

Ultrastructural Study in Canine Prostatic Hyperplasia

— Effect of Antiandrogen —

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Ultrastructural changes in canine prostates after treatment with chlormadinone acetate (CMA) were investigated. Old male beagle dogs (5-8 years old) were divided into two experimental groups; group 1 consisted of benign prostatic hyperplasia (BPH) controls, and group 2 received 0.3mg/kg/day CMA orally for 6 months. In group 1 animals, the most striking ultrastructural changes were detected in the rough endoplasmic reticulum (rER) and Golgi complexes. The secretory granules were lined up along the apical plasma membrane, and exocytosis was frequently seen. In group 2 animals, the cytoplasm was electron-lucent and contained relatively few, poorly developed organelles. The rER was sparse and consisted of a few scattered, short profiles studded with ribosomes. The Golgi complexes were inconspicuous. The secretory granules were markedly decreased in both number and size. Furthermore, mitochondrial degeneration such as swollen or disappeared mitochondrial cristae or decreased electron density of the matrix were frequently seen in the smooth muscle cells. Based on our data, atrophy after treatment with CMA may be due to shrinkage of both glandular and stromal compartments in the prostate tissue.

Key Words: Chlormadinone acetate (CMA), Benign prostatic hyperplasia (BPH), Glandular epithelium, Smooth muscle cell, Mitochondria

INTRODUCTION

Chlormadinone acetate (CMA) and cyproterone acetate have been used in medical-treatment of human benign prostatic hyperplasia (BPH) or prostatic carcinoma [2,3,8,9]. The atrophic effects of CMA on the prostate of rats and dogs have been previously reported from our laboratory [5,6]. Using spontaneous BPH dogs we found that immunostaining of androgen receptor (AR) in both epithelial and stromal fibro-muscular cell nuclei was remarkably decreased by CMA treatment [7], which may be attributed to the decrease in the number of AR and /or antibody binding sites for AR. Therefore, we postulated that atrophy of the prostate after treatment with CMA is the result of effects not only on the epithelium but on the stroma. This report mainly deals with the ultrastructural changes in canine prostates (spontaneous BPH) after treatment with CMA.

MATERIALS AND METHODS

Animals

Male beagle dogs were used. The animals received dry dog food (CD-1, CLEA Japan, Inc.) and water *ad libitum*. Eight old dogs (5-8 years old) were considered to have BPH on the basis of biopsy.

Experiments

Four old animals in group 1 were used as untreated controls. Four old animals in group 2 were administered 0.3mg/kg/day CMA orally as crystalline powder in gelatin capsules for 6 months. All animals were sacrificed by exsanguination under pentobarbital anesthesia at the end of the experimental period.

Electron microscopic examination

Small pieces of prostates were fixed in 0.1M phosphate-buffered 2.5% glutaraldehyde (pH 7.4) for 3hr at 4°C under constant

agitation. The fixed tissues were washed with 0.1M phosphate buffer. After post-fixation in 0.1M phosphate-buffered 1% OsO₄ (pH7.4) for 1hr at 4°C, they were dehydrated in a graded ethanol series and embedded in Quetol 812. Ultrathin sections were prepared with an LKB ultra-microtome, were double stained with uranyl acetate and lead citrate and were observed under a JEOL 1200EX electron microscope.

RESULTS

Group 1 (BPH control) : Representative ultrastructural features of the glandular epithelial cells are shown in Figs 1 and 2. The most striking ultrastructural changes were detected in the rough endoplasmic reticulum (rER) and Golgi complexes in the epithelial cells. The rER showed dilated cisternae, which were often displaced by neighboring organelles. The Golgi complexes were

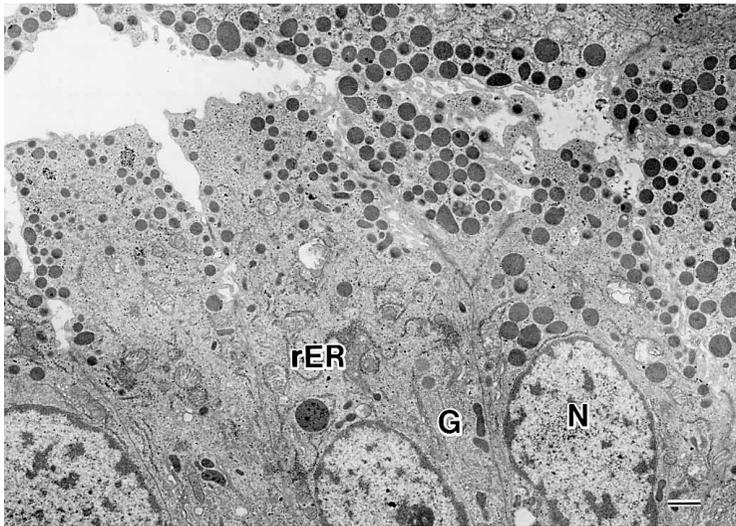


Fig. 1 Electron microscopic view of glandular epithelial cells in the prostate of BPH dogs. G: Golgi complexes, rER: Rough endoplasmic reticulum, N: Nucleus, Bar : 1 μ m, \times 7,000

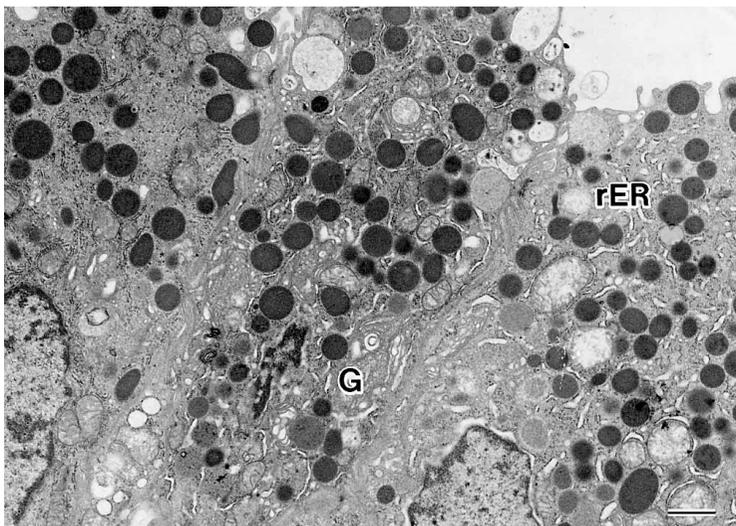


Fig. 2 Well developed rough endoplasmic reticulum (rER) and Golgi complexes (G) are characteristically seen. Bar : 1 μ m, \times 10,000

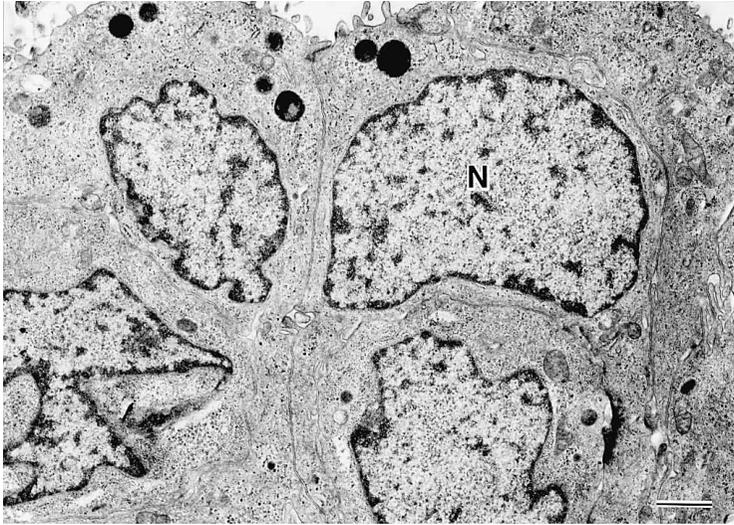


Fig. 3 Electron microscopic view of glandular epithelial cells in the prostate of BPH dogs after treatment with CMA. Atrophic glandular epithelial cells have electron-lucent cytoplasm containing relatively few, poorly developed organelles. N: Nucleus, Bar : 1 μ m, \times 12,000

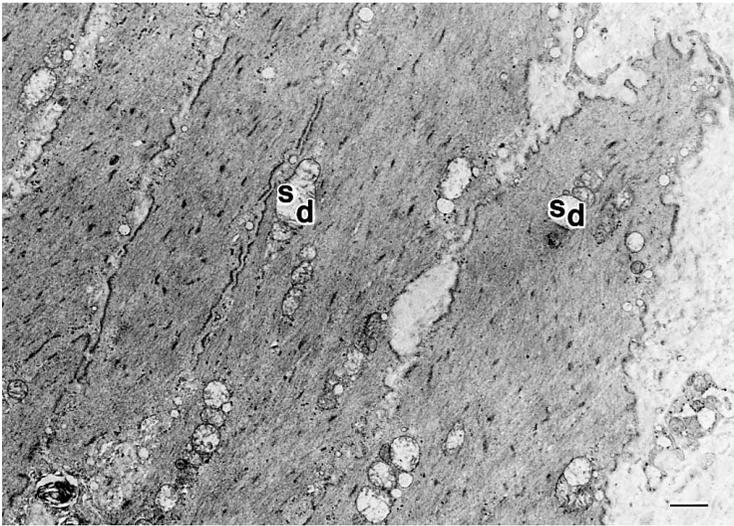


Fig. 4 Electron microscopic view of smooth muscle cells in the prostate of BPH dogs after treatment with CMA. Mitochondrial degeneration such as swollen (s) or disappeared mitochondrial cristae (d) or decreased electron density of the matrix are frequently seen. Bar : 1 μ m, \times 8,000

extensively developed and contained a few forming granules and some lysosomes. The secretory granules appeared to be slightly decreased, but were normal in shape and interior electron density. Occasionally, secretory granules were lined up along the apical plasma membrane, and exocytosis was frequently seen.

Group 2 (CMA) : The cytoplasm of the glandular epithelial cells were electron-lucent and contained relatively few, poorly developed organelles (Fig. 3). The rER were sparse and consisted of a few scattered, short profiles studded with ribosomes. The Golgi complexes were inconspicuous. The secretory granules were markedly decreased in both

number and size. Furthermore, mitochondrial degeneration such as swollen or disappeared mitochondrial cristae or decreased electron density of the matrix were frequently seen in the smooth muscle cells (Fig. 4).

DISCUSSION

The hyperplastic glandular epithelium was characterized by well developed rER and Golgi complexes. In addition, exocytosis was frequently seen, which appears to indicate enhanced secretory and metabolic activities in the prostate. Previously, Amuller *et al.* [1] reported that at the subcellular level, hyperfunction of the prostate was reflected by an increase in the amount of rER, in the size of the Golgi apparatus, in the number of Golgi vesicles, and in the number, size and electron density of secretory granules, in addition to high secretory activity of the glandular epithelium.

CMA induced the marked atrophy of the glandular epithelium, and loss of secretory and metabolic activities was also evident. Furthermore, the majority of stromal smooth muscle cells revealed mitochondrial degeneration such as swollen or disappeared mitochondrial cristae or decreased electron density of the matrix. Previous studies using spontaneous BPH dogs indicated that immunostaining of androgen receptor (AR) in the nuclei of both epithelial and stromal fibro-muscular cells was remarkably decreased by CMA treatment [7]. Based on our data, atrophy after treatment with CMA may be due to shrinkage of both glandular and stromal compartments in the prostate. In humans, prostatic hyperplasia (BPH) is the result of an increase in both glandular and stromal compartments. Therefore, it is suggested that effects on both compartments are required to achieve the intended clinical benefits in patients treated with CMA.

The precise mechanism by which CMA induces atrophy of the prostatic stromal compartments is not clear from the present investigation. In this context, quantitative evaluation of prostatic compartments by morphometric analysis, and immunohisto-

chemistry of 5 α reductases type I and type II in the prostate seem important in solving this problem. Further work along this line is now in progress in our laboratory.

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