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β-Adrenergic receptor manipulation and acid phosphatase and zinc levels in the ventral prostate of the adult rat

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The influence of 15-day treatments with the β-adrenergic receptor agonist isoproterenol (120 μg/kg/d) or the antagonist propranolol (1.00 mg/kg/d) on acid phosphatase and zinc levels in the ventral prostate was examined in intact rats, rats simultaneously injected with dexamethasone (0.25 mg/kg/d) and animals chemically castrated with a single dose of ethane dimethanesulphonate (75 mg/kg). Isoproterenol-treatment significantly increased acid phosphatase concentration in the ventral prostate of intact rats, whereas propranolol prevented a glandular zinc loss induced by dexamethasone administration. These results demonstrate that the levels of both biochemical parameters in the prostate can be altered by β-adrenergic receptor manipulation. The responsiveness of the two secretory processes is different and depends on the functional status of the ventral prostate.

1. Introduction

Growth and function of the prostate gland are primarily controlled hormonally by androgens. However, the abundance of adrenergic [1–4] and muscarinic [1, 5] receptors and nerve fibers [6–9] suggests that the autonomic nervous system may play a role in the growth and secretory functions of the prostate.

The prostate contains a high density of α_1- and β_2-adrenergic receptors. α_1-adrenergic receptors are present in the prostatic stroma, on the smooth muscle cells surrounding the acini and ducts, and they are responsible for the contractions of prostatic tissue elicited by catecholamines or α_1-adrenergic agonists [1, 2, 10–12]. β_2-Adrenergic receptors are localized on the glandular epithelial cells [3, 4, 12, 13], but their physiological relevance is virtually unknown. It is believed that these receptors have a role in the synthesis of secretory proteins in the rat prostate [14] and that the decrease in their number is responsible for the atrophic changes in the gland of rats with experimentally induced diabetes [13]. Furthermore, some of the long-term effects of prostatic denervation may be due to the lack of stimulation by β_2-adrenergic receptors [15]. We have reported that chronic administration of the β-adrenergic receptor agonist isoproterenol or the antagonist propranolol influenced the structure of the adult rat ventral prostate either by affecting blood testosterone levels or directly, which depended on the gonadal status of the animals [16–18].

The aim of this work was to examine the effects of 15-day β-adrenergic receptor stimulation with isoproterenol or blockade with propranolol on some biochemical parameters of prostate secretory activity, i.e. the acid phosphatase and zinc levels in the ventral lobe of the gland. These components of prostatic epithelial cells and their secretion are present in high concentrations, which may vary with changes in glandular function as well as in pathological conditions [19]. As it is known that the number of β_2-adrenergic receptors in the prostate depends on blood androgen levels [4, 12, 20, 21], the effects of isoproterenol and propranolol were followed in intact and chemically castrated rats. Chemical castration was performed with ethane dimethanesulphonate (EDS), a specific cytotoxic agent for adult rat Leydig cells [22]. In addition, since the stress response involves the activation of neural and hormonal networks that lead to an increase of catecholamine and glucocorticoid secretions, groups of intact animals simultaneously treated with dexamethasone were also used.

2. Investigations and results

The 15-day isoproterenol treatment (120 μg/kg/d) only affected the acid phosphatase level in the ventral prostate of intact rats (Fig.). It evoked a small, but statistically significant increase in the concentration of this enzyme (18%, $P < 0.01$).

Administration of propranolol (1.00 mg/kg/d) for 15 days influenced zinc level in the ventral prostate, but only in rats simultaneously treated with dexamethasone (Fig.). Both the concentration and glandular content were significantly increased (173%, $P < 0.05$ and 155%, $P < 0.05$, respectively) when compared with controls receiving dexamethasone only.

Dexamethasone-treatment (0.25 mg/kg/d) by itself decreased the levels of both measured biochemical parameters of the ventral prostate (Fig.). The acid phosphatase concentration was reduced by 34% ($P < 0.05$) and this reduction was expressed in the glands of rats simultaneously treated with isoproterenol or propranolol (46%, $P < 0.01$ and 42%, $P < 0.01$, respectively) as well. The zinc concentration and content in the ventral prostate were decreased in rats treated with dexamethasone (61%, $P < 0.05$ and 58%, $P < 0.05$, respectively), but not in rats treated with dexamethasone and isoproterenol or propranolol.

Chemical castration, induced by an injection of EDS (75 mg/kg), also reduced the levels of both biochemical parameters in the ventral prostate (Fig.). The decreases for acid phosphatase concentration and content were 42% ($P < 0.01$) and 87% ($P < 0.01$), respectively, and for zinc concentration and content 46% ($P < 0.05$) and 86% ($P < 0.01$), respectively. The alterations in acid phosphatase and zinc concentrations were not observed in the ventral prostates of isoproterenol- or propranolol-treated chemically castrated rats.

As can be seen in the Table, neither isoproterenol nor propranolol affected the body weight or the weights of the ventral prostate, seminal vesicle plus coagulating gland or the testis. However, all animals treated with dexamethasone had significantly lower body weights than the controls. Application of EDS also reduced body weight gain and, as expected, the weights of the testis and accessory sex glands.

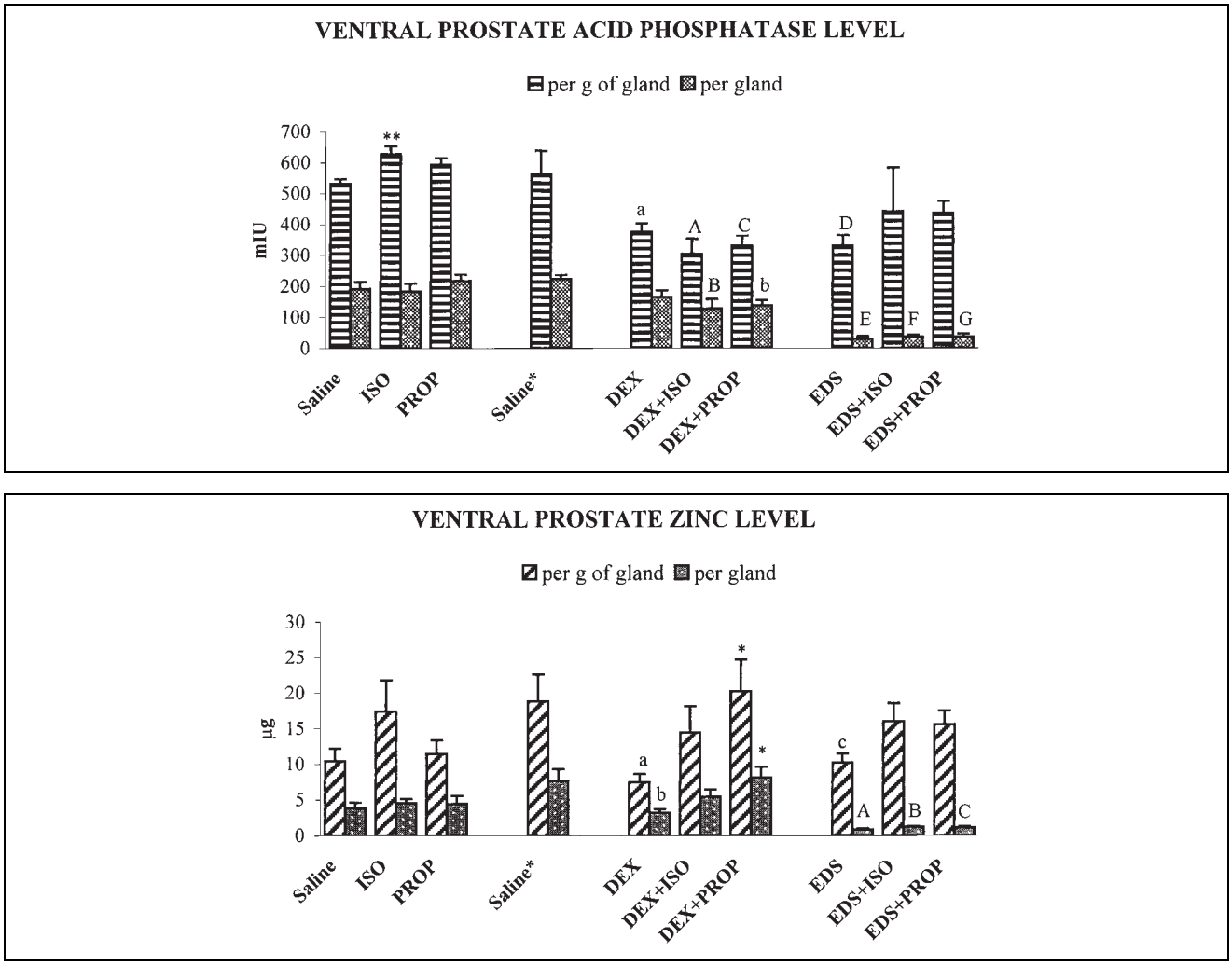

Fig.: Acid phosphatase (top) and zinc (bottom) levels in the ventral prostate of intact, dexamethasone-injected (DEX) and chemically castrated (EDS) rats treated with isoproterenol (ISO) or propranolol (PROP) for 15 days. Saline*, common control for DEX and EDS groups.
Results are expressed as means + SEM for 5 animals in each group.
*, $P < 0.05$ vs. DEX and **, $P < 0.01$ vs. Saline. Small letters, $P < 0.05$ and Capitals, $P < 0.01$ vs. Saline*

3. Discussion

The present work reveals that acid phosphatase and zinc levels in the ventral prostate of adult rats can be altered in response to β-adrenergic receptor agonist or antagonist administration, depending on the functional status of the gland.

In intact rats exposed to the β-adrenergic receptor agonist isoproterenol for 15 days, the acid phosphatase concentration in the ventral prostate was significantly increased. This finding supports the view that β-adrenergic receptors in the prostate may have a role in the synthesis of secretory proteins, prostate binding protein being one of them [14]. Such a role is also indicated by the distribution of β-adrenergic receptors in the epithelial cells of the prostate [3, 4, 12, 13].

Since isoproterenol was administered systematically, the question arises whether the observed effect on acid phosphatase concentration in the gland was direct or not. The levels of this enzyme in the prostate depend on an adequate androgen supply [23]. On the other hand, β-adrenergic receptor agonists and antagonists can alter the secretion of testosterone. Experiments with isolated Leydig cells have established clearly that one direct effect of β-adrenergic receptor activation is stimulation of testosterone secretion [24, 25], but the results obtained *in vivo* are inconclusive. Recently, we reported that isoproterenol, under the same experimental conditions, significantly decreased serum testosterone concentration and consequently evoked atrophic changes in the epithelial component of the gland in intact rats which were detected morphometrically [18]. In view of these data, the enhanced acid phosphatase concentration, described here, could not be attributed to altered serum testosterone levels.

Isoproterenol-treatment did not affect prostatic acid phosphatase in rats in which the level of this enzyme in the ventral prostate was significantly reduced either by chemical castration or by administration of dexamethasone. The same isoproterenol treatment partially prevented the morphological atrophic changes in the ventral prostate of chemically castrated rats [18], and, from the results of this work, it is obvious that it could not fully restore the secretory activity of epithelial cells. The decreased sensitivity of the ventral prostate of chemically castrated rats is presumably the result of a reduced number of β-adrenergic receptors in the gland following testosterone deprivation [4, 12, 20, 21]. It seems less likely that the density of these receptors is decreased in dexamethasone-treated rats because glucocorticoids, including dexamethasone, increase β-adrenergic responsiveness and receptor density in

Table: Body and reproductive organ weights in intact, dexamethasone-injected (DEX) and chemically castrated (EDS) adult rats treated with isoproterenol (ISO) or propranolol (PROP) for 15 days

Treatment	Body weight (g)	Reproductive organ weights (mg)		
		Testis	Ventral prostate	Seminal vesicle + Coagulating gland
Saline	356 ± 6	1218 ± 76	356 ± 32	1045 ± 111
ISO	348 ± 22	897 ± 130	290 ± 35	1015 ± 123
PROP	364 ± 9	1198 ± 111	365 ± 35	1097 ± 84
Saline*	376 ± 17	1145 ± 86	410 ± 35	1162 ± 57
DEX	304 ± 22**	1143 ± 66	445 ± 40	969 ± 152
DEX + ISO	270 ± 14**	1118 ± 76	396 ± 55	1109 ± 164
DEX + PROP	262 ± 15**	1259 ± 78	424 ± 52	975 ± 169
EDS	298 ± 30*	800 ± 89*	86 ± 19**	257 ± 54**
EDS + ISO	358 ± 14	802 ± 82	83 ± 13**	241 ± 15**
EDS + PROP	340 ± 17	903 ± 78	80 ± 20**	270 ± 69**

Saline*, common control for DEX and EDS groups.
Results are presented as means ± SEM for 5 animals in each group.
*, $P < 0.05$ and **, $P < 0.01$ vs. Saline*.

several organs through modulation of gene expression [26–28]. However, data on the effect of glucocorticoids on prostatic β-adrenergic receptors are lacking. Anyhow, the results of this and our previous work [29] are in agreement with findings showing that elevated glucocorticoid levels suppress reproductive function in males [30].

Administration of the β-adrenergic receptor antagonist propranolol for 15 days influenced zinc levels in the ventral prostate, but only in dexamethasone-treated rats. In these animals propranolol precluded the loss of glandular zinc evoked by dexamethasone. The mechanism of this effect is difficult to explain at present. It is known that zinc accumulation in citrate-producing prostate epithelial cells is regulated by testosterone and by prolactin [31]. It has also been reported that malignant prostate cells are unable to accumulate high zinc levels [32]. Here we demonstrate that both chemical castration and dexamethasone significantly decreased zinc levels in the ventral prostate. This reduction in dexamethasone-treated rats most probably is not mediated by testosterone. A shorter treatment with the same dose of dexamethasone did not influence serum testosterone levels [29] and since reproductive organ weights were unaltered by dexamethasone in the present experiments, we can assume that serum testosterone was not decreased either.

Propanolol did not affect glandular acid phosphatase levels, suggesting that β-adrenergic receptors could be involved in the control of the secretion of this enzyme only when the sympathetic innervation to the gland is stimulated. Alternatively, it is possible that the duration of β-adrenergic receptor blockade was not sufficient, because a 21-day long denervation of the rat prostate resulted in a marked decrease of secretory activity, including the acid phosphatase levels in the gland [15].

The physiological significance of the effects of β-adrenergic receptor manipulation on the rat prostate observed in this work remains to be determined. Both acid phosphatase and zinc are androgen-dependent secretory products of prostatic cells [23, 31]. However, it is possible that in conditions in which the activity of the sympathetic nervous system is enhanced, their levels in the gland may additionally be regulated by catecholamines via β-adrenergic receptors. Moreover, an excessive stimulation of these receptors might have a role in the etiology of pathological changes in the prostate.

In conclusion, manipulation with β-adrenergic receptors influences the levels of acid phosphatase and zinc in the rat ventral prostate. The responsiveness of the secretory processes to the treatment applied may be different and depends on the functional status of the gland.

4. Experimental

4.1. Experimental animals and treatments

Adult male Wistar rats, weighing 250–300 g at the beginning of the experiment, were housed under conditions of controlled temperature (19 to 21 °C) and light (12 h on, starting at 09.00 h) with constant access to a standard diet and tap water.

The rats were randomly divided into three groups for the subcutaneous (s.c.) administration of isoproterenol hydrochloride (SIGMA) or propranolol hydrochloride (ICN-Galenika) for 15 consecutive days:

I. Intact animals;
II. Animals simultaneously injected s.c. with dexamethasone (0.25 mg/kg/d; ICN-Galenika);
III. Chemically castrated rats. Chemical castration was performed with a single intraperitoneal injection of ethane dimethanesulphonate (EDS; 75 mg/kg) given at the beginning of the treatment. Since EDS is not commercially available, it was synthesized by the procedure described by Jackson and Jackson [22]. The vehicle for EDS was dimethylsulphoxide in water (3 : 1 v/v).

The daily doses of isoproterenol and propranolol were 120 μg/kg and 1.00 mg/kg, respectively. These doses have already been shown to affect the rat ventral prostate structure [16–18]. Control rats received s.c. an equal volume of sterile saline.

One hour after the final injection the rats were anaesthetized with ether, killed by decapitation and the ventral prostate, the seminal vesicles plus coagulating glands and the left testis, were dissected out and individually weighed. One lobe of each ventral prostate was frozen and kept at −70 °C until biochemical analysis.

4.2. Determination of acid phosphatase and zinc in the ventral prostate

Ventral prostate tissue was homogenized in 1 ml phosphate-buffered saline, pH 7.4, according to Wang et al. [15] using an automatic glass homogenizer. Acid phosphatase and zinc levels in the ventral prostate were determined with commercial kits DIALAB, Cat. No. 188 002 and RANDOX, Cat. No. ZN 2341, respectively.

The results are presented per g of wet weight of ventral prostate (concentration) and per gland (content).

4.3. Statistical analysis

Data are expressed as means ±SEM. Statistical analysis was carried out using one-way analysis of variance (ANOVA) followed by the least significant difference test (LSD).

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