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Original Article

Variations in Gonadal Steroids in Workers Occupationally Exposed to Toxicants at Automobile Workshops and Petrol Filling Stations

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ABSTRACT

The most at risk for occupational toxicity brought on by exposure to heavy metals and PAHs among various vocations are gas station attendants and auto workers. The gonadal and its regulating hormonal pattern were identified in the current investigation in gas station attendants and car employees. **Objective:** This study's goal is to ascertain the impact of various occupational toxicants on the ovarian health of gasoline station attendants (PPA) and car technicians (AMM). Gonadal steroids and the hormones that control them were examined for this reason and their relationship to gonadal function was established. **Methods:** For this, blood samples from 19 gas station attendants and 29 auto mechanics were obtained from various gas stations and car shops, respectively. The University of the Punjab in Lahore provided the blood samples for the 24 controls. Using commercially available ELISA kits, the levels of serum estradiol, follicle stimulating hormone (FSH), luteinizing hormone (LH), and testosterone were examined. The significance of changes was evaluated using the one-way ANOVA test. **Results:** When compared to the control group, there was a little decrease in the levels of estradiol, follicle-stimulating hormone, luteinizing hormone, and testosterone among fuel station attendants and car employees. **Conclusions:** Pertinently, reduced reproductive and their regulatory hormonal levels predispose future risk of manifesting reproductive health issues.

INTRODUCTION

Occupational environments are major source of different varieties of harmful chemical substances. In Pakistan due to poor economic and health conditions, a number of uneducated people are forced to do work in these hazardous working environments. Due to the lack of awareness, these people are not familiar of basic health conditions. Chemicals are omnipresent anthropogenic pollutants they are poisonous, carcinogenic, and can induce mutations in all organisms, including humans [1]. Automobile workers belong to such occupational section of the society which are most likely to get harmed by chronic toxicity of lead and the reason behind that is their daily working processes which include motor vehicle spray painting, assembly, welding, brazing and processing of radiators. The toxicity of lead become a community problem when the family members especially children get harmed by indirect method and source of this indirect toxicity is uniforms or clothes of workers which they use at their occupational sites [2]. Polycyclic aromatic hydrocarbons (PAHs) may covalently attach with proteins and deoxyribonucleic acid (DNA), which results in biochemical disturbance and cell damage in various animals and cause cancer in human. The main source of these harmful pollutants in the environment includes forest fire, petroleum leakage, burning of oil and coal. These pollutants have adverse effects on male reproductive system [3]. Low antioxidant capacity and free radical genesis, in occupationally exposed workers,

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evidences an early biochemical sign of a deranged metabolic state [4]. Gonadal functions are controlled via feedback loops which involves hypothalamic periodic pulses of gonadotropin releasing hormone (GnRH) to the adenohypophysial cells which in turn synthesize luteinizing hormone (LH) and follicle stimulating hormone (FSH) [5]. LH acts on the Leydig cells where they start testosterone synthesis, and FSH which stimulates sperm production [6]. Unusual synthesis of prolactin by pituitary tumors can effect and depress the production of both LH and FSH resulting in gradual decrease in testosterone formation in the testes [7]. Infertility is an increasing problem all over the globe, affecting 8–15% of couples in reproductive life [8]. Impotency in men is manifested by different lethal factors which encompasses, testicular blockage, metabolic alterations, and environmental toxins [9,10]. Occupational and unintentional exposure to chemicals, use of alcohol, drugs, and use of androgenic steroids are capable of exerting deep oppressive effect on the formation of sperm and androgens by the testes [11]. Estimation of reproductive hormones and oxidative indices interaction in serum of males occupationally exposed to chemicals is important in envisaging those who may develop serious disease together with infertility [12]. According to current research, oxidative stress is caused by an imbalance between the levels of the antioxidants peroxidative and antioxidative in plasma. As a consequence, the metabolic and functional problems of the male reproductive cells are reduced in many types of infertility [13,14]. This study's goal is to ascertain the impact of various occupational toxicants on the ovarian health of gasoline station attendants (PPA) and car technicians (AMM). Gonadal steroids and the hormones that control them were examined for this reason and their relationship to gonadal function was established.

METHODS

Male attendants and mechanics' blood samples were gathered from several Lahore gas stations and vehicle repair businesses, respectively. Healthy controls were sampled from the University of the Punjab's Quaid-e-Azam Campus with age and sex matching. A comprehensive Proforma was prepared to know the entire medical history of participants and their demographic features. After giving detailed explanation to every participant about the aim and purpose of research work, written consent was taken by them. Questionnaire was filled by each participant individually before taking the blood sample from them. Demographic data included systolic and diastolic blood pressure, history of smoking, age, gender, weight, height, any kind of drug addiction, medication, HBV, HCV, HIV screening and any past ailment history. Inclusion criteria setup for the petrol pump workers and automobile mechanics was at least, more than six months (6-8 hours daily) of exposure in the hazardous occupational environments having dust and exhaust fumes, petroleum vapors and aromatic hydrocarbons. Total forty-eight male exposed subjects (Age; 17-60 years) were recruited in this research work. Nineteen were petrol pump attendants, twenty-nine were automobile mechanics or workers and twenty-four were healthy control individuals, selected from general population. Blood samples of both workers and control samples were analyzed for their LH FSH, estradiol and testosterone level by using ELISA kits. Subjects whose sampling was done were categorized in two groups as follow:

Control group Healthy Males
Group I Petrol Pumps attendants (PPA)
Group II Auto-Mobile Mechanics (AMM

RESULTS

Comprehensive presentation of studied parameters in all studies groups are depicted in Table I.

Parameters	Controls (n=24)	Petrol Pump Attendants (n=19)	AMM (n=29)	P-value
Estradiol (pg/mL)	32.63 ± 2.37	27.76 ± 2.77	31.15 ± 1.98	0.3
FSH(mIU/mL)	5.26 ± 0.64	5.19 ± 0.65	4.80 ± 0.75	0.8
LH(mIU/mL)	4.59 ± 0.58	3.38 ± 0.41	3.38 ± 0.41	0.1
Testosterone (ng/mL)	4.91 ± 0.44	4.43 ± 0.52	4.43 ± 0.43	0.7

Table 1: Levels of Gonadal and its regulatory hormones in comparable groups. Values are mean ± SEM.

Parameters	Control vs Petrol Pump Attendants	Control vs Automobile Workers	Petrol Pump Attendants vs Automobile Workers
Estradiol (pg/mL)	14.92↓	4.53 ↓	12.21↑
FSH(mIU/mL)	1.33 ↓	8.74 ↓	7.51 ↓
LH(mIU/mL)	26.36↓	26.36↓	-
Testosterone (ng/mL)	9.77 ↓	9.77 ↓	-

Table 2: Presenting percentage difference of comparable groups

Non-significant decrease of 14.92 % was evidenced in petrol pump attendants when compared to control. In control vs automobile worker's comparison mild decrease of 4.53 % was evidenced in automobile workers. Additionally, mild elevation of 12.21 % of Estradiol was present in automobile mechanics when compared with petrol pump attendants as shown in figure 1 and table 2.

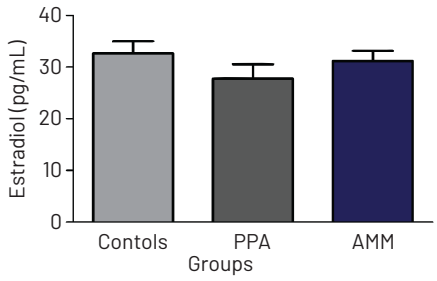

Figure1: Comparison of Estradiol (pg/mL) in studied groups.

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Values are mean ± SEM

Non-significant decrease of 1.33% was evidenced in FSH levels in PPA when compared to controls. Controls vs AMM comparison demonstrated non-significant decrease of 8.74% in FSH levels as compared to controls. Moreover, mild decrease of 7.51% was observed in automobile mechanics when compared to PPA as shown in Figure 2, and Table 2.

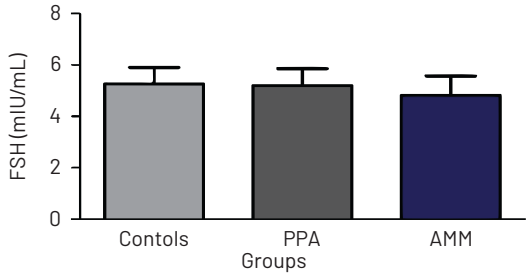

Figure 2: Comparison of Serum Follicle Stimulating Hormone (mIU/mL) in comparable groups. Values are mean ± SEM.

Levels of LH demonstrated non-significant decline of 26.36% in petrol pump attendants when compared to controls. A non-significant decrease of 26.36% was evidenced in automobile mechanics as compared to controls. Additionally, no difference was found in automobile mechanics when compared with petrol pump attendants as shown in Figure 3 and Table 2.

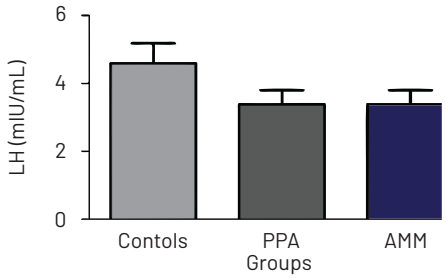

Figure 3: Comparison of Luteinizing Hormone (mIU/mL) in studied groups. Values are mean ± SEM.

There was a non-significant decrease of 9.77% in testosterone levels in both petrol pump attendants and automobile workers as compared to controls. Moreover, no difference was evidenced in automobile mechanics when compared to petrol pump attendants as shown in Figure. 4 and Table 2.

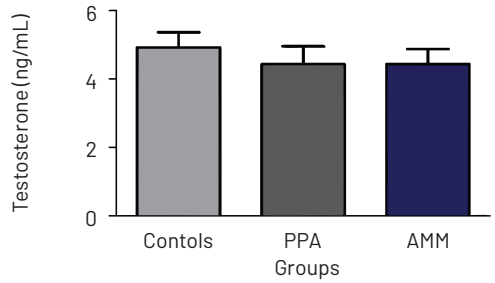

Figure 4: Comparison of Serum Testosterone (ng/mL) in studied groups. Values are mean ± SEM.

DISCUSSION

The present study was designed to determine the effect of different occupational toxicants on the gonadal health of petrol pumps attendants and automobile mechanics. Estrogens produced in testis interacts with estrogen receptors (ER), perpetuating the initiation of transcription of specific genes. Estrogen receptors (ERα and ERβ) are present in most of the cells of testis and in some other parts of the genital tract. Hence, the role of estrogens in physiology of male reproduction is of great concern [15-19]. A non-significant decrease in Estradiol concentration is observed among the members of exposed groups (Both petrol pump attendants and automobile workers). Follicle-stimulating hormone (FSH) plays an important role in reproduction of mammals. It stimulates testicular and ovarian functions through a G-protein-coupled receptor on the surface of target cells. In females, FSH induces the maturation of ovarian follicles by targeting a FSH receptor (FSHR) expressed only on granulosa cells. In males, FSH supports spermatogenesis and also stimulates sertoli cell proliferation in testes. FSH clinically used in treatment of infertile men and ovulatory women [20,21]. In our study, mild decrease in FSH concentrations was observed among the individuals of exposed groups (Both petrol pump attendants and automobile workers). Decreased level of FSH may results in number of ailments like azoospermia, oligospermia and infertility. Azoospermia is defined as a "disease in which there is absence of sperm in minimum two different samples ejaculated by same subject (including the centrifuged sediment) [22]. In the general population, 10 to 15% of couples suffer from infertility issues [23]. Of these infertile males, 10 to 20% (or 1% of all men in the general population) suffer from azoospermia (24). Detailed history, hormone profile, physical examination, genetic predisposition imaging play important role to conclude the classification of the azoospermia clinically [22,25]. Luteinizing hormone (LH) is produced in all classes of vertebrates (fishes to mammals). Basophilic cells known as gonadotrophs in the anterior pituitary gland produced and stored LH. In males, LH targets the interstitial cells (Leydig cells) present in testis, which results in production of androgens. Additionally, secondary function of the LH is to promote spermatogenesis through androgens [26]. A non-significant decrease in LH concentrations is observed among the members of exposed groups (Both petrol pump attendants and automobile workers). Decrease in LH results in the lower secretion of sex steroids, failure of ovulation and luteinization and atrophy of interstitial cells, whereas, excessive secretion of LH results in hyperplasia of testicular cells (interstitial) which is followed by atrophy, increased secretion of estrogen or androgen, super-ovulation, and accelerated sexual maturation. Low blood LH

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level causes different human diseases like craniopharyngioma and adrenogenital syndrome [27]. The testis secret male sex hormones (dihydrotestosterone, androstenedione and testosterone) which are collectively referred as androgens, which include. Testosterone is abundant as compare to all other hormones, which made it more significant. Testosterone changes into more active hormone dihydrotestoterone in the target tissues. Testosterone is produced by interstitial cells of Leydig, present in the interstices of seminiferous tubules which make up 20% of the mass of the adult testes. Leydig cells are almost absent in the childhood and testes almost produce no testosterone at that time, but it is abundant in newly born male infants for first few months of life and in adult's male at any time after puberty, in both these time testes secrete large amount of testosterone [28]. In this study, non-significant decrease in Testosterone concentrations is observed among the members of exposed groups (Both petrol pump attendants and automobile workers). Decrease in the testosterone level can cause some serious problem like infertility, oligospermia and erectile dysfunction (ED). Wide interest has been showed in de ciency of testosterone in men with ED. Some physiologists favor the determination of level of testosterone only under certain circumstances for example when there is bilateral testicular atrophy or a decrease in libido [29]. Recently it was shown that a permissive role is played by in erectile function. The functioning of nitric oxide synthase relays on sufficient levels of androgen, and deficiency of androgens might affect the functioning of gene i.e. phosphodiesterase type-5(PDE-5)[30]. Recently it was reported that some patients of hypogonadism and ED might respond to androgenic supplements but they do not respond to phosphodiesterase type-5 (PDE-5) inhibitors [31]. Therefore, clarification is required on some points but it seems clear that in erectile mechanism testosterone plays important role. Low level of testosterone can cause decreased bone mineral density and muscle mass, central obesity, increased fat mass, decreased energy and libido, insulin resistance, dysphoria and irritability [32]. The occurrence of clinical deficiency of androgen (low testosterone levels and symptoms) was reported recently to be nearly 6%to12% in elderly and middle-aged men [33]. Testes are one of the complex organ in mammals which are characterized by two major functions: production of spermatozoa and synthesis of steroid hormones. It is familiar fact that maintenance of spermatogenesis and normal testicular development are controlled by gonadotrophins and testosterone whose effects are transformed by factors which are locally-produced, and among them estrogens are noticeably concerned [34]. The reproductive tract of the male has high levels of estrogens as compare to general blood compartment [35], therefore it favors the fact that testis is the source of estrogens [36]. Hypogonadotropic hypogonadism is described by low FSH serum levels in relationship with generally, low LH levels and low serum testosterone levels. According to study in University of Illinois, almost half of men who have suffered from non-obstructive azospermia (NOA) also suffer from hypogonadotropic hypogonadism. This result shows that hypogonadotropic hypogonadism may be significantly more common in the infertile males than was formerly believed. Genetic hypothalamic diseases, for instance Kallmann syndrome, and acquired pituitary deficiencies or congenital, such as pituitary tumors (functional or nonfunctional) or empty sella syndrome, can result hypogonadotropic hypogonadism. In men who suffered from azoospermia with decreased libido, gynecomastia, anosmia, visual field deficits or headaches, should be suspected for hypogonadotropic hypogonadism [37].

CONCLUSIONS

It is concluded that petrol pump attendants and automobile workers are being affected by occupational toxicity, although a non-significant decrease is observed in this study but chronic exposure to such harmful toxicants may lead towards a prominent decrease in gondal and their regulatory hormones concentration, which may cause infertility in them. Chronic exposure to occupational toxicants may also cause some other medical problems like dysfunction of kidney, liver and other major systems of the body like nervous, reproductive and endocrine. Therefore, it is recommended that they should take some precautionary measures to save themselves from such harmful toxicants. The use of facial masks, while, working in such sites and proper cleaning of the occupational sites can also prove helpful in this regard.

Conflicts of Interest

The authors declare no conflict of interest.

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