)	0.007
No	60 (70.6%)	63 (74.1%)	76 (89.4%)	
Hypertension				
Yes	23 (27.1%)	27 (31.7%)	7 (8.2%)	0.0005
No	62 (72.9%)	58 (68.3%)	78 (91.8%)	
Diabetes				
Yes	30 (35.3%)	43 (50.6%)	15 (17.6%)	0.0001
No	55 (64.7%)	42 (49.4%)	70 (82.4%)	

Abbreviations: BMI – body mass index, IR – interquartile range. Data are presented as mean±SEM or median (IR). One-way analysis of variance (ANOVA) with Tukey post-hoc test was used to analyze statistical differences among groups (p<0.05).

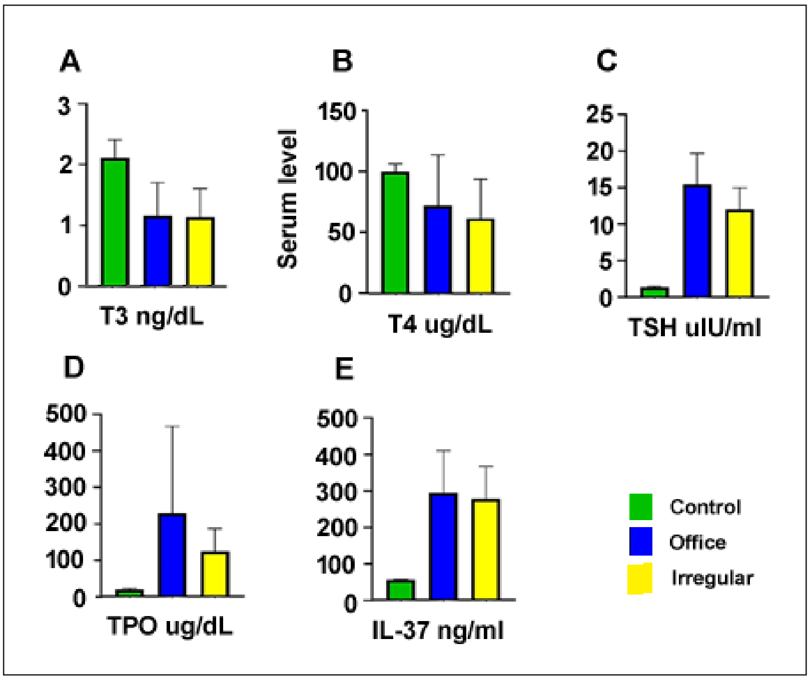

Figure 1. Serum levels of triiodothyronine (T3) (**A**), thyroxine (T4) (**B**), thyroid-stimulating hormone (TSH) (**C**), thyroid peroxidase (TPO) (**D**), and interleukin-37 (IL-37) (**E**) in the office shift workers, irregular shift workers, and control group.

Table 2
Laboratory results of study participants (office staff, irregular staff, and controls

Parameter	Type of shift work		Controls (N=85)	p-value
	Office (N=85)	Irregular (N=85)		
T3 (ng/mL)	1.14±0.12	1.162±0.110	2.11±0.10	<0.0001
T4 (nmol/L)	61.37±7.80	71.78±10.12	99.72±1.50	0.002
TSH (µIU/mL)	15.46±4.20	12.04±2.90	1.40±0.10	<0.01
TPO (µg/dL)	124.80±65.98	229.20±17.24	19.62±1.23	0.002
IL-37 (ng/mL)	294.80±21.05	278.00±16.22	56.50±0.28	<0.0001

Abbreviations: IL-37 – interleukin 37; T3 – triiodothyronine; T4 – thyroxine; TPO – thyroid peroxidase; TSH – thyroid-stimulating hormone. Data are presented as mean±SEM. One-way analysis of variance (ANOVA) with Tukey post-hoc test was used to analyze statistical differences among groups ($p<0.05$).

Table 3
Pearson correlation coefficients (r) between laboratory results of study participants

Parameter	TSH	IL-37	TPO	T3	T4
TSH	1	–	–	–	–
IL-37	0.183	1	–	–	–
TPO	–0.019	0.085	1	–	–
T3	–0.425*	–0.502*	–0.099	1	–
T4	–0.283*	–0.250	–0.163	0.292*	1

Abbreviations: IL-37 – interleukin 37; T3 – triiodothyronine; T4 – thyroxine; TPO – thyroid peroxidase; TSH – thyroid-stimulating hormone. *$p<0.05$.

and the office shift group (1.14±0.12 ng/mL) compared to the control group (2.11±0.1 ng/mL) ($p<0.0001$). Also, T4 serum levels were higher in the irregular shift group (71.78±10.12 nmol/L) and the office shift group (61.37±7.80 nmol/L) compared to the controls (99.72±1.5 nmol/L) ($p=0.002$). Mean serum levels of TSH in the irregular office shift group (12.04±2.9 µIU/mL) were lower than the TSH levels in the office shift group (15.46±4.2 µIU/mL) compared to the control (1.4± 0.102µIU/mL) ($p<0.01$). The mean TPO serum levels in the office shift group (124.8±65.98 µg/dL) and irregular shift group (229.2±17.24 µg/dL) were significantly increased compared to the control group (19.62±1.23 µg/dL) ($p=0.002$). IL-37 serum levels were significantly higher in the office shift group (294.8±21.05 ng/mL) and the irregular shift group (278.0±16.22 ng/mL) compared to the controls (56.5±0.28ng/ml) ($p<0.0001$) (Table 2).

ROC curve analysis. ROC analysis showed high predictive accuracy for TSH (AUC=0.898; cutoff = 1.99 ng/mL, sensitivity = 83.33%, and specificity = 80.95%), TPO (AUC=1; cutoff = 26 ng/mL, sensitivity = 100%, and specificity = 83.33%), and IL-37 (AUC=1; cutoff=58.25 ng/mL, sensitivity=80.95%, and specificity=100%) (Figure 2).

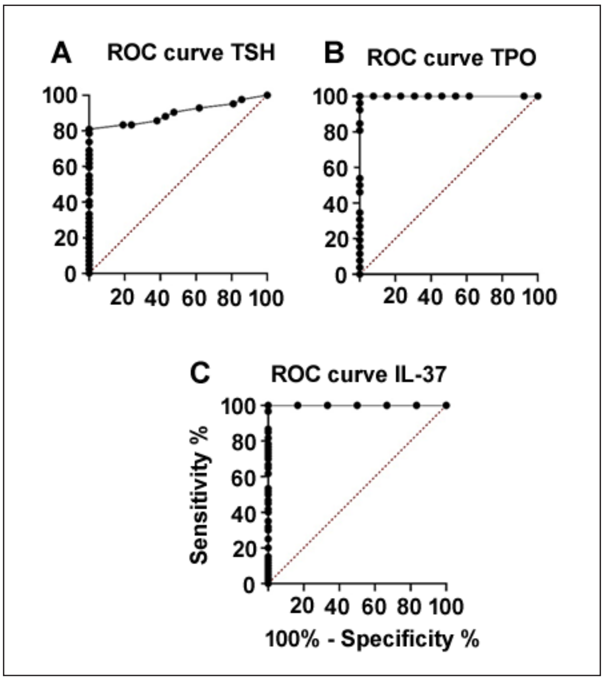

Figure 2. ROC (receiver operating characteristic curve) data that represent sensitivity and specificity of the studied groups for thyroid-stimulating hormone (TSH) (**A**), thyroid peroxidase (TPO) (**B**), and interleukin-37 (IL-37) (**C**).

Correlation analysis. Pearson's Chi-square analysis revealed significant negative correlation between TSH and T3 (r=−0.425) and TSH and T4 (r=−0.283) (both p<0.05). Significant negative association was found between IL-37 and T3 levels (r =−0.502) (p<0.05). T4 levels significantly positively correlated with T3 levels (r=0.292) (p<0.05) (Table 3).

Discussion

Shift work has become a significant health problem in recent years. Strong evidence links shift work to an increased risk of various diseases, including cardiovascular diseases, chronic kidney disease, type 2 diabetes mellitus, and cancer (Wong et al. 2023).

The current research demonstrated that shift workers, whether office or irregular shift workers, were overweight based on their BMI (25–29.9 kg/m^2). Despite this, there were no significant differences in their BMI compared to the control group. From this observation, it can be hypothesized that other non-work-related factors may affect body weight among this group of individuals. These results suggest that variables above the work schedule may impact the body weight of shift workers. The fact that the BMI was consistent across each group, despite the differing work patterns, further indicates that body mass regulation in shift workers can be complicated, and that several interrelated lifestyle and physiological factors may be involved beyond activity levels (e.g., eating, sleep and metabolic adaptation related to the disruption of circadian rhythms). To step this more in depth, one Iraqi study conducted by Alsaffar et al. (2024) showed significant changes in the expression of hormones related to appetite (leptin and ghrelin, and their respective receptors) in a cohort of obese Iraqis; emphasizing the importance of hormones associated with appetite in aiding weight management. An endocrine model provides a unique rationale that can perhaps explain the differences in overall weight patterns seen in our sample, suggesting that perhaps it is not merely the consumption of food through a period of shift work, but rather a potential indirect mechanism involved with hormonal processes related to appetite and overall metabolic processes. While Gomero et al. (2018) found that obesity was implicitly a factor with respect to work absenteeism, it is still unclear the implications of incremental obesity with respect to work performance. Both Gomero et al. (2018) and Alsaffar et al. (2024) demonstrated the need for a multifactorial understanding of weight management in occupational health, in particular with shift workers and the implications of their health behavior on their weight and subsequent health outcomes.

The current study showed that TSH, TPO, and IL-37 levels were higher in the office shift group and irregular shift work group compared to the non-work group. This type of work can lead to serious health problems, like sleep disorders, metabolic disorders, heart diseases, mental health problems, and some types of cancers. These health disorders arise due to the disruption of the circadian clock system, which is associated with alterations of gene expression (Silva et al. 2020).

Chronic stressful conditions during shift work may induce an imbalance in immune system leading to overproduction of glucocorticoids and catecholamines through activation of the hypothalamus-pituitary-adrenal (HPA) axis. This process, in turn, may result in the development of abnormal thyroid function (Vaivode et al. 2023). Acute stress can cause a decrease in circulating T3 levels within 120 min post-stress initiation. This decrease in T3 induced by stress likely results from reduced secretory activity of the thyroid gland rather than alteration in the ratios of conversion pathways of T4 to T3 or rT3 (Helmreich et al. 2006). Ahmed and Ramzan (2013) showed how work-related stress affects job performance in Pakistan's banking sector. The study using data from 144 workers found a negative link between work-related stress and worker performance.

The outcomes of multivariate screening indicated that working hours and age were associated with thyroid dysfunction in medical personnel in radiotherapy units, and in older healthcare workers, longer years of work were positively associated with thyroid dysfunction, which is consistent with some previous studies (Lee et al. 2020).

Wu et al. (2021) demonstrated that the risk of thyroid function abnormalities was higher in smokers than in non-smokers, and their TSH levels were significantly higher compared to non-smokers. Long working hours can lead to increased tobacco consumption, and subsequently nicotine and alkaloids in tobacco can cause a chronic inflammatory state in the body and stimulate the production of mediators by immune cells, which leads to accelerated deterioration of thyroid function (Cho et al. 2018). In addition, eating food, especially animal foods, late in the day for those who work shifts is also a bad habit and a risk factor that can affect thyroid function. (Bellastella et al. 2022).

Depression during shift work is another risk factor for thyroid dysfunction in medical workers. The induction of inverse psychological factors such as depression leads to disruption of neuroendocrine functions such as the HPA axis, leading to disorders of thyroid hormone secretion and affecting normal thyroid function (Dai et al. 2023).

Another powerful stressor that can trigger thyroid hormone release is physical activity. However, research findings on the effects of training programs (chronic exposure) on basal, resting thyroid levels do not drastically differ from those in sedentary normal populations. A few studies reported that the rate of T4 secretion was higher in exercise-trained individuals than in untrained individuals, although this is not a universal finding. The roles of thyroid hormones (T3, T4, TSH) in combination with prolactin in exercise are multifaceted, but one overlapping and common function is their combined aid and support of the tissue inflammatory responses after exercise (Hackney and Saeidi 2019).

In the present study, a group of office shift worker showed significantly increased TSH levels compared to irregular shift workers. Zhang et al. (2019) demonstrated that night shift workers had significantly higher TSH levels (0.303 mIU/L) compared to non-night shift workers over the 5-year study period (2011–2015).

Shift work represents a major disruption to the circadian system, which is entrained to the natural light-dark cycle, and consequently affects the body's ability to regulate metabolic hormones by changes in the biological clock machinery. In line with this mechanism, many studies have shown that nightshift workers exhibit dysregulated levels of multiple hormones, including leptin, melatonin, prolactin, testosterone, and thyroid hormones (Dragoi et al. 2024).

Interleukin-37 belongs to the interleukin-1 (IL-1) family with the ability to suppress inflammation associated with increased innate and adaptive immune responses by inhibiting the production of pro-inflammatory cytokines, thereby reducing the pathological effects of inflammation. Interleukin-37 precursor is produced upon pro-inflammatory stimuli to modulate inflammation and reduce extreme tissue destruction (Wang et al. 2018; Su and Tao 2021). In the present study, IL-37 serum levels were significantly increased in the office shift group compared to irregular shift workers, and were significantly higher in subjects of both shift work groups compared to controls. Elevated IL-37 levels may be related to secretion of catecholamines and higher blood pressure along with stress as consequences of chronic work, which may lead to cardiovascular diseases. The combination of physical and psychological stress can cause long-term inflammation and subsequent various diseases (Huang et al. 2013; Chu et al. 2024).

There are no previous studies available examining the effects of shift work on IL-37 levels. However, other studies have examined the association between shift work and production of cytokines such as IL-1β, IL-8, IL-10, and tumor-necrosis factor alpha (TNF-α) (Liu et al. 2021). Aquino-Santos et al. (2020) found that chronic alteration of circadian rhythm in shift work scale policemen resulted in impaired lung function, beyond to impair pulmonary and systemic immune function. They showed that shift work policemen had higher levels of nitric oxide (NO) and IL-2 in the exhaled breath condensate, indicating chronic lung inflammation, compared to civil men. Policemen presented also increased levels of IL-2 and lower levels of IL-10, clearly displaying a clinical status of low-grade inflammation. Another study revealed that only night shift work was significantly associated with high levels of IL-6 in Japanese workers. The initial cause is likely insufficient sleep and circadian rhythm disruption, which creates chronic stress and may be linked to elevated IL-6 levels. Lack of sleep and circadian rhythm disruption are much more common in night workers than in other workers (Amano et al. 2018).

Furthermore, long-term shift work and higher exposure to environmental and occupational stress reduce antioxidant capacity, which may increase the effect of increased oxidative stress production due to high workload (Zimet et al. 2016). Walker et al. (2016) have reported that inflammation and changes of the immunity system were linked to mood changes and decreased well-being, and therefore focus on the need to improve workplace risk management.

Limitations of our study include the relatively small sample size and the restriction to specific workplaces, which could potentially limit its generalizability. The cross-sectional design precludes establishing causality between shift work and changes in hormonal or immunological function. Only a limited number of biomarkers were assessed, and lifestyle factors including nutrition, stress, and sleep quality were not comprehensively assessed. Further longitudinal studies involving larger and more diverse groups are needed to confirm the results obtained.

Conclusion. This study indicate that the shifting work can affect the health of workers. It was observed that the increase of serum levels for IL-37 and TSH in shift work workers can be a biomarker in the effect of shift work and workplace on the immunological and hormonal statuses of workers.

Conflict of interest. *The authors declare no conflict of interest.*

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