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Impact of Benzo[a]Pyrene Exposure on Thyroid Gland of Individuals Workers In Oil Refining

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Abstract

Benzo[a]pyrene is very well known as a toxic substance. Exposure to several types of serious toxic impacts is associated with all individuals. This research determined the Benzo[a] pyrene level in the OR employees because they are subjected to pollutants all the time. For that reason, the Benzo[a]pyrene concentration was quantified in specimens of blood obtained from 70 occupationally exposed people (Polluted Group) and 70 persons (Control Group). Additionally, they evaluated the effects of Benzo[a]pyrene on thyroid function (TF). The findings showed that PG people had considerably greater blood benzo[a]pyrene levels ($P<0.05$) than CG persons. TSH and TT3 level differences between the PG and CG weren't significant in terms of statistics ($P>0.05$). TT4 and TRAb levels, however, were considerably greater in the PG than in the CG. The research found that levels of thyroid hormones (THs) are affected by long-term exposure to petroleum fuels, with higher [TR-Ab] in the PGs group than in the CGs.

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1. Introduction

Benzo[a]pyrene [BaP], like a typical hysteretic of Polycyclic Aromatic Hydrocarbons (PAHs), poses a great risk to the environment and people's health [1]. Polycyclic Aromatic Hydrocarbons (PAHs) are carcinogenic and also pose a risk to people and wild creatures [2]. BaP is a category of chemicals that result from the incomplete burning of organic matter [3]. BaP, another PAHs, is discovered in various environmental wildlife like air, soil, water, sediment, and also enters the body easily through skin contact, swallowing, and inhalation [4]. They can be present in fuel therefrom, such as heating oil, gasoline, and diesel, as well as in industrial operations like oil refining [5]. Oil refineries (ORs) are common sources for contamination as PAH's. The research field has provided much guidance that offers the presence of PAHs on the surface of the surrounding soil ORs. These compounds exhibit negative impacts on environment, like water also air contamination, which are still under investigation are also a reason for anxiety. They have also studied potential routes of exposure and assessed risks to the health of people and the environment [6]. According to authors, OR's impact is a serious risk to individuals when subjected to high levels of contaminants during an extended period of time [7,8]. Changes in the levels of thyroid hormones (THs) can impact the whole body due to THs are basis for controlling metabolism, development, also growth [9]. However, factors such as the type and concentration of contaminations, the density and length of exposure, also human susceptibility specify the precise impacts of ORs on THs [2]. Regarding to thyroid gland, epidemiological study has attached exposure to PAH to decrease levels of circulating also thyroxine tissue (TH), which may be because direct poisoning to the thyroid hormone or interference with the expression of TH binding proteins or iodothyronine deiodinase2 (DIO2) production [10]. BaP's carcinogenic qualities and reproductive toxicity to marine animals prompted much of the research on the material, although little is known about how it affects thyroid function. After receiving a BaP injection, mullet liza abu fish shown dramatic changes

in TSH concentration, TH plasma level, and thyroid follicular pathology [11]. However, the exact mechanism causing thyroid toxicity is yet unknown. Animal studies have offered additional proof of these findings because contact with PAHs changed the peroxidation of lipids and disrupted the signaling pathway, as well as generating toxicity in fish [12]. Petroleum or natural oil derivatives contain some chemicals that combine to form PAH's. Additionally, they could be released during various stages of the shipment, treatment, and upgrading of oil [13]. Types of contamination (such as polycyclic aromatic hydrocarbons and heavy elements) are typical signs of oil and coal combustion [14]. Benzo(a)pyrene is the most common examinations that are used for marker of poly_cyclic aromatic hydrocarbons (PAH's) contamination [15]. Structure of T3, T4 and BaP as shown in figures (1, 2 and 3). The objective of the research is to measure the OR employees' blood benzo[a]pyrene concentrations and investigate their effects on thyroid function.

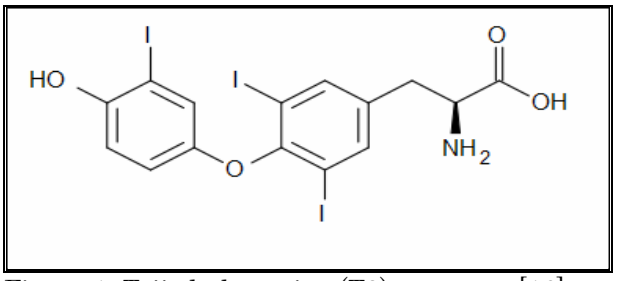

Figure 1. Triiodothyronine (T3) structure [16].

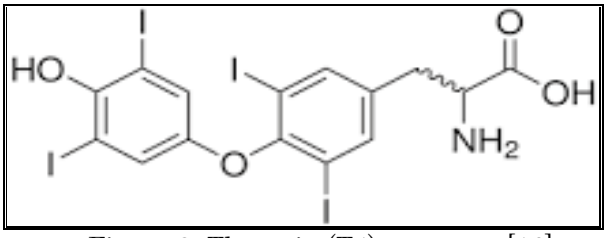

Figure 2. Thyroxin (T4) structure [16].

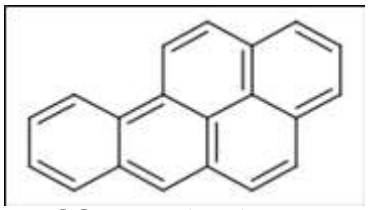
Figure 3. Benzo[a]pyrene (BaP) structure [17].

2. Materials and Methods

2.1. Exclusion criteria

A total of 140 people without thyroid or endocrine disease symptoms participated in the study; smokers and pregnant women were not included.

2.2. Blood sample collection

Seventy healthy individuals between the ages of 25 and 60 and seventy OR workers from the Al-Dura Refinery in Baghdad and aged 35 to 60 years participated. Samples were gathered in the laboratories of Abdul Mageed Private Hospital from December 2023 to March 2024. After drawing five mL of venous blood, it was allowed to clot at an ambient temperature (25°C) for ten to fifteen minutes, also centrifuged for ten min at 4000 rpm. The sera were then separated into sterilised Eppendorf tubes, also kept until examination at -20°C. The institutional ethics committee gave its approval to this work (20234740).

2.3. Hormone and Analytical Assessment

Employing the gas chromatographic method (Shimadzu® GC-2010, Japan), the amount of benzo[a]pyrene in the specimens of serum was determined. Additionally, they used the Roche cobas® e_411 auto analyzer (Germany) to measure the levels of thyroid-stimulating hormone (TSH), TT4, and TT3, while TRAb levels were gauged using enzyme-linked immunosorbent assay (ELISA) kits.

2.4. Measurement of human serum Benzo[a]pyrene

2.4.1. Sample preparation

Human serum of benzo[a]pyrene was measured by gas chromatography (GC) with a flame ionization detector. The analysis requirements of benzo[a]pyrene were as follows:

Table 1.The analysis requirements of Benzo[a]pyrene.

Parameter	Conditions of GC
Column Oven (ZB - 5)	100 (hold 1 min) - 300 °C (hold 2 min) (10 °C / min)
Detector	Flame Ionization Detector
Injection Volume	1 µl
Injection Temperature	280 °C
Pressure	100kpa
Temperature Detector	330 °C
Carrier Gas	He (Helium)
Flow Rate	5 mL/min
Oven program (Temperature)	(40 heighted to 120) °C for one minute, at the speed of 25 °C/min, after that (160) °C at the speed of ten °C/min, also lastly to (300) °C at the speed of 5 °C/min, it is kept at the last temperature for fifteen minutes.

2.4.2. Procedure

A liquid-liquid extraction method using a 4:1 ratio of n-hexane also dichloro-methane was used. Extraction (10 mL) also serum (2 mL) were put into the sealed bottles & shaken at 300 rpm for 20 seconds. Utilizing the pipette containing a pipette tip inside a sterile amber bottle, the organic matter was absorbed and heated. Extract was cleaned up into a column (15 cm x 1cm, length and internal diameter) with silica gel slurry as the stationary phase, pre-prepared with distilled water, also n-hexane/DCM before the sample was eluted, collected as well as concentrated in the stream of nitrogen gas [18].

2.5. Body Mass Index (BMI)

The BMI value was calculated using the BMI formula. BMI= Weight (kg) / Height (m). Weight status was divided by BMI value, underweight (< 18.50), normal (18.05-24.90) and overweight (25.01-29.9) [33].

2.6. Statistical analysis

Statistically analyzed the data utilizing SAS/STAT® 9.1. The mean ≝ standard deviation (STD) was used to express each outcome. The independent t- test was used to evaluate the significant differences between the two groups' mean values. For each of the identified parameters, the correlation coefficients (r) were computed. The statistical program MedCalc was utilized to analyze the data. Values showing P<0.05 were deemed statistically significant.

3. Results and Discussion

Standard criteria were used to select the subjects of the present study, in which the CG and PG were comparable in age and BMI distribution to prevent these parameters from influencing the results of the study. Table 2 shows the most current traits of every participant in this research. According to the statistics, the mean age of the CG group was 45.28±8.22, but the PG group's mean age was 49.68±9.30. Additionally, the mean BMI for those in CG was 26.35±2.87 kg/m2, but for those in PG was 26.73±3.32 kg/m2.

Table 2. Displays characteristics of the research participants.

Parameter	Polluted group	Control group
Age (year)	49.68±9.30	45.28±8.22
BMI (Kg/m2)	26.73 ±3.32	26.35±2.87

Table 3 shows TT4, TSH, TT3, Benzo[a]pyrene, and TRAb results as mean ± SD. Differences in TSH levels were not significant (P>0.05) between CG (1.86±0.94 µIU/mL) and PG (2.33±1.86 µIU/mL). Total triiodothyronine (TT3) levels were non-significant (P>0.05) between CG (1.69±0.28 nmol/L) and PG (1.86±0.63 nmol/L). Total thyroxine (TT4) levels were significantly higher (P<0.05) in PG (118.27±23.14 nmol/L) compared to CG (99.76±20.16 nmol/L). Thyroid-stimulating hormone receptor autoantibody (TRAb) levels were also significantly higher (P<0.05) in PG (0.76±0.78 IU/L) compared to CG (0.43±0.20) IU/L. The levels of benzo[a]pyrene in the PG subjects (3.24±1.00 ppb) were substantially higher (P<0.05) than those in the CG subjects (0.12±0.03 ppb).

Table 3. Displays characteristics of the research participants.

Parameter	Polluted group	Control group	p-value
TT3 (nmol/L)	1.86±0.63	1.69±0.28	0.07
TT4 (nmol/L)	118.27±23.14	99.76±20.16	<0.0001
TSH (ulU/mL)	2.33±1.86	1.86±0.94	0.11
TRAB (IU/L)	0.76±0.78	0.43±0.20	0.005
Benzo[a]pyrene (ppb)	3.26±1.00	0.11±0.03	<0.0001

Crude oil naturally contains benzopyrene. In this investigation, the benzo[a]pyrene amounts in the PG people were observed to be 3.24±1.00 ppb. Because of a previous study that illustrated the detrimental effects of ORs on people's health, many refineries are located outside of urban areas [19]. Nonetheless, some ORs are located in urban areas nearer to the customers. According to a previous study, Jordanians who lived close to an OR had negative health impacts, such as skin conditions, respiratory problems, and a generalized sense of being unwell [20]. There are a lot of ORs in Baghdad, as in many other cities in developing countries. It is unclear, therefore, how these important amenities affect the health of people. Additionally, those who are surrounded by the atmosphere at ORs on a regular basis may have volatile organic compounds (VOCs) that have not been assessed by any study in the past. As a result, by measuring the amount of PAHs, in the present investigation, the researchers looked at how OR affected people's health. Privatization of these VOCs might result from a number of causes. Because of their

poisoning and potential negative impacts on health, PAHs also TPHs are especially hazardous volatile organic compounds [21]. They are known carcinogens and are attached to many health problems, such as cardiovascular disease [22], infertility [23], dyslipidaemia [24], also liver malfunction [25]. To ascertain the link between benzo[a]pyrene and other variables in the blood specimens gathered for this research, a correlation analysis using Pearson's r was employed. The results in Table 5 showed that the levels of PGs and the variables under study did not significantly correlate, also show a positive relationship between serum benzo[a] pyrene and other parameters. Among the studied parameters of the PG and CG except the significant positive relationship between TT3 and TT4 in CG, as shown in Table 4.

Table 4. Correlation between thyroid hormone and benzo[a]pyrene at control group.

Control group	TT3		TT4		TSH		TRAB	
Variables	R	p	r	P	r	p	r	p
TT3	-	-	0.416**	0.003	-0.085	0.555	-0.131	0.366
TT4	0.416**	0.003	-	-	-0.098	0.498	-0.191	0.185
TSH	-0.085	0.555	-0.098	0.498	-	-	0.013	0.927
TRAB	-0.131	0.366	-0.191	0.185	0.013	0.927	-	-
Benzo[a]pyrene (ppb)	0.096	0.509	0.045	0.755	-0.002	0.989	-0.033	0.821

** : Highly significant

Table 5. Correlation between thyroid hormone and benzo[a]pyrene at polluted group.

Polluted	TT3		TT4		TSH		TRAB	
Variables	r	p	r	p	R	p	r	p
TT3	-	-	0.190	0.186	-0.028	0.848	-0.095	0.511
TT4	0.190	0.186	-	-	-0.048	0.738	-0.248	0.082
TSH	-0.028	0.848	-0.048	0.738	-	-	0.001	0.993
TRAB	-0.095	0.511	-0.248	0.082	0.001	0.993	-	-
Benzo[a]pyrene	0.036	0.803	0.209	0.145	0.000	0.999	0.167	0.246

PG results indicated that long exposure to ORs may negatively impact TF [26]. Several studies have investigated the relationship between Thyrotropin (TSH) levels and exposure to industrial contaminations like ORs [27]. This is due to these pollutants have the possible to interfere with metabolism also the production of Thyroid Hormone (TH) as well as induce disturbances like hyper- or hypothyroidism [28]. Serum TSH levels of PG were not significant in terms of statistics ($P > 0.05$). Hormone of TSH levels in PG were substantially lower than those in CG, according to Rafad et al. (2022) [27]. Simultaneous results to the present research were mentioned by Mohammed et al. (2023) [29]. Total Triiodothyronine (TT3) levels of the PG and CG were not significantly various from one another ($P>0.05$), but they weren't like that mentioned by Mohammed et al. (2023) [29], who found significant differences in serum TT3 levels for PG and CG. Thyroid hormone levels can be affected by several factors like sample size, variability among subjects, or exposure levels could have influenced these outcomes. Sutuyeva et al. (2020) [30], however, exposure to petroleum products has been reported to significantly reduce TF levels, particularly T3 also T4. According to the results, the high serum TRAb concentration of PG can be return to thyroid dysfunction due to direct and prolonged exposure to benzene and air pollutants, leading to the increased synthesis of TSH [31]. PG results show a positive relationship, high exposure to ORs may lead to hypothyroidism. Zaccarelli-Marino et al. (2019) [32], similarly, people living near industrial contaminants have been reported to have significantly lower levels of TF.

4. Conclusions

This research used a novel comparative analytical technique to identify the complex relationship among exposure to BaP, TRAb, and TH in OR workers. The results revealed a significant increase in the level of BaP in the OR staff. This raises significant concerns about the possibility of health risks not just for OR personnel but also for those who reside close to these pollutants. The study also revealed that prolonged exposure to petroleum fuels disrupts TRAb and TH levels, which may affect other body systems. Current knowledge on the health effects of ORs remains deficient, which requires more research.

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