

Aging process of epithelial cells of the rat prostate lateral lobe in experimental hyperprolactinemia induced by haloperidol

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Abstract

The aim of the study was to examine the influence of hyperprolactinemia, induced by haloperidol (HAL) on age related morphology and function changes of epithelial cells in rat prostate lateral lobe. The study was performed on sexually mature male rats. Serum concentrations of prolactin (PRL) and testosterone (T) were measured. Tissue sections were evaluated with light and electron microscopy. Immunohistochemical reactions for Anti-Proliferating Cell Nuclear Antigen (PCNA) were performed. In rats of the experimental group, the mean concentration of: PRL was more than twice higher, whereas T concentration was almost twice lower than that in the control group. Light microscopy visualized the following: hypertrophy and epithelium hyperplasia of the glandular ducts, associated with increased PCNA expression. Electron microscopy revealed changes in columnar epithelial cells, concerning organelles, engaged in protein synthesis and secretion.

Key words: aging, lateral lobe, rat prostate, hyperprolactinemia.

Introduction

Due to the progress of aging of males in the industrialized civilization, disorders of the prostate gland, including inflammatory disorders, hyperplasia of the secretory epithelium, benign hypertrophy and malignancy become increasing prob-

lems in the population. The aetiology is, in general, searched for in paracrine endocrinological disorders. One of the reasons is hyperprolactinemia, revealing physiologically among men after the 60th year of life, or either as a symptom of various diseases or a side effect of various drug treatments. Influence of PRL on the prostate acts on dual level - as that of a pituitary hormone and that of cytokine, exerting either autocrine or paracrine effects. Prolactin membrane bound receptor (PRLR) is widely dispersed in the rat prostate - dorsal, lateral, ventral lobes [4], as well as in the human prostate [3]. An *in vivo* rodent grafted pituitary indicated a local direct proliferate, trophic effect of PRL on the prostate, independently of the circulating androgen levels, especially in the lateral lobe [1]. The metabolism of the prostate gland is also affected by PRL: in rats, the androgen independently stimulates citrate production, especially in the lateral lobe and changes cellular and mitochondrial zinc levels [2]. Androgens directly control most of the proteins, secreted by epithelial cells of the lateral lobe of the rat prostate. The aim of our study was to examine the influence of hyperprolactinemia on age-related changes in the morphology and function of epithelial cells of the rat prostate lateral lobe.

Material and Methods

The studies were performed on sexually mature 18 months old Wistar rats, divided into experimental and control groups, with 10 animals in each. Hyperprolactinemia was induced by intraperitoneal injections of a Haloperidol (Polfa Warszawa, Poland) 2.0 mg/kg body weight, administered for 14 days. Animals of the control group received saline in an equal volume. Serum levels of PRL were measured by ELISA (Spi-Bio, France), and T levels were measured by RIA (Orion Diagnostics, Finland). The prostate lateral lobe was fixed in 4% neutral buffered formalin or Bouin's fluid and embedded in paraffin. Histological analysis was performed on serial sections, obtained from prostatic samples stained with either H-E or P.a.s. For

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Figure 1A. Cross section through the gland and stroma of prostate lateral lobe from an 18-mth control rat.

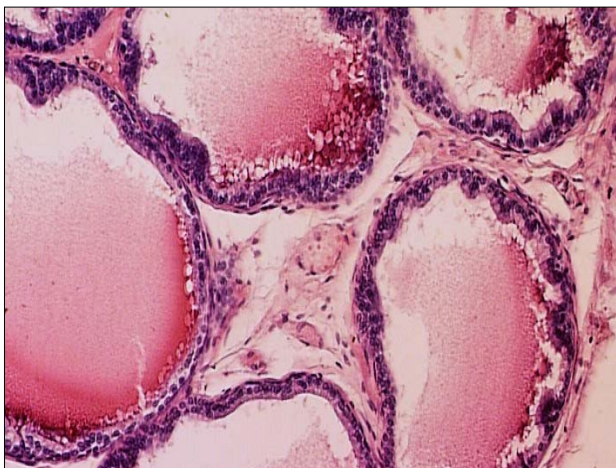


Figure 2A. The Golgi apparatus in the columnar epithelial cells of the lateral lobe of a control prostate.

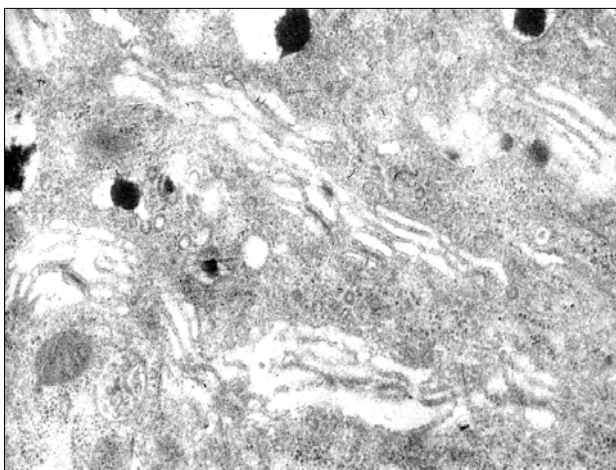
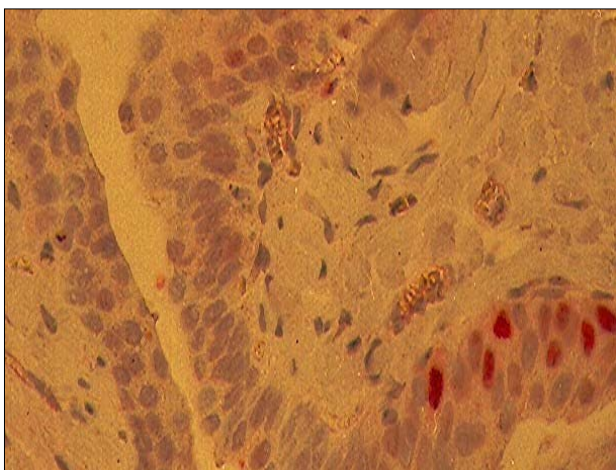


Figure 3A. Immunohistochemical expression of PCNA in the epithelial cells of the prostate lateral lobe in a control rat,



immunohistochemical detection of Proliferating Cell Nuclear Antigen (PCNA) in the prostate lateral lobe, DAKO EnVision System Alkaline Phosphatase (DAKO, Denmark). was used. Tissue sections were incubated with: primary antibody Monoclonal Mouse Anti-PCNA Clone PC10 and Fast Red (DAKO, Denmark) was used to visualize the reaction. Negative controls con-

Figure 1B. Papillary infoldings and multistratified cell clusters in HAL rat (100x).

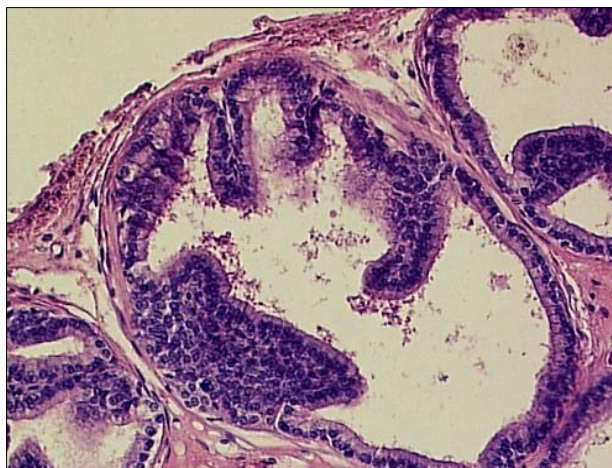


Figure 2B. Dilated cisterns GA and residual material in HAL rat, the lateral prostate (7000x).

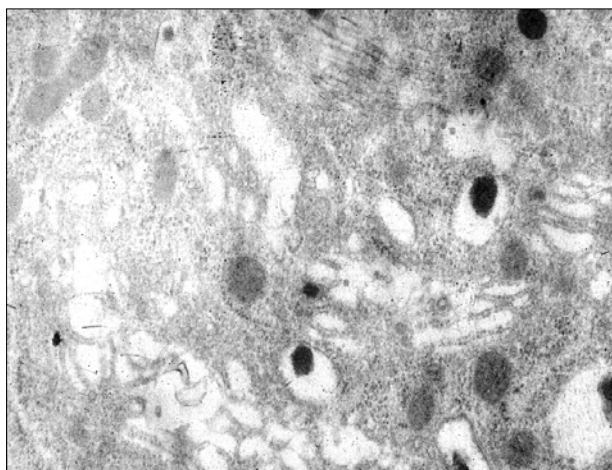
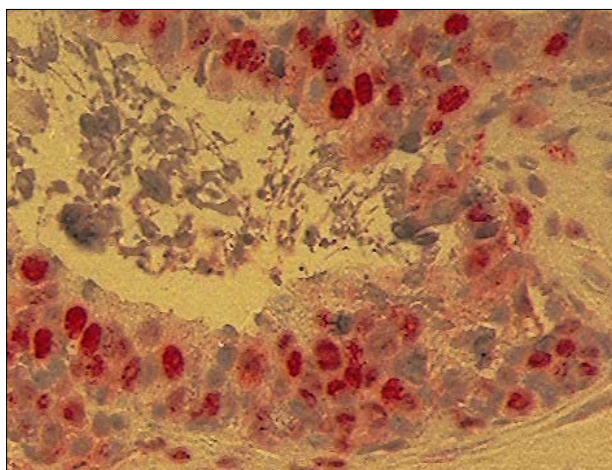


Figure 3B. and after HAL (400x).



sisted of omissions of the primary antibody. For transmission electron microscopy (TEM), samples of the lateral lobe of the prostate were routinely prepared and finally examined under a JEM-1200 EX. Variations in plasma hormone levels were analyzed by the Cochran-Cox test. Statistical significance was established at levels of $p < 0.05$.

Results

In rats of the experimental group, the mean concentration of PRL (79.7ng/ml) was almost twice higher, whereas the mean concentration of T (1.35ng/ml) was twice lower, compared to respective values in the control group (PRL 45.7ng/ml, T 3.42ng/ml). Also noticed were: hypertrophy of the glandular ducts, hyperplasia of epithelium, forming papillary infoldings and multistratified cell clusters. Within the hyperplastic areas in the connective tissue, we noticed inflammatory infiltrations, consisting of: lymphocytes, histiocytes and plasmocytes, surrounding the glands. The characteristic microscopic image, as described above, occurred focally, while other areas of the observed gland were unaffected.

Most of the changes, observed in electron microscopy, concerned intracellular organelles, engaged in protein synthesis and secretion - rough endoplasmatic reticulum, Golgi apparatus (GA) and the luminal secretory surface of epithelial cells. Immunohistochemical expression of PCNA antigen reveals a correlation to hyperplastic activity of the epithelium. An increased presence of PCNA was observed in papillary infoldings and clusters of epithelium in acini.

Discussion

Eighteen (18) months old rats are relative to 60 years old humans. Hyperprolactinemia brings into prominence the effect of PRL on pathology of the prostate gland [5]. Spontaneous prostate disease is described only in primates, whereas in rodent animal models, it is induced by chemical treatment or genetic modulations [1, 6, 10]. The prostate gland in rats consists of three lobes: ventral, lateral and dorsal, among which only the dorsal and the lateral lobe are homological to their human equivalents. Prolactin levels increase with age, whereas testosterone parallelly decreases, indicating the age-dependent role of PRL in the proliferation control of prostatic cells, causing a development of benign prostate hyperplasia, dysplasia and, possibly, prostate carcinogenesis in the human [7].

An essential role in the unbalance of apoptotic cell death and the proliferation rate in the ageing prostate, clinically observed in elderly men, is played by androgens. In the cross talk between PRL and T up-regulation of the androgen receptor (AR) [10], an influence on 5 α -reductase occurs [9]. In 24 months old Brown Norway rats, despite a decline of T serum levels, prostatic hyperplasia reveals in dorsal and lateral lobes of the prostate, whereas it does not in the ventral lobe. As it has been revealed by results of immunohistochemical, and genetic studies, the epithelial cell proliferation rate correlates with AR expression: stronger in the dorsolateral prostate and weaker in the ventral lobe, when compared to respective values in the young rat [11]. As it was assessed in our previous studies, hyperprolactinemia is one of the causes, increasing AR expression in rat prostate epithelial cells [10]. Rats over 12 months reveal an inflammatory invasion, low acinar epithelium devoid secretory activities, acinies often have smaller diameters with a thick sur-

rounding fibromuscular stroma and lumen, filled up with proliferating or exfoliated epithelium [8]. That was corresponding to the results, obtained in our study on 18 month old rats. We observed: large secretory vacuoles, containing dense bodies, large excessively dilated cisterns of rough endoplasmatic system and the Golgi apparatus in the supranuclear region, and only a few secreted granules. PRL plays an essential role in prostate pathology. The results, as obtained in animals, put aside the detailed knowledge of the mechanism ruling in biology of the prostate gland. The influence of PRL takes place directly, modulating the growth, proliferation and metabolism of prostate cells, as well as indirectly, via sex steroid hormones and their receptors.

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