Histological Effects of Smokeless Tobacco onthe Endometrial Glandsof the Orally Treated Female Swiss Albino Rats

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ABSTRACT

Introduction: Tobacco use is the leading cause of mortality all over the world. Smokeless tobacco contains higher quantities of nicotine than most cigarettes. Over the past few decades the use of tobacco among women of reproductive ages has increased. Exposure to the toxins present in the tobacco interferes with the endometrial receptivity, endometrial angiogenesis and uterine blood flow.

Material and Methods: 30 adult female Swiss albino rats were randomly selected. They were divided into three groups (n=10 each). Group A was kept as control. Experimental groups B&C consisted of rats which were given 5 % & 10% of smokeless tobacco in their feed respectively. Feed and water were given ad libitum. On 31stday the animals were sacrificed. Uterus of all the animals were removed and weighed. The tissues were processed for histological examination under light microscopy using H & E and Trichrome stains.

Results: A significant decrease in the weight of the uterus was observed (P value ≤ 0.001). The histological changes in the uterus of experimental groups revealed severe cystically dilated sub mucosal glands. Endometrial glands also showed marked atrophy (P value ≤ 0.001).

Conclusion: From this study it can be concluded that the smokeless form of tobacco causes adverse effects on the endometrium of the female Swiss albino rats. Long term use of this form of tobacco may lead to adverse reproductive outcomes or other pathological conditions of uterus.

Key Words: Swiss albino rats, smokeless tobacco, uterus.

I. INTRODUCTION

The use of smokeless tobacco is a worldwide practice. Numerous variations have been observed in the nature of the product used as well as in the customs associated with its use[1].

The smokeless form of tobacco consists of crudely divided tobacco leaf which is mixed with sugar and syrup and is usually packaged in brightly colored packets. The most commonly available preparations of smokeless tobacco in subcontinent region are gutka, zarda, paan, mainpuri ,gul, and mishri[2].

Studies conducted on females in the region of South Asia have concluded that due to easy accessibility and lack of awareness regarding the tobacco products there is an increased consumption of smokeless tobacco products2,3.

Nicotine which is an important alkaloid present in smokeless tobacco was also identified in the endometrial fluid .Animal models exposed to toxic metabolites of tobacco have shown abnormal endometrial maturation4.Exposure to the toxins present in the tobacco interferes with the endometrial receptivity, endometrial angiogenesis and uterine blood flow[5].

Material and methods:This study was carried out in the Sindh Agricultural University, TandoJam. 30 adult female Swiss albino rats were randomly selected. They were divided into three groups (n=10in each group). Animals in Group A were kept as control. Experimental Groups B& C consisted of rats which were given 5 % & 10% of smokeless tobacco in their feed. Feed and water were provided ad libitum. The experiment was carried out for 30 days.On 31st day the animals were sacrificed by cervical dislocation. Uterus of all animals were removed and weighed. The tissue were then processed for histological examination under light microscopy using H & E and Trichrome staining methods.

Statistical Analysis:The statistical analysis was done by using SPSS version 16.0. Measures of central tendency

were applied. Chi- Square test and student-t test were applied to compare different groups.

Results: A significant decrease in the uterine weight of Group B was also noted with a mean value of 0.596±0.009 gm. However a highly significant reduction in the uterine weights of the oral smokeless tobacco treated rats was observed in the experimental group C with a mean value of 0.499 ±0.011gm as compared to the controls having mean value of 1.115± 0.005 gm.(p-value <0.001) (Table No: 1)

Histological changes in the uterus:Examination of the uterus of the control rats showed the normal histological structure of the endometrial glands and surrounding stroma (Photomicrograph No.1).A significant reduction in the size of endometrial gland was also observed in the experimental groups (photomicrograph No.2).The endometrial glands of the group C also revealed severe cystically dilated sub mucosal glands. Marked atrophy was also observed in theendometrial glands(P value ≤ 0.001, Fig:01, photomicrograph No.3).

II. DISCUSSION

It is well known that through neural stimulus to GnRH, hypothalamus regulates the rhythmic release of pituitarygonadotrophins, i.e., FSH, LH and prolactin. Investigations on constituents of the tobacco indicate that nicotine being a centralnervous system influencing drug inhibits the release of gonadotrophins from pituitary.Uterine growth depends upon the ovarian estrogen secretion. Estrogen primarily acts upon the surface epithelium and the glands within endometrium[7]

Progesterone acts on estrogen primed uterus and prepares the uterine epithelium from proliferative to secretory state. In the present investigation, reduced thickness of endometrium and reduction in the size of endometrial glands indicates the inhibition of ovarian steroid biosynthesis necessary for growth of the uterus and reproductive cyclicity.[7]

Toxins present in the tobacco interfere with the endometrial receptivity, endometrial angiogenesis and uterine blood flow[6].

In our study the use of smokeless tobacco produced endometrial degeneration along with edema and fibrosis. Necrosis, atrophy and cystic dilatation in endometrial glands were also observed in the uterus of the tobacco treated rats.

Similar findings wereobserved showing that the uterus of the animals receiving nicotine administration exhibited marked reduction in the thickness of both endometrium and myometrium with an observable reduction in the endometrial glands[8].

Another study done by Zhang et al., 2007 also shows that the compounds found in tobacco are capable of altering the epithelial cell layer of endometrium and myometrium. Tobacco was also found to produce stromal inflammation and cellular edema.The reduced thickness of the endometrium and myometrium and decrease of number of the endometrial glands observed in the present study may result from the impaired hormonal support (estrogen and progesterone), since estrogen is responsible for the proliferation of the uterine muscles and progesterone is responsible mainly for the development of the uterine glands and glycogen deposition, however the direct toxicity of nicotine or its related metabolites cannot be excluded. The later factor may explain the degenerative and atrophic changes observed specially in the uterine epithelium and glands[9]. It has also been suggested that CYP1A2 and CYP1B1 are not induced by(benzo (a) pyrene) in the endometrial cells.These findings are in accord with the present study which also clearly exhibits that use of tobacco leads to impairment in the endometrial receptivity and angiogenesis.

III. CONCLUSION

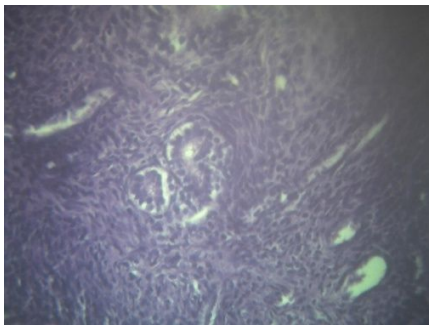
Based on the histological findings in this study it can be easily stated that the exposure to the oral smokeless tobacco alters every stage of the uterine activity.

IV. RECOMMENDATIONS

In the light of present study it is recommended that measures should be taken to discourage females in the reproductive age regarding use of smokeless tobacco. Preventive strategies including education aboutthe effects of tobacco at adolescent level and change in society norms should be carried out. Further studies should be carried out to observe the effects of smokeless tobacco on the fertility rate and its effects on the fetal outcomes.

TABLE NO: 1

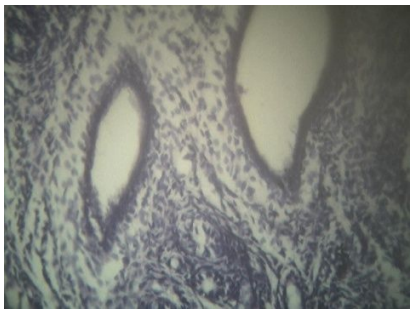
Comparison of weight of uterus between Group A and Group C using Student t-test p-value<0.05*			
Statistics	Weight of uterus		P-value*
	Group A(n=10)	Group C (n=10)	
Mean	1.115	0.499	0.001
Std. Dev	0.005	0.011	0.001



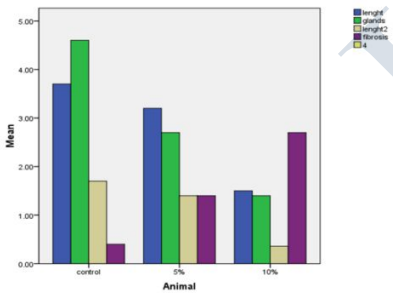
Photomicrograph No.1: uterus of group A showing healthy endometrial glandsx 10



Photomicrograph No.2: uterus of group B showing mildly atrophied endometrial glands x 10



Photomicrograph No.3: uterus of group C showing cystically dilated endometrial glands x 40



(Figure 01)

Comparison of histological findings of uterus of rats of control, Group B & C.

REFERENCES

[1] Backinger,C.L., Fagan, P., Matthew, E and Grana, R (2003) Adolescent and young adult tobacco prevention and cessation: current status and future directions. Journal of Tobacco Control, Vol.12, pp. 46-53.

[2] Srivastava, M., Parakh, P and Srivastava, M. (2010) Predictors and prevalence of nicotine use in females: a village-based community study Indian Journal of Psychiatry, Vol.19, pp. 125-129.

[3] Warren, C.W., Riley, L., Asma, S., Eriksen, M.P., Green, L., Blanton, C., Loo, C., Batchelor, S and Yach, D (2000) Tobacco use in youth: a surveillance report from the Global Youth Tobacco Survey Project. Bulletin of World Health Organization. Vol. 78, pp. 868-876.

[4] Cooke, J.P and Bitterman, H. (2004) Nicotine and angiogenesis: a new paradigm for tobacco-related diseases. Annals of Medicine, Vol. 36, pp. 33-40.

[5] Zhang, W., Yang, J., Wang, J., Xia, P., XU ,Y., Jia, H and Chen Y.(2007) Comparative studies on the increase of uterine weight and related mechanisms of cadmium and p-nonyl phenol, Journal of Toxicology, Vol,241, pp. 84-91.

[6] Xiao, D., Huang, X., Yang, S., Zhang, L (2007) Direct effects of nicotine on the contractility of the uterine artery in pregnancy.Journal of Pharmacology and Experimental Therapeutics, Vol.322, pp. 180-185

[7] AnastasiaA. Varvarigou., Spyros G. Liatsis.,PavlosVassilakos, George DandNicholas G. Beratis(2009)Effect of maternal smoking on cord blood estriol, placental lactogen, chorionic gonadotropin, FSH, LH, and cortisol,J. Perinat. Med. 37 (2009) 364–369.

[8] El-Meligy, M.M.S., Hady, R.H.A., AbdelSamaei., A.R and. SaadEldien, H.M.(2007) effect of nicotine administration and its withdrawal on the reproductive organs, fertility,and pregnancy outcome in female rats.Journal of Forensic Medicine and Clinical Toxicology, Vol. 15, No. 1, pp. 95-132.

[9] Bao H, Vepakomma M, Sarkar MA. Benzo(a)pyrene exposure induces CYP1A1 activity and expression in human endometrial cells. J Steroid BiochemMolBiol 2002;81:37-45

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