Effect of Tobacco Alkaloids on the Endocrine System

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Alkaloids in tobacco and cigarette smoke causes malfunction of the endocrine system. Under certain conditions, smoking may be a risk factor for the development of hormone dependent pathology. Researching the relationship between smoking and sex steroid hormone status was based on the investigation of serum androgens and estrogens in men, smokers and nonsmokers. Comparative determinations were made in men without apparent endocrine pathology, on the one hand and, on the other hand, in patients with prostatic pathology characterized by impaired metabolism of sex steroid hormones. The results of this study could contribute to develop effective ways to quit smoking or to relieve the negative effects of smoking on the body.

Keywords: cigarette smoke, alkaloids, endocrine system, sex steroid hormones

Smoking is a public health problem. Every year, tobacco is responsible for the deaths of 3.5 million persons. It is estimated that in the decade 2020-2030, tobacco will kill 10 million people a year [1]. Increase public awareness about the harmful effects of tobacco is a prudent and beneficial approach for health.

Tobacco smoke is a heterogeneous, complex and dynamic aerosol, which contains at least 7357 chemical compounds [2]. Many of these compounds are chemically active and have negative effects on hypothalamic-pituitary-adrenal axis and hypothalamic-pituitary-gonadal axis, on the respiratory, circulatory systems, on muscles and bones, on the immune response [3-7]. Harmful compounds identified in tobacco smoke could be: alkaloids, nitrosamines, polycyclic aromatic hydrocarbons, volatile compounds, heavy metals, aromatic amines, heterocyclic amines, many additives [1-3, 5].

According to the literature, the major alkaloids of tobacco leaves are: nicotine (nonprotonated, monoprotonated, diprotonated), N-methylanabasine, anatabine, anabasine, nornicotine, cotinine, 2 -3 dipyridyl (fig. 1) [2,3].

Nicotine, the main pyrolidonic alkaloid present in Nicotiana tabaccum is a neurotransmitter responsible for tobacco dependence. Nicotine has the ability to alter steroidogenesis in both women and men (fig. 2). Nicotine causes dopamine increasing in the brain by inhibiting monoamine-oxidase. A high level of dopamine is associated with maintaining the sensation of pleasure. Another effect of nicotine is the increased activity of acetylcholine, by using the acetylcholine receptors, and so, preventing apoptosis [2-4, 6, 8, 9].

Nitrosamines, tobacco and tobacco smoke specific substances formed during processing, drying and storage of tobacco, influence metabolic and neuroendocrine system functions. Researches in this area have shown that the main volatile nitrosamines identified in tobacco and in tobacco smoke are: N-nitroso-R-amine (R = methyl, methyl ethyl, diethyl) and N-nitroso-pyrrolidine. Non-volatile nitrosamines specific to tobacco, are numerous: N-nitrosonornicotine, methylnitrozoamine-pyridyl-butanone, N-nitrosoanatabine, N-nitrosoanabasine, nitroso-methyl-aminobutyric acid. Nitrosamines directly affect cellular DNA, including genes that protects against cancer [2, 3].

Recent studies have identified at least 539 polycyclic aromatic hydrocarbons in cigarette smoke and 400-500 volatile compounds (oxides of nitrogen, carbon, sulfur,

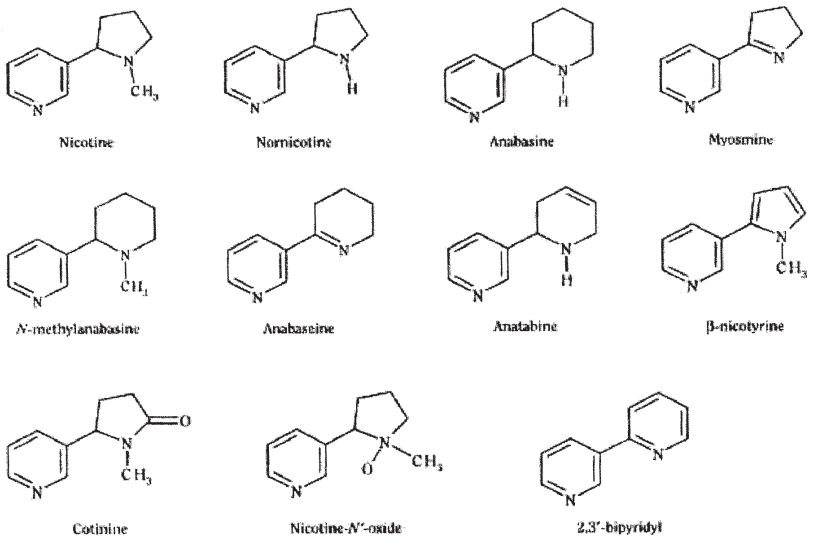

Fig. 1.Tobacco alkaloids (3)

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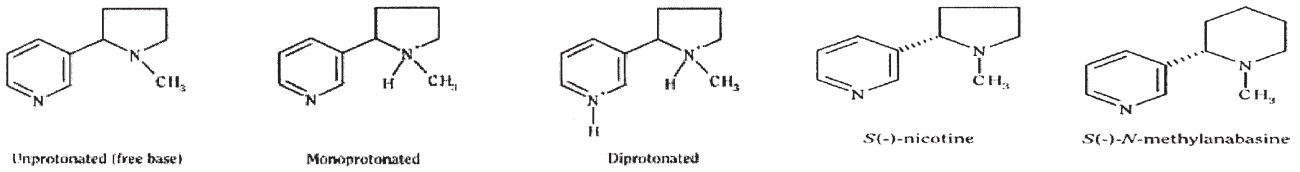

Fig. 1.Tobacco alkaloids (3)

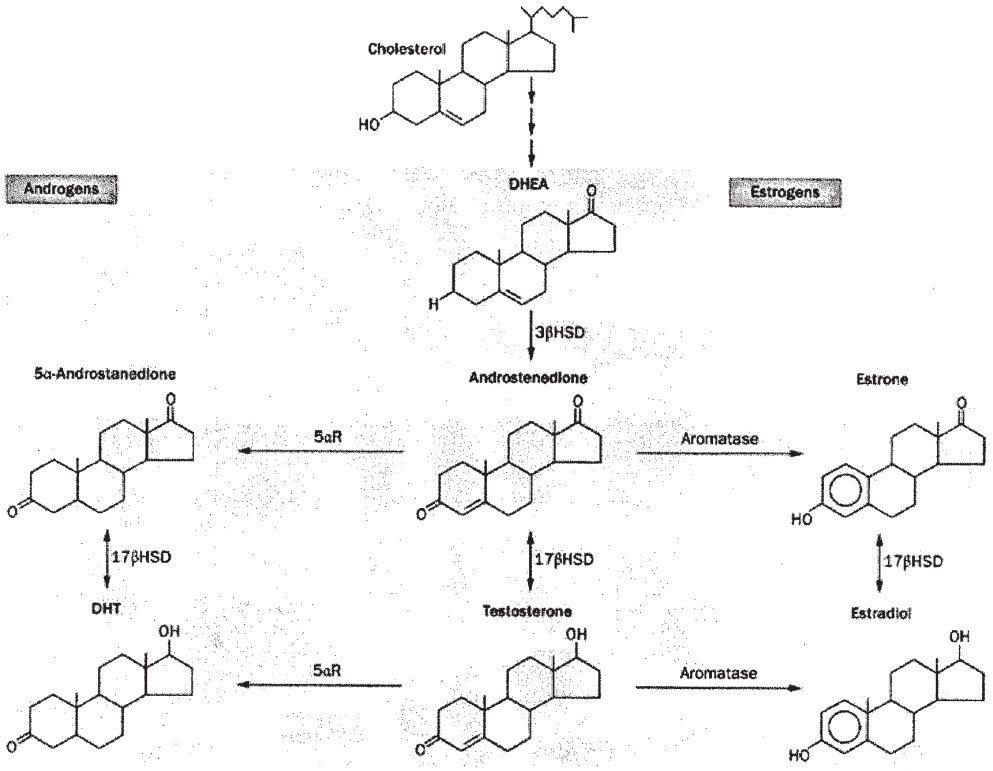

Fig. 2. Sexual hormones biosynthesis starting with cholesterol.
Abbreviations: 5αR, 5α-reductase; DHEA, dehydroepiandrosterone; DHT, dihydrotestosterone; HSD, hydroxysteroid dehydrogenase (9)

aromatic hydrocarbons, aldehyde and ketones, aliphatic hydrocarbons, nitrates, acids and bases) (2, 3).

The toxicity of tobacco and tobacco smoke is given by the presence of heavy metals, non-metals, complex metals, radioactive elements or isotopes nonradioactive. Frequently have been reported in tobacco presence of cadmium, copper, lead, mercury, nickel, arsenic, antimony, radon, polonium, arsenic, cadmium, nickel which affects the body ability to repair damaged DNA (2,3).

Aromatic amines present in cigarette smoke are accumulating in the body and increase the risk of cancer. Among them are included: dimethylaniline, toluidine, ethylaniline, naphthylamine, aminobiphenyl, methyl-naphthylamine (2.3).

Heterocyclic aromatic amines identified in the tobacco are derived of indole, imidasole, pyridil, dipyridil, quinones. These aromatic compounds have as precursors creatinine, amino acids, peptides, proteins, sugars (2,3).

Additives utilized for improving taste and increasing the attraction of smoking increases the harm of smoke.

The working hypothesis in this study was the fact that smoking affects the hormonal secretion and of various hormone regulators.

Studying the relationship between smoking and steroidogenesis was focused on the following objectives:

- investigating the serum levels of androgens and estrogens in men, smokers and nonsmokers, without apparent endocrine pathology ;

- investigating the serum levels of sexual steroid hormones in men, smokers and nonsmokers, with benign prostatic hyperplasia (BPH) (pathophysiological situations characterized by altered sex hormone status);

- assessing correlations between the change of serum levels of sex hormones and the clinical features of monitored subjects, on the one hand, and, on the other hand, between serum levels of sex hormones and a number of parameters involved in their metabolism .

A better understanding of the effects exerted by smoking on the endocrine system would allow the application of specific therapies and obtaining some useful information for developing effective approaches to smoking cessation.

Experimental part

Materials and methods

The effect of smoking on metabolism of steroid sex hormones was examined by determining serum levels of estradiol, testosterone, dihydrotestosterone through standardized working methods (ELISA).

For performing this analysis, there were selected 80 men aged between 50 and 65 years, with adequate nutritional status (table 1). Participants in the study were grouped as follows:

- control group - smokers and nonsmokers ;

- group with benign prostatic hyperplasia - smokers and nonsmokers .

Statistical analysis of data was done using SPSS software and followed:

- establishing a relationship between serum levels of steroid sex hormones in smokers and nonsmokers in normal conditions (control group) and in pathological conditions maintained by hormonal imbalance (benign prostatic hyperplasia);

- determining the causal relationship between the serum variation of sex hormones and a number of factors which considers the severity of prostate disease (International

Parameters	Control		BPH	
	Nonsmokers	Smokers	Nonsmokers	Smokers
Age (years)	58.1±4.3	57.3±6.2	59.1±_3.9	55.9±4.2
BMI(Kg/mp)	23.9±1.5	24.0±0.6	23.6±0.7	22.9±1.1
Glucose (mg/dl)	86.2±6.2	88.1±9.1	91.1±5.2	84.9±5.8
ASAT(U/L)	18.9±5.1	23.1±8.4	27.3±6.2	25.0±3.8
ALAT(U/L)	23.9±3.5	24.9±3.9	26.1±3.8	23.1±4.4
Chol (mg/dl)	188.1±11.2	169.9±9.8	183.2±11.3	179.1±12.4
HDL chol (mg/dl)	43.8±4.6	44.6±6.1	48.1±8.2	46.2±5.2
LDLchol (mg/dl)	93.4±11.3	99.1±12.0	103.5±13.1	98.8±11.1
CRP (mg/dl)	0.33±0.39	0.86±0.59	0.85±0.45	1.54±0.89
PSA (ng/dl)	0.9±0.5	1.1±0.5	4.1±1.1	4.3±0.9
Zn (ug/dl)	94±11	91±13	72±16	75±19
T (ng/ml)	4.9±1.4	5.3±2.1	3.6±1.5	3.2±2.1
DHT(pg/ml)	423±102	406±98	501±68	523±87
E2 (pg/ml)	30.8±4.2	28.0±6.1	37.8±7.3	43.3±11.2
IPSS	3.2±1.3	5.2±2.1	21.4±6.6	23.2±5.7

BMI- body mass index; ASAT - aspartate aminotransferase; ALAT - alanine aminotransferase; Chol – cholesterol; CRP – C reactive protein; PSA – prostatic specific antigen; Zn – zinc; T – testosterone; DHT – dihydrotestosterone; E2 – estradiol; IPSS- International Prostatic Symptom Score;

Table 1
BASAL CHARACTERISTICS OF PARTICIPANTS FROM THE STUDY

Hormones		Nonsmokers		Smokers	
		IPSS<20	IPSS>20	IPSS<20	IPSS>20
T	r	0.08	-0.26	-0.09	-0.30
	p	0.36	0.09	0.23	0.07
DHT	r	0.13	0.24	0.12	0.27
	p	0.51	0.07	0.21	0.00
E2	r	0.09	0.22	0.18	0.28
	p	0.57	0.16	0.35	0.06
DHT/T	r	0.11	0.26	0.14	0.41
	p	0.31	0.05	0.16	0.00
E2/T	r	0.15	0.11	0.09	0.18
	p	0.61	0.15	0.36	0.12

T – testosterone; DHT – dihydrotestosterone; E2 – estradiol; r- correlation coefficient ; p- statistical significance ; IPSS- International Prostatic Symptom Score;

Table 2
CORRELATIONS BETWEEN SERUM LEVEL OF SEXUAL HORMONES AND IPSS SCORE IN PATIENTS WITH BPH

Prostate Symptom Score - IPSS , PSA - prostate specific antigen) or certain factors that stimulate or suppress the synthesis of sex hormones (concentrations serum zinc and C-reactive protein).

Results and discussions

In the control group, there was an insignificant increase in serum levels of testosterone in smokers compared with nonsmokers (5.3 ± 2.1 ng/mL versus 4.9 ± 1.4 ng/mL, p> 0.05). In patients with BPH, there was not a significant reduction in serum levels of testosterone in smokers compared to nonsmokers (3.2 ± 2.1 ng/mL versus 3.6 ± 1.5 ng / mL, p> 0.05) (table 1). Statistically significant reductions were obtained between variation of testosterone in patients with BPH and controls (3.4 ± 2.0 ng/mL versus 5.1 ± 1.9 ng/mL, p <0.05).

Serum dihydrotestosterone showed statistically not significant variations between smokers and nonsmokers both in control group (406 ± 98 ng/mL versus 423 ± 102 ng/mL, p> 0.05) and in BPH group (523 ± 87 ng/mL versus 501 ± 68 ng/mL, p> 0.05) (table 1). Increases with statistical significance were obtained between variation of dihydrotestosterone in patients with BPH and in control group (511 ± 109 ng/mL versus 414 ± 116 ng/mL,p <0.05).

Serum estradiol showed statistically significant increases in patients with BPH compared to control (39.6 ± 13.2 pg/mL versus 28.9 ± 7.3 pg/mL, p <0.05). In contrast, no statistically significant differences were obtained between variations in serum estradiol in smokers and nonsmokers or to control (28.0 ± 6.1 pg/mL versus 30.8 ± 4.2 pg/mL, p> 0.05) or in patients with BPH (43.3 ± 11.2 pg/mL versus 37.8 ± 7.3 pg/mL, p> 0.05) (table 1).

Based on these results it can be estimated that smoking induced increasing of testosterone synthesis and reduced production of dihydrotestosterone and estradiol in the control group.

These effects were not found in patients with BPH, a situation which could be explained by the intervention and other mechanisms in the regulation of sexual steroid hormone synthesis and secretion. Smoking stimulates also the synthesis of CRP in the analyzed groups (table 1).

Modification of serum levels of sex hormones was analyzed according to IPSS score and serum concentrations of PSA, CRP and zinc. It must be mentioned that there was no significant correlation between serum levels of sex steroid hormones and values, within the normal range, of IPSS, PSA, CRP, zinc. Therefore there will be discussed further, the established statistical associations

Hormones		Nonsmokers		Smokers	
		PSA<4	PSA>4	PSA<4	PSA>4
T	r	0.04	-0.16	0.13	-0.44
	p	0.83	0.07	0.41	0.03
DHT	r	0.07	0.10	0.16	0.27
	p	0.67	0.05	0.19	0.00
E2	r	0.17	0.12	0.07	0.19
	p	0.57	0.16	0.35	0.06
DHT/T	r	0.26	0.34	0.21	0.42
	p	0.14	0.04	0.26	0.00
E2/T	r	0.18	0.26	0.15	0.29
	p	0.35	0.07	0.53	0.02

T – testosterone; DHT – dihydrotestosterone; E2 – estradiol; r- correlation coefficient ; p- statistical significance ; PSA – prostatic specific antigen;

Table 3
CORRELATIONS BETWEEN SERUM LEVEL OF SEXUAL HORMONES AND PSA(ng/mL) IN PATIENTS WITH BPH

Hormones		Nonsmokers		Smokers	
		PSA<4	PSA>4	PSA<4	PSA>4
T	r	0.14	-0.53	-0.04	-0.39
	p	0.77	0.02	0.56	0.00
DHT	r	0.08	0.18	0.13	0.24
	p	0.08	0.11	0.62	0.01
E2	r	0.14	0.08	0.17	0.00
	p	0.94	1.00	0.42	1.00
DHT/T	r	0.06	0.35	0.23	0.62
	p	0.76	0.00	0.61	0.00
E2/T	r	0.12	0.29	0.16	0.49
	p	0.38	0.08	0.10	0.03

T – testosterone; DHT – dihydrotestosterone; E2 – estradiol; r- correlation coefficient ; p- statistical significance ; PSA – prostatic specific antigen; CRP – C reactive protein ;

Table 4
CORRELATIONS BETWEEN SERUM LEVEL OF SEXUAL HORMONES AND CRP (mg/dL) IN PATIENTS WITH BPH

Hormones		Nonsmokers		Smokers	
		Zn<60	Zn>60	Zn<60	Zn>60
T	r	0.22	0.00	0.49	0.19
	p	0.63	0.96	0.00	0.04
DHT	r	-0.16	0.11	-0.33	-0.12
	p	0.10	0.75	0.03	0.09
E2	r	0.14	0.08	0.17	0.00
	p	0.94	0.87	0.33	0.96
DHT/T	r	-0.26	0.09	-0.29	-0.13
	p	0.03	0.63	0.00	0.15
E2/T	r	-0.11	0.16	-0.14	0.07
	p	0.68	0.57	0.89	0.67

Table 5
CORRELATIONS BETWEEN SERUM LEVEL OF SEXUAL HORMONES AND ZINC (ug/dL) IN PATIENTS WITH BPH

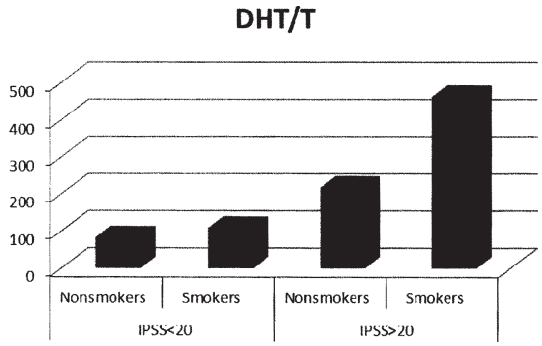

Fig. 3. Graphical representation of DHT/T ratio according to IPSS score in patients with BPH.

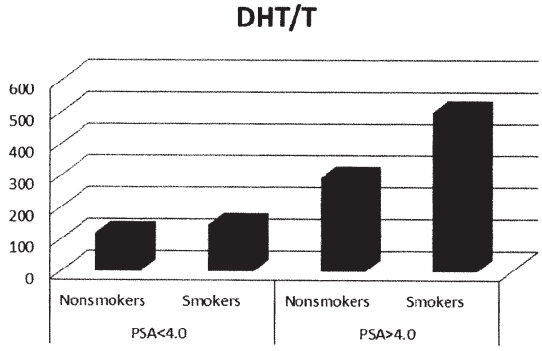

Fig. 4. Graphical representation of DHT/T ratio according to PSA (ng/mL) level in patients with BPH.

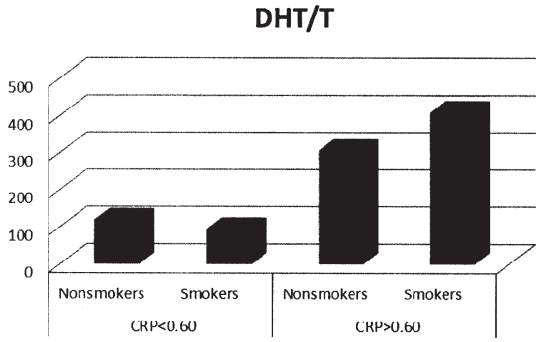

Fig. 5. Graphical representation of DHT/T ratio according to CRP(mg/dL) level in patients with BPH.

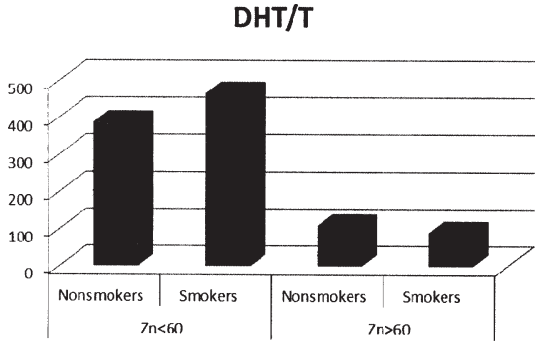

Fig. 6. Graphical representation of DHT/T ratio according to Zn(ug/dL) level in patients with BPH.

between sex hormone levels and IPSS, PSA, CRP, zinc only in patients with BPH stratified according to normal or pathological values of these factors (tables 2-5).

The effect of smoking on the metabolism of steroid hormones in patients with BPH is supported by the following results:

- getting a negative association between serum levels of testosterone and IPSS> 20 (table 2), PSA> 4ng/mL (table 3), CRP> 0.60 mg / dL (table 4). Between the level of testosterone and zinc <60mg/dL was obtained a positive association (table 5). These correlations are stronger in monitored smokers compared with nonsmokers;

- highlighting a positive correlations between levels of dihydrotestosterone and IPSS> 20 (table 2), PSA> 4ng/mL (table 3), CRP> 0.60 mg / dL, respectively, negative correlation with zinc <60ug/dL (table 5) only for smokers;

- highlighting a positive associations with statistical significance between serum levels of estradiol and PSA> 4 ng/mL only in smokers (table 3);

- statistically significant variation of the ratio between serum levels of testosterone and dihydrotestosterone (DHT / T) and IPSS> 20 (fig. 3), PSA> 4 ng/mL (fig. 4), CRP > 0.60 mg/dL (fig. 5), zinc <60 ug/dL (fig. 6), only in smokers;

- statistically significant variation in the ratio between serum levels of estradiol and testosterone (E2 / T) and PSA> 4 ng/mL (fig. 8) and CRP> 0.60 mg / dl (fig. 9) only for smokers. It was no statistically significant relationship

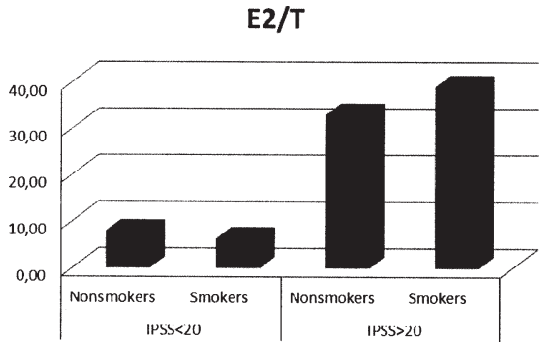

Fig. 7. Graphical representation of E2/T ratio according to IPSS score in patients with BPH.

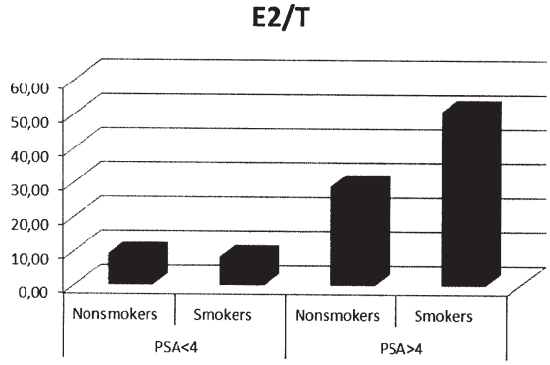

Fig. 8. Graphical representation of E2/T ratio according to PSA (ng/mL) level in patients with BPH.

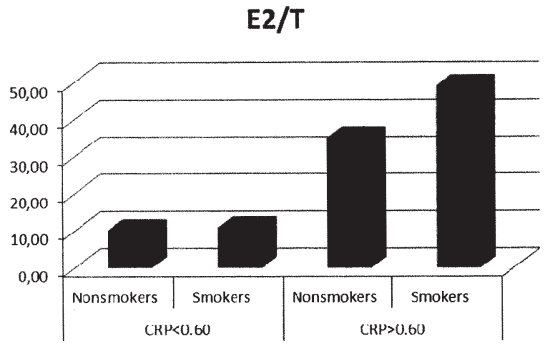

Fig. 9. Graphical representation of E2/T ratio according to CRP(mg/dL) level in patients with BPH.

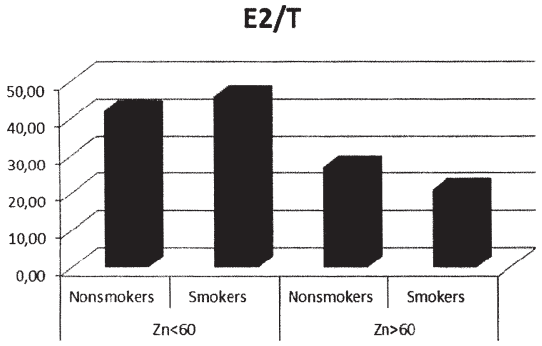

Fig. 10. Graphical representation of E2/T ratio according to Zn(ug/dL) level in patients with BPH.

between E2 / T and IPSS (fig. 7), respectively E2 / T and zinc (fig. 10).

Epidemiological and experimental data show that the risk of developing BPH is higher in smokers compared with nonsmokers. There was evoked a number of effects caused by smoking on the functioning pituitary, thyroid, adrenals, gonads and ovaries. Smoking activates the hypothalamic - pituitary- adrenal and gonadal axis, stimulates synthesis of adrenocorticotropic hormone (ACTH), activates cortisol synthesis, modifies the affinity of nicotinic receptors from ligands, affects the synthesis of acetylcholine, dopamine and prolactin [6, 10, 11]. In the literature there are some conflicting results on the effect of tobacco and tobacco smoke on serum levels of sex hormones. Nicotine affects the level of total and free testosterone, dehydro-epiandrosterone (DHEA), dehydroepiandrosterone sulfate (DHEAS), sex hormone binding globulin (SHBG), luteinizing hormone (LH) [11-15]. Increasing of testosterone levels in smokers was justified by some authors by stimulating SHBG synthesis [12]. High levels of androgens in smokers women were explained by the ability of nicotine to inhibit aromatase [13]. In experimental models, it has been shown that nicotine and cotinine can act as competitive inhibitors of 3 alpha -dehydrogenase steroid (enzyme that converts dihydrotestosterone to andro-stanediole- 3 alpha) [14]. Nicotine also affects the hepatic metabolism of testosterone by inactivating 7 alpha hydroxylase and by stimulating 6 beta hydroxylase [1].

Recent studies have shown that PSA level was higher in smokers compared with nonsmokers. Effects of smoking on PSA were investigated in two age groups (25-30 years and 50-70 years) and results were similar [15- 17]. As a result, smoking promotes the synthesis and secretion of PSA, representing a risk factor in the development of prostate disease.

Conclusions

Toxins and chemicals of which we are exposed every day could induce malfunction of the endocrine and immune systems. Pesticides, herbicides, xenoestrogens, heavy metals, radiation, drug abuse, being overweight, may affect steroid hormone metabolism. The results presented above, related to the impact of smoking on the human body, suggests that alkaloids could be a major cause of spoilage steroidogenesis. The authors appreciate that the study of the effect of smoking on the synthesis and secretion of sex hormones requires a more complex analysis concerning the exerted influence by certain enzyme modulators or inflammatory stimuli. In this context, the authors proved that serum levels of zinc and CRP exerted a significant effect on the synthesis of sex hormones. Zinc is necessary for the conversion of androstenedione to testosterone and dihydrotestosterone in regulating the expression of estrogen receptor and androgen receptor, in the aromatisation of testosterone to estrogen, in synthesis of growth factors and in signaling regulation. Pro-inflammatory factors that are increasing as response to prostatic lesions play a role in the clearance of cellular debris and in accelerating antigens and microorganisms phagocytosis.

Smoking causes changes in the synthesis and secretion of sex steroid hormones that could promote, in a particular context, the development of prostatic pathology. The major objective of the worldwide scientific community is represented by finding means to mitigate negative effects caused by smoking on the body.

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