Original Article

ASSESSMENT OF THYROID HORMONES AND TESTOSTERONE AMONG TRAMADOL–ABUSERS IN GAZA STRIP–PALESTINE

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ABSTRACT

Introduction: Tramadol is a prescription opioid used to treat moderate pain. But among Tramadol-abusers in Gaza Strip, little is known regarding Tramadol's effect on their body functions such as levels of thyroid hormones and testosterone**.** The aim of the study was to investigate the effect of tramadol abuse on the level of thyroid hormones and testosterone among tramadol-abusers in Gaza Strip. **Methods:** This study used a prospective case-control design performed on a population of Tramadol overdosed patients, aged 18 to 35 years old who were admitted to psychiatric clinics in the Gaza Strip. Questionnaires on sociodemographic and clinical data were collected from 196 males (98 cases from tramadol abusers who followed before pre-detoxification and one-month later post detoxification and an equal number of healthy individuals with matching age as control. TSH, FT4, FT3, and Testosterone levels were measured, statistically analyzed. **Results**: There was an association between tramadol abuser's pre-treatment and healthy subjects for thyroid parameters and testosterone. T-test showed tramadol abusers pre-detoxification were significantly increased in TSH levels than controls ($P<0.001$) conversely, they were significantly decreased in serum FT4, FT3, and testosterone levels than controls also paired t-test illustrated level of TSH for pre-detoxification were significantly higher than post-detoxification. Additionally, there is no significant correlation between controls and tramadol abusers' groups (pre & post-treatment) in hematological profile ($P>0.05$) except in red cell distribution width (RDW) that showed a positive significant correlation between controls and tramadol abusers groups in both pre & post-treatment ($P=0.021$). **Conclusion:** The study concluded that tramadol abusing have a significantly decreased level of Testosterone, FT4, and FT3 levels and a significantly high level of TSH, also we conclude that the level of testosterone, T3, and T4 in pre detoxification of tramadol abusers was significantly lower than controls and significantly lower than its results after detoxification.

Keywords: opioids, tramadol, thyroid function, testosterone, Gaza Strip

Introduction

Tramadol (Tramal TM) is a synthetic opioid analgesic (painkiller) that is frequently prescribed to manage moderate to severe levels of pain such as that experienced after surgery or in chronic conditions like arthritis (Gay-Escoda et al., 2019). There has been an increasing rate of recognizing problems connected to prolong use of opioids (Rhodin et al., 2010), these opioids including narcotics, opium, morphine, meperidine, and tramadol which have high rate consumption around the world (Lewis et al., 1997).

In present days tramadol is widely used for the treatment of different pain disorders and it is the most commonly prescribed opioid in the world (Rosenblum et al., 2008). It is considered a centrally effective drug used for the treatment of moderate to severe pain, including cancer pain, the pain of surgery, muscle and joint pains, its effectiveness is less than morphine and pethidine and is stronger than ibuprofen, it is distributed in the body with a mean distribution half-life of 1.7 hours (Macintyre et al., 2010).

According to Australian studies, tramadol use has increased almost 10 times between 1998 and 2004 (Mehrpour et al., 2015). It has been approved for use in many countries for many years such as the united states since 1995 and in France since 1997 (Shadnia et al., 2008). Nowadays Tramadol has a clear prevalence in various Arabic countries such as Egypt. it is noticed that tramadol abuse in the Egyptian community was increased in the last years (Fawzi, 2011). Other neighboring countries suffer from this problem increasingly alarming phenomenon of Tramadol such as the United Arab Emirates which has been on the top in the phenomenon of selling and trafficking of Tramadol since January 2010.

Also, Tramadol abuse has been heavily demonstrated in Gaza, it is the most used one (Fawzi, 2011a; Progler, 2010). Rough estimates suggested that there are 15,000 addicted males to tramadol in Gaza (30% of males between 14 and 30 years old). even Israeli reports noted “some supplies of the drug are smuggled into Gaza from Egypt through tunnels, tramadol being the most used one (Fawzi, 2011b). Tramadol is currently available as tablets, oral drops, and solutions for injection. Tramadol is completely and rapidly absorbed after oral use (Shilo et al., 2008). Despite being a centrally acting analgesic agent, increasing the rate of opioids consumption especially tramadol is one of the major challenges in most countries. It is considered one of the most common causes of poisoning in people especially in adult male patients with a previous history of drug addiction and psychological problems which is considered the cause of mortality and morbidity in many countries (Mehrpour et al., 2015).

There are limited data available about its side effects and manifestations in overdose cases. The side effects of poisoning with the tramadol are drowsiness, nausea, vomiting, restlessness, headache, constipation, and some rare side effects related to the respiratory system, liver, and kidney (Subedi et al., 2019). Many medications especially morphine in overdose states affect

the body because of its toxicity and many of it affect thyroid hormones and testosterone (Caplan et al.,2007) The effect of long-term consumption of tramadol on endocrine systems especially the thyroid and testosterone hormones have not investigated so much (Katz & Mazer, 2009).

Some researches shows a direct effect of tramadol abuse with hormonal secretions, such as Thyroid hormones, Testosterone, LH and FSH which thus interferes with the menstrual cycle in women and decrease of the normal pulsatile release of LH leading to a negative effect on the testes of males caused by the decrease of testosterone levels (Mehrpour et al., 2015). Since Tramadol-Abusers are highly prevalent in the Gaza population (Diab et al., 2020) Assessment of the effect of thyroid hormones and testosterone among Tramadol-Abusers in such a population may help to determine the potential effects of Tramadol-Abusers metabolism (Tafesh, 2013). This study aimed to investigate the effect of tramadol abuse on the level of thyroid hormones and testosterone among tramadol-abusers in Gaza Strip.

Methods

The present study is a prospective case-control study, with inclusion criteria of known patients with Tramadol abuse, with age between 18-35 years, further a period of poisoning is more than three years. Patients who received other types of opioids, take hormone replacement therapy or corticosteroid therapy, and patients with a history of thyroid gland disease or fertility problems were excluded from this study.

The sample of the study consisted of 196 males as participants divided equally into two groups: (cases group) consisted of 98 with tramadol abuse problems, aged from 18 to 35 years old who followed before treatment (pre-detoxification) and one-month later post detoxification and an equal number of healthy individuals with matching age as control. The researcher conducted face-to-face interviews with the participants, which were used for filling the questionnaires designed for matching the study need of the study population. Most questions are designed as yes/no questions, which offers a dichotomous choice (Biner & Kidd, 1994). The questionnaire included questions on sociodemographic data (age, education, governorates, marital status, smoking, and employment status), also clinical data (route of poisoning, time of the poisoning, the dosage of poisoning, and clinical findings).

Blood samples (5 ml each) drawn into a vacationer blood collection tube from the study groups' cases and control. About 2 ml of blood was placed into an ethylene diamine tetraacetic acid (EDTA) vacutainer tube to perform complete blood count (CBC) for cases and controls. Serum samples were obtained to determine the biochemical parameters. CBC of cases and control was determined using Orphee for analysis depending on the electrical impedance principle. A complete system of reagents of control and calibrator according to standard protocols (Orphee for analysis, Mythic 18). The hormones serum samples included TSH, free T_4, free T3, and

Testosterone were measured quantitatively by ELISA technique (Enzyme-Linked Immune Sorbent Assay) using Biorex reagent KIT according to (Szaniawska & Spencer, 1995).

Treatment of addiction to tramadol goes through three consecutive stages; the first stage depended on the process of detoxification (withdrawal of tramadol), the second stage of psychological specialist, and the third stage of behavioral and cognitive therapy. The first stage included the period of withdrawal of the drug from the body, which spends a period of 7 days to 12 days according to the dose used in the past and the duration of the use of drugs. For most tramadol addiction, most cases can treat at home but in some cases, it may be necessary to enter the hospital to control the withdrawal symptoms. This phase treated suddenly with the patient giving some drugs that relieve withdrawal symptoms (such as drugs that help to sleep using antidepressants which called strong benzodiazepine such as Xanax, lorocare, or clonazepam) in addition to spasmolytic drugs also the medicines of the digestive system (McQuay et al., 2016). The second stage included the use of Antidot for opiates such as Naltrexone 50 mg orally. The third stage included encouragement and increased motivation to continue treatment and non-return of drugs. This stage is carried out by a psychiatrist or psychologist and the time after finishing directly from the phase of detoxification (McQuay et al., 2016).

Ethical and administrative considerations

The approval letter was obtained from the Helsinki Committee in the Gaza Strip-Palestine. For ethical consideration, the necessary approval to conduct this study was obtained from the Helsinki committee with ethical approval number PHRC/HC/115/16 in Gaza Strip. Also, an approval letter to conduct the study was obtained from the Ministry of Health (MOH). Before filling the questionnaire, assurance of voluntary participation was maintained, then the interviewer described the study aims, and explained all the questions for the participants. Informed consent was obtained from the participants with their signatures.

Statistical analysis

Data were computer analyzed using SPSS (Statistical Package for the Social Science version 20). The sample distribution of the study variables and the cross-tabulation were applied. Chi-square (X^2) was used to identify the significance of the relations associations, and interactions among various variables. The paired and student t-test, Pearson's correlation test, and correlation graphs plotting were applied. The results in all the above-mentioned procedures were accepted as statistically significant when the p-value was less than 5% (P<0.05).

Results

The study population comprised 196 participants divided equally into two groups: (case group) consisted of 98 with tramadol abuse problems, aged from 18 to 35 years old who followed before treatment (pre-detoxification) and one-month later post detoxification and equal number healthy individuals. The age was matched between cases and controls; with mean age 27.5±4.3 years in controls whereas that of chronic users of tramadol cases was 27.9±4.0 years (*P*>0.05). Table 1 shows the sociodemographic data among the study population. The number of controls from Gaza, Deir El-Balah, Khan Younis, and Rafah Governorates were 46 (46.9%), 3 (3.1%), 39 (39.8%), 10 (10.2%), respectively. In addition, the number of tramadol abusers from Gaza, Deir El-Balah, Khan Younis and Rafah Governorates were 28 (28.6%), 23 (23.5%), 30 (30.6%), 17 (17.3%), respectively, with statistically significant differences (χ^2 = 22.75, *P*< 0.001).

Analysis of the educational status of the controls and tramadol abusers showed that 37.8% and 71.4% have high education, 43.9% and 23.5% have finished secondary school, 18.3 %, and 5.1% have passed the primary school, respectively. The high educational level was associated with chronic users of tramadol (χ2 = 23.59, *P*< 0.001). Regarding employment status, 64.3% controls and 79.6% cases reported that they were employed compared to 35.7 controls and 20.4 cases who had not, respectively. Chi-square test showed employed in cases higher statistically significant than controls (χ2 = 5.69, *P*> 0.05). The numbers of single, married, widow and divorced in controls were 44 (44.9%), 45 (45.9%), 3 (3.1%), 6 (6.1%) and 49 (32.6%), respectively whereas in cases they were 15 (15.3%), 64 (65.3%), 10 (10.2%) and 9 (9.2%), respectively. There was a statistically significant association between different groups (χ^2=21.94, *P*< 0.001). The prevalence of smoking was higher statistically significant in cases than controls (92 (93.9%) *vs.* 82 (83.7%), respectively, χ^2 = 5.12 and *P*= 0.024.

Table 1:Baseline information among the study population (*n*=98)

Sociodemographic	Controls	Cases	Statistical test	*P*-value
Governorates n (%)				
Gaza	46 (46.9)	28 (28.6)	$\chi^2 = 22.75$	<0.001*
Deir El-Balah	3 (3.1)	23 (23.5)		
Khan Younis	39 (39.8)	30 (30.6)		
Rafah	10 (10.2)	17 (17.3)		
Education n (%)				
Higher education	37 (37.8)	70 (71.4)	$\chi^2 = 23.59$	<0.001*
Secondary school	43 (43.9)	23 (23.5)		
Primary school	18 (18.3)	5 (5.1)		
Employment Status n (%)				
Employed	63 (64.3)	78 (79.6)	$\chi^2 = 5.69$	0.017*
Unemployed	35 (35.7)	20 (20.4)		
Marital status n (%)				
Single	44 (44.9)	15 (15.3)	$\chi^2 = 21.94$	<0.001*
Married	45 (45.9)	64 (65.3)		
Widowed	3 (3.1)	10 (10.2)		
Divorced	6 (6.1)	9 (9.2)		
Smoking n (%)				
Smokers	82 (83.7)	92 (93.9)	$\chi^2 = 5.12$	0.024*
Non-smokers	16 (16.3)	6 (6.1)		
Age (years) Mean±SD (min-max)	27.5±4.3 (18-35)	27.9±4.0 (18-35)	t = 0.71	0.479

χ^2: chi-square test; **t:** Student's t-test;
**P*-value significant at $P \leq 0.05$.

Clinical data among tramadol abusers were presented in (Table 2) Rote of administration in all cases was by tablet. The average duration of using tramadol among cases was 5.7±1.8 years and a dose of tramadol was 3.8±2.2 tablets per day.

Table 2: Clinical data among cases (*n*=98)

Clinical data	Cases
Rote of administration n (%)	
Tablet	98 (100)
Oral drops	0 (0.0)
Injection	0 (0.0)
Period of Using (years)	
Mean±SD (min-max)	5.7±1.8 (4-10)
Dose (tablet/day)	
Mean±SD (min-max)	3.8±2.2 (1-17)

SD: standard deviation

Table 3 illustrate the mean values of hematological parameters among controls and tramadol abusers (pre-and post-treatment). The mean values of WBCs, Hb, RBCs, MCV, MCH, MCHC, RDW, and PLT demonstrate that there were no statistically significant differences between controls and pre-treatment and post-treatment of tramadol abusers showed by Student's t-test (*P*> 0.05). The results illustrate that there was no association between hematological parameters studied between the controls and tramadol abusers pre-and post-treatment.

Table 3: Haematological parameters among controls and tramadol abusers

Parameters	Control	Cases		% change	P-value
		Pre-treatment	**Post-treatment**		
WBCs (K/µL) (min-max)	6.9±1.6 (4.6-10.4)	7.0±2.0 (3.9-12.5)	7.2±2.1 (3.9-12.3)	1.44	0.721 [a]
				4.26	0.263 [b]
				2.82	0.225 [c]
RBC (M/µL) (min-max)	4.9±0.4 (4.3-6)	4.9±0.6 (3.4-6.2)	5.0±0.6 (3.8-6.6)	0.00	0.957 [a]
				2.04	0.425 [b]
				2.04	0.421 [c]
Hb (g/dl) (min-max)	14.6±0.9 (13.5-16.5)	14.3±1.5 (10.2-16.7)	14.5±1.4 (10.5-17.3)	-2.05	0.130 [a]
				-0.69	0.344 [b]
				1.39	0.187 [c]
HCT (%) (min-max)	42.8±2.8 (38.3-48.5)	42.4±3.9 (29.5-51.4)	42.7±3.4 (34.3-50.4)	-0.94	0.479 [a]
				-0.23	0.858 [b]
				0.71	0.485 [c]
MCV (fL) (min-max)	86.9±5.2 (76.4-96.4)	86.7±9.6 (62.9-105.5)	86.4±10.7 (67.5-110.8)	-0.23	0.856 [a]
				-0.58	0.658 [b]
				-0.35	0.790 [c]
MCH (pg) (min-max)	29.7±1.7 (26.5-32.6)	29.3±3.8 (20.8-38.2)	29.2±3.6 (23.4-39.8)	-1.36	0.386 [a]
				-1.70	0.202 [b]
				-0.34	0.730 [c]
MCHC (%) (min-max)	34.2±0.7 (33.0-36.0)	33.9±3.2 (27.1-43.8)	34±3.5 (26.2-43.7)	-0.88	0.430 [a]
				-0.59	0.564 [b]
				0.29	0.872 [c]
RDW (%) (min-max)	13.3±0.9 (11.3-14.6)	13.3±1.2 (10.4-15.8)	13.3±1.1 (10.5-15.7)	0.00	0.813 [a]
				0.00	0.590 [b]
				0.00	0.604 [c]
PLT (K/µL) (min-max)	238.6±62.1 (152-446)	241.4±71.5 (133-508)	238.7±57.6 (135-450)	1.17	0.771 [a]
				0.04	0.987 [b]
				-1.12	0.401 [c]

RBC=Red blood cell; HGB=hemoglobin; Hct=Hematocrit; MCV=mean corpuscular volume; MCH=mean corpuscular hemoglobin; MCHC=mean corpuscular hemoglobin concentration; RDW=red cell distribution width. All values were expressed as the mean ±SD. K/µL=thousand cells per microliter; M/µL= million cells per microliter.

[a, b] The significance of difference was checked by independent-sample T-test (compare all vs. control), significant at $P \leq 0.05$. [c] the difference in tramadol abusers were checked by paired-sample T-test (compare post vs. pre-treatment), significant at $P \leq 0.05$.

Table 4 and Figure 1 shows the mean values TSH, FT4, FT3, and testosterone among controls and tramadol abusers pre- & post-treatment. The mean levels of TSH were significantly increased (2.8±2.0 vs. 1.9±1.0 mIU/L, % of change = 47.4, $P < 0.001$, respectively) in tramadol abuser before treatment compared to control. In contrast, the mean levels of serum FT4, FT3, and testosterone were significantly decreased in tramadol abuser before treatment compared to control (1.1±0.3 vs. 1.4±0.3 ng/ml, 2.6±0.8 vs. 2.8±0.8 ng/ml, and 4.4±1.2 vs. 5.1±1.1 pg/ml, respectively $P \leq 0.05$).

The mean levels of FT4 were significantly higher in the post-treatment tramadol abusers group than control (1.5±0.5 vs. 1.4±0.3 ng/ml, % change= -7.14, $P \leq 0.05$). Meanwhile, there was no significant difference in the mean of TSH, FT3, and testosterone among post-treatment of tramadol abusers compared to control (1.9±1.0 vs. 2.0±1.0 mIU/L, 2.8±0.8 vs. 2.9±1.2 ng/ml, 5.1±1.1 vs. 4.9±1.5 pg/ml, $P > 0.05$, respectively). Clearly, paired t-test illustrated that the mean levels of TSH in tramadol abuser post-treatment were significantly decreased compared to tramadol abuser pre-treatment (2.0±1.0 vs. 2.8±2.0 mIU/L, % change = -28.6, $P = 0.036$, respectively). While, the mean levels of FT4, FT3, and testosterone for tramadol abuser post-treatment were significantly increased compared to tramadol abuser pre-treatment with percent of change 36.37%, 11.54%, and 11.36%, respectively.

Table 4: Serum levels of TSH, FT4, FT3, and testosterone among controls and tramadol abusers (pre & post-treatment) (*n*=98)

Hormones	Controls	tramadol abusers		% change	P-value
		Pre-treatment	Post-treatment		
TSH (mIU/L) (min-max)	1.9±1.0 (0.5-4.2)	2.8±2.0 (0.6-9.5)	2.0±1.0 (0.4-5.0)	47.4	0.001 [a *]
				5.3	0.396 [b]
				-28.6	0.036 [c *]
FT4 (ng/ml) (min-max)	1.4±0.3 (0.8-1.8)	1.1±0.3 (0.1-1.9)	1.5±0.5 (0.3-3.0)	-21.4	0.001 [a *]
				7.14	0.003 [b*]
				36.37	0.001 [c *]
FT3 (ng/ml) (min-max)	2.8±0.8 (1.8-4.3)	2.6±0.8 (0.8-4.4)	2.9±1.2 (0.8-5.8)	-7.14	0.026 [a *]
				3.57	0.617 [b]
				11.54	0.04 [c *]
Testosterone (pg/ml) (min-max)	5.1±1.1 (3.4-8.5)	4.4±1.2 (1.9-7.5)	4.9±1.5 (1.9-9.0)	-13.72	0.001 [a *]
				-3.92	0.346 [b]
				11.36	0.001 [c *]

SD: standard deviation; **TSH:** thyroid-stimulating hormone; **FT4:** Free thyroxine; **FT3:** free triiodothyronine. All values were expressed as the mean ±SD

[a,b] The significance of difference was checked by the independent-sample T-test (compare all vs. control), significant at $P \leq 0.05$. [c] The difference in tramadol abusers were checked by paired-sample T-test (compare post vs. pre-treatment), significant at $P \leq 0.05$.

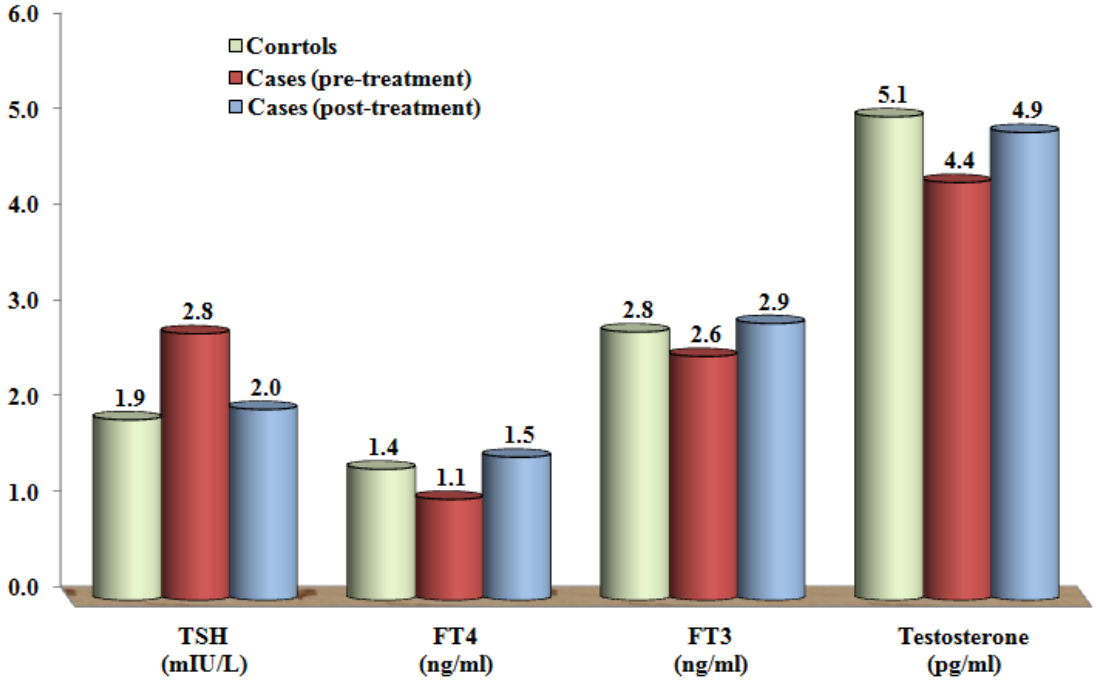

Figure 1: Serum levels of TSH, FT4, FT3, and testosterone among controls and tramadol abusers (pre & post-treatment).

The correlation between hormonal and haematological parameters studied in the study population in Table 5. Pearson correlation showed a significant positive correlation between TSH levels and PLT counts in the study population (r = 0.169, *P* = 0.018; Fig. 2). On the other hand, no significant correlation was found between TSH levels and WBCs (r= 0.033, *P*= 0.645), RBC (r= 0.022, *P*= 0.763), Hb (r= -0.017, *P*= 0.815), HCT (r= -0.012, *P*= 0.867), MCV (r= -0.049, *P*= 0.493), MCH (r= -0.054, *P*= 0.449), MCHC (r= 0.008, *P*= 0.911); and RDW (r= 0.028, *P*= 0.699). Obviously, Pearson correlation of the present study showed positive significant correlations between FT4 levels and Hb concentrations (r= 0.150, *P*= 0.036; Fig. 2). In comparison, no significant relation between FT4 levels and WBCs (r= 0.11, *P*= 0.125), RBC (r= 0.135, *P*= 0.059), HCT (r= 0.136, *P*= 0.057), MCV (r= -0.054, *P*= 0.451), MCH (r= -0.021, *P*= 0.768), MCHC (r= 0.023, *P*= 0.751), RDW (r= 0.094, *P*= 0.19); and PLT (r= -0.048, *P*= 0.506). As can be expected, there was no significant correlation between FT3 levels and hematological parameters studied; WBCs (r= -0.063, *P*= 0.38), RBC (r= -0.024, *P*= 0.743), Hb (r= 0.11, *P*= 0.124), HCT (r= 0.073, *P*= 0.312), MCV (r= 0.097, *P*= 0.178), MCH (r= 0.123, *P*= 0.086), MCHC (r= 0.051, *P*= 0.48), RDW (r= 0.105, *P*= 0.144) and PLT (r= -0.022, *P*= 0.755). After all, testosterone levels revealed a significant positive correlation with RDW (r= 0.164, *P*= 0.021; Fig. 2), nevertheless there is no significant correlation in testosterone levels with WBCs (r= 0.007, *P*= 0.923), RBC (r= 0.008, *P*= 0.914), Hb (r= 0.114, *P*= 0.112), HCT (r= 0.031, *P*= 0.671), MCV (r= 0.019, *P*= 0.791), MCH (r= 0.095, *P*= 0.187), MCHC (r= 0.098, *P*= 0.171), and PLT

(r= -0.07, *P*= 0.329). In brief, the present study showed no significant correlation between hormonal and hematological parameters studied except positive significant correlation between TSH levels & PLT counts, FT4 levels & Hb concentrations; and testosterone levels & RDW.

Table 5: The correlation between hormonal and haematological parameters among the study population.

Parameters	**TSH (mIU/L)**		**FT4 (ng/ml)**		**FT3 (ng/ml)**		**Testosterone (pg/ml)**	
	r	***P*-value**	**r**	***P*-value**	**r**	**P-value**	**r**	*P*-value
WBCs (K/µL)	0.033	0.645	0.110	0.125	-0.063	0.380	0.007	0.923
RBC (M/µL)	0.022	0.763	0.135	0.059	-0.024	0.743	0.008	0.914
Hb (g/dl)	-0.017	0.815	0.150	**0.036***	0.110	0.124	0.114	0.112
HCT (%)	-0.012	0.867	0.136	0.057	0.073	0.312	0.031	0.671
MCV (fL)	-0.049	0.493	-0.054	0.451	0.097	0.178	0.019	0.791
MCH (pg)	-0.054	0.449	-0.021	0.768	0.123	0.086	0.095	0.187
MCHC (%)	0.008	0.911	0.023	0.751	0.051	0.480	0.098	0.171
RDW (%)	0.028	0.699	0.094	0.190	0.105	0.144	0.164	0.021*
PLT (K/µL)	0.169	**0.018***	-0.048	0.506	-0.022	0.755	-0.070	0.329

r: Pearson correlation; *P-value significant at *P*≤ 0.05.

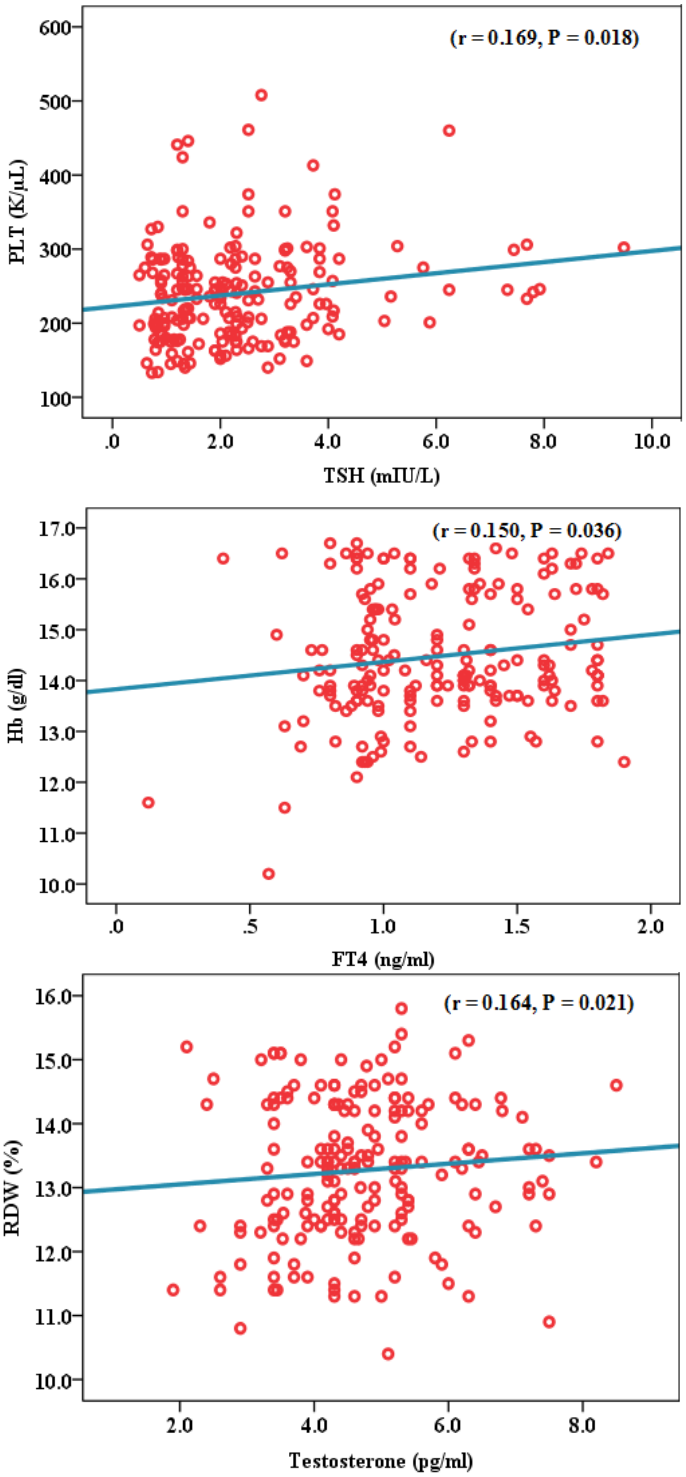

Figure 2: The correlation between hormonal and haematological parameters studied in the study population between a) TSH and PLT; b) FT4 and Hb; and c) Testosterone and RDW

The correlation between TSH, FT4, FT3, and testosterone are pointed out in Table 6. Serum TSH levels exhibited negative correlation with FT4 (r = -0.375, *P*< 0.001; Fig. 3); FT3 (r= -0.309, *P*< 0.001; Fig. 3) and Testosterone (r = -0.455, *P*< 0.001; Figure 3). On the contrary, serum FT4 levels showed positive correlation with FT3 (r = 0.502, *P*<0.001; Fig. 3) and testosterone (r = 0.610, *P*<0.001; Fig. 3). Likewise, there was a positive correlation between FT3 and testosterone (r= 0.713, *P*<0.001; Fig. 3). Summarily, a negative significant association was found between serum TSH and other studied hormones (FT4, FT3, and testosterone). In contrast, FT4 was positive relations between FT3 and testosterone. Also, there were positive relations between FT3and testosterone.

Table 6: The correlation between studied hormones in the study population

Hormones	**TSH (mIU/L)**		**FT4 (ng/ml)**		**FT3 (ng/ml)**		**Testosterone (pg/ml)**	
	r	***P*-value**	**r**	***P*-value**	**r**	***P*-value**	**r**	*P*-value
TSH (mIU/L)	-	-	-0.375	<0.001*	-0.309	<0.001*	-0.455	<0.001*
FT4 (ng/ml)	-0.375	<0.001*	-	-	0.502	<0.001*	0.610	<0.001*
FT3 (ng/ml)	-0.309	<0.001*	0.502	<0.001*	-	-	0.713	<0.001*
Testosterone (pg/ml)	-0.455	<0.001*	0.610	<0.001*	0.713	<0.001*	-	-

r: Pearson correlation *P-value significant at *P*≤ 0.05.

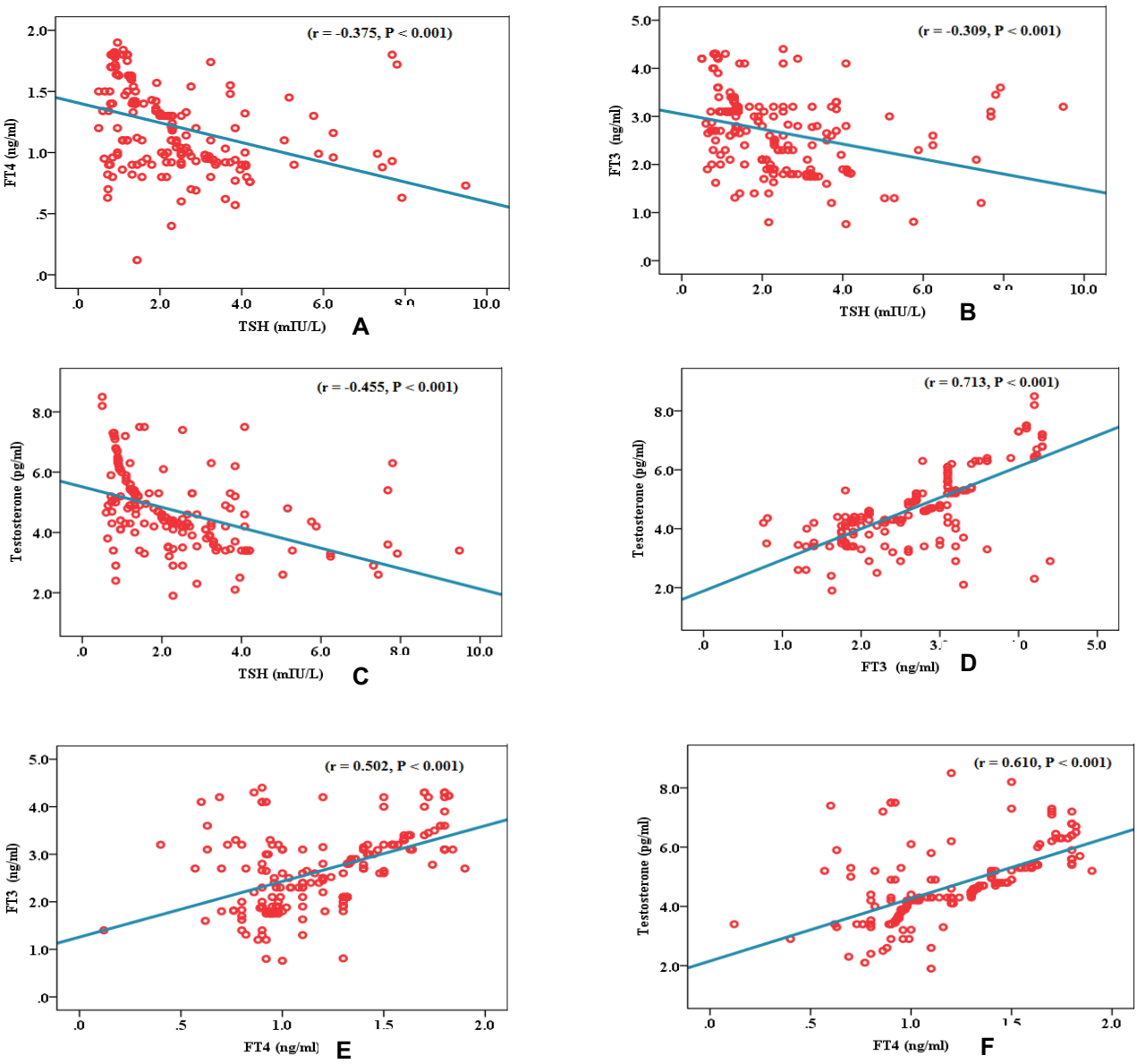

Figure 3: The correlation between TSH, Thyroid hormones, and Testosterone among the study population
(a): The correlation between TSH and FT4 among the study population
(b): The negative correlation between TSH and FT3 among the study population
(c): The negative correlation between TSH and testosterone among the study population
(d): The positive correlation between FT4 and FT3 among the study population
(e): The positive correlation between FT4 and testosterone among the study population
(f): The positive correlation between FT3 and testosterone among the study population

Table 7 illustrated the relation between dose of poisoning and studied parameters (hematological and hormonal). Pearson correlation test revealed negative significant correlations between dose of poisoning and FT4 (r= -0.218, *P*= 0.031) and FT3 (r= -0.211, *P*= 0.037). However, there was no significant correlation between dose of poisoning and others parameters (WBCs (r= -0.042, *P*= 0.68), RBC (r= -0.021, *P*= 0.841), Hb (r= -0.176, *P*= 0.083), HCT (r= -0.133, *P*= 0.193), MCV (r= -0.093, *P*= 0.364), MCH (r= -0.137, *P*= 0.18), MCHC (r= -0.07, *P*= 0.492), RDW (r=-0.035, *P*= 0.730), PLT (r= -0.008, *P*= 0.935), TSH (r= -0.027, *P*= 0.794) and Testosterone (r= -0.136, *P*= 0.182)). Obviously, duration of poisoning was no significant correlation with either hematological or hormonal parameters in the study parameters (r= 0.011, *P*= 0.913 for WBCs; r= 0.1, *P*= 0.325 for RBC; r= 0.056, *P*= 0.585 for Hb; r= 0.063, *P*= 0.54 for HCT; r= -0.055, *P*= 0.593 for MCV; r= -0.052, *P*= 0.610 for MCH; r= 0.001, *P*= 0.994 for MCHC; r= -0.106, *P*= 0.298 for RDW; r= 0.035, *P*= 0.732 for PLT , r= 0.035, *P*= 0.735 for TSH; r= 0.118, *P*= 0.247 for FT4; r= -0.093, *P*= 0.364 for FT3; and r=-0.142, *P*= 0.163 for testosterone). Therefore, neither hematological nor hormonal affected by duration of poisoning.

Table 7: The relationships of poisoning duration and dose with studied parameters

Parameters	**Dose of poisoning (tablet/day)**		**Duration of poisoning (years)**	
	r	**P-value**	**r**	**P-value**
WBCs (K/µL)	-0.042	0.680	0.011	0.913
RBC (M/µL)	-0.021	0.841	0.100	0.325
Hb (g/dl)	-0.176	0.083	0.056	0.585
HCT (%)	-0.133	0.193	0.063	0.540
MCV (FL)	-0.093	0.364	-0.055	0.593
MCH (pg)	-0.137	0.180	-0.052	0.610
MCHC (%)	-0.070	0.492	0.001	0.994
RDW (%)	-0.035	0.730	-0.106	0.298
PLT (K/µL)	-0.008	0.935	0.035	0.732
TSH (mIU/L)	-0.027	0.794	0.035	0.735
FT4 (ng/ml)	-0.218	**0.031***	0.118	0.247
FT3 (ng/ml)	-0.211	**0.037***	-0.093	0.364
Testosterone (pg/ml)	-0.136	0.182	-0.142	0.163

r: Pearson correlation; *P-value significant at *P*≤ 0.05.

Table 8 illustrated the hematological profile correlation between studied tramadol abusers (Post- and pre-treatment). Pearson correlation showed positive significant correlation between pre- and post-treatment of tramadol abusers in WBCs (r = 0.661, *P*< 0.001), RBCs (r = 0.391, *P*< 0.001), HGB (r = 0.848, *P*< 0.001), HCT (r = 0.486, *P*< 0.001), MCV (r = 0.271, *P*= 0.007), MCH (r = 0.315, *P*= 0.002), MCHC (r= 0.483, *P*< 0.001), RDW (r = 0.753, *P*< 0.001) and PLT (r = 0.906, *P*<0.001) **Fig. 11.** Meanwhile, there is no significant correlation between controls and tramadol abusers groups (pre & post-treatment) in hematological profile (*P*>0.05) except in RDW that showed positive significant correlation between controls and tramadol abusers groups in both pre & post treatment (*P*<0.05).

Table 8: The correlation between hematological profiles among the study population

Parameters	Post& Pre-treatment		Pre-treatment & controls		Post-treatment & controls	
	r	P-value	r	P-value	r	P-value
WBCs (K/µL)	0.661	**<0.001***	-0.140	0.169	-0.125	0.221
RBC (M/µL)	0.391	**<0.001***	-0.071	0.487	0.014	0.888
Hb (g/dl)	0.848	**<0.001***	-0.155	0.127	-0.103	0.312
HCT (%)	0.486	**<0.001***	-0.032	0.756	-0.075	0.461
MCV (fL)	0.271	**0.007**	-0.106	0.297	0.023	0.822
MCH (pg)	0.315	**0.002**	-0.059	0.566	0.103	0.314
MCHC (%)	0.483	**<0.001***	-0.003	0.978	-0.101	0.324
RDW (%)	0.753	**<0.001***	0.527	**<0.001***	0.748	<0.001*
PLT (K/µL)	0.906	<0.001*	0.112	0.270	0.148	0.147

r: Pearson correlation; *P-value significant at *P*≤ 0.05.

Discussion

Tramadol as being one of the opioids which are widely used for the treatment of different degree of pain disorders (Ibrahim, 2018)**,** it is considered the most commonly prescribed opioids in the world (Fischer et al., 2019; Rodieux et al., 2018), but by a long term of tramadol abusing it became the most common causes of drug poisoning in people leading to psychological problems which are considered the causes of mortality and morbidity in many countries (Kunzler et al., 2018). Many medications that belong to opioids especially morphine in overdose states affect the body because of its toxicity and many of its effects on thyroid hormones and testosterone (Sikka & Bartolome, 2018). It is noteworthy that tramadol has been being one of the most common scenes of drug poisoning due to drug abuse, notably among adult males worldwide (Bamigbade & Langford, 1998), as well in the Gaza Strip, and elsewhere in the middle east. When overdosed, it is, as believed, associated with significant morbidity and mortality (Elmanama et al., 2015).

The present study demonstrated that there was an association between some socio-demographic characteristics and tramadol abusing which included age, smoking, the Governorates, employment status, and marital status. This indicates that the socio-demographic characteristics affected results as confiding. There was a matching between tramadol abusing and controls in most of these characteristics. In our results, there was an association between marital status and tramadol abusing and that may be due to the use of tramadol as a way for treating premature ejaculation (Abdel-Hamid et al., 2016). Also, in the present study, tramadol abusers' data showed that about (71.4%) of the abusers were a high educational level this percentage is critical especially with the difficult socioeconomic status along with a life of hardship and unemployment.

The results showed that the Gaza governorate had a high percentage of tramadol abusing this may be due to area or patient selection also this study demonstrated that the route of ingestion of tramadol was the oral route of intoxication; this is probably because tablets are widely available in Gaza Strip, cheapest and easiest to use. Dispersion of injectable forms of tramadol is limited to non-private or private pharmacies and hospitals (Leppert, 2009). This is in agreement with a study performed in Iran, showing that the majority of people seeking tramadol from pharmacies are young adults taking tramadol orally, with the criteria for drug addiction (Zabihi et al., 2011). Most patients were addicted to a tramadol dose of more than 500 mg using opioid overdose according to previously published data that considered the total daily dose should not exceed 400mg for adult therapeutic blood levels of 0.1-0.8 mg/L (Clarot et al., 2003).

To the best of our knowledge, there are no studies aimed to assess the hematological parameters for follow up of tramadol administration, neither other opioids, until the date of the writing of this manuscript. It was found that the drug abuse exhibited no significant observations in the mean level of WBCs, RBCs, Hb, and HCT MCV, MCH, MCHC, and PLT parameters. This is probably explained as the drug has no side effect on either hemopoiesis or iron metabolism by any means.

Our current results showed that the mean levels of TSH were significantly higher among tramadol abusers at baseline compared to control ($P \leq 0.05$) also it is demonstrated that TSH after treatment was significantly lower than before treatment. This study is the first to test thyroid hormones on tramadol abusers in Gaza Strip, and the results show similarity to other studies at the TSH levels due to reduction of thyroid function, which in turn led to decreased FT3 and FT4, because of the known consequences of Mu opioid peptide (MOP) receptor agonists, which affects the neuroendocrine system causing reduced hypothalamic-pituitary-gonadal axis (Rhodin et al., 2010). From other previous studies, the administration of opioids showed an altered hypothalamus-thyroid axis similar results were documented (Gozashti et al., 2014).

The mean levels of free T_4 and free T_3 were significantly lower among tramadol abusers compared to control at baseline. While after detoxification treatment the mean levels were significantly higher compared to the baseline of the study. When studying thyroid hormones, there were significant alterations in the thyroid axis. FT3 and FT4 were lowered, whereas TSH was increased in addicted patients compared to their pretreatment results and control cases. From other previous studies, the administration of opioids showed an altered hypothalamus-thyroid axis (Gozashti et al., 2014). Studies on morphine and heroin showed lowered hypothalamus-thyroid axis activity (Gozashti et al., 2014). A recent study on opium showed different findings, for instance, FT3 and Total T3 were decreased, whereas FT4 and Total T4 were increased and so TSH (Gozashti et al., 2014).

Studying testosterone showed significantly lowered levels of the hormone among drug abusers compared to their pretreatment results and the control group (Table 6). This finding in agreement with (Ahmed & Kurkar, 2014); (Abdellatief et al., 2015) who found significantly reduced plasma levels of testosterone in albino adult male rats which were subcutaneously injected with 40 mg/kg of tramadol, three times per week for 8 weeks compared with the control group.
These results were somehow predicted at the time other studies on opioids showed similar results (Handa et al., 1994), which also can be explained as a result of the reduction of the hypothalamic-pituitary-gonadal axis.

Conclusion: Finally, we conclude that serum hormonal levels of TSH, FT4, FT3, and testosterone exhibited a positive correlation between tramadol abuser's pre-detoxification and post detoxification, where the level of testosterone in pre detoxification of tramadol abusers was significantly lower than controls and was significantly lower than its results after detoxification. There is no significant correlation between controls and tramadol abusers' groups (pre & post-treatment) in hematological profile except in RDW that showed a positive significant correlation between controls and tramadol abusers' groups in both pre & post-treatment. There was no significant correlation between the duration of poisoning and hematological or hormonal parameters in the study parameters. The hormones level including (TSH, Testosterone, FT4, and FT4) should be testing as monitoring for tramadol abusers. More research concerning other hormone tests may be performed to increase awareness for people about the toxic effects of tramadol.
Our study has some limitations: First, sample collection was relatively difficult as many patients refuse to participate in the study and difficulty obtaining samples from females due to the very limited of female Tramadol abusers. Second, the delay of performing some hormonal and biochemical parameters into some days due to unavailability of kits reagents. Third, the delay of some samples due to electricity off. And four, the lack of relevant studies made the comparison somewhat difficult.

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Conflicts of Interest:
The authors declare no conflicts of interest.

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