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Experimental Study of the Efficiency of Impaza on the Model of Chronic Aseptic Prostatic Inflammation

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We compared the efficacy of Impaza (antibodies against endothelial NO-synthase in ultra-low doses) and *Serenoa repens* on the rat model of chronic aseptic prostatic inflammation. Administration of *Serenoa repens* in a dose of 50 mg/kg for 1.5 months prevented the development of prostate sclerosis and increased luminal area, but did delay the development of atrophic processes. In animals treated with Impaza (3 ml/kg for 1.5 months), atrophic changes in the prostate gland were practically absent. These findings indicate that Impaza can be used in complex therapy of abacterial prostatitis.

Key Words: *erectile dysfunction, chronic prostatitis, ultra-low doses of antibodies against endothelial NO-synthase*

Inflammatory diseases of the prostate gland (PG) are most common in men. According to recent reports, 2.2-16.0% men in North America, Europe, and Asia suffer from prostatitis. Relapses are reported in 50% patients with a history of prostatitis [6,9,12]. The risk of inflammation increases with age: by 8% every 5 years [8]. Chronic abacterial prostatitis (CAP) is more prevalent. It is diagnosed in 80-90% cases, while acute and chronic bacterial inflammation PG is diagnosed in 5-10% cases [14]. The relationship between chronic prostatitis (CP) and probability of sexual dysfunction was demonstrated [13].

Impaza is a medication containing ultra-small doses of antibodies to endothelial NO-synthase. Impaza enhances endothelial NO synthase activity, restores NO production by the endothelium during sexual stimulation, increases the levels of cyclic guanosine monophosphate (cGMP) in smooth muscles, and facilitates their relaxation, which improves blood filling of the cavernous bodies and ensures growth of intracavernous pressure (penis enlargement in different phases of the copulative cycle) and erection of sufficient strength and duration [4]. Several preclinical and clinical studies confirmed the efficacy and safety of the drug in the treatment of erectile dysfunction [2,4]. However, the use Impaza in the treatment of erectile dysfunction under conditions of CP remains an open question.

Here we studied the efficacy of Impaza treatment on the rat model of CAP.

MATERIALS AND METHODS

Experiments were carried out on 3-month-old outbred male rats (n=27) weighing 300-350 g. Inflammation was induced by applying a silk ligature on the anterior lobe of PG under anesthesia (thiopental 60 mg/kg in-

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traperitoneally). Impaza dose was chosen on the basis of previous studies of its specific activity (stimulating effect on sexual behavior) in Wistar rats, which showed the effect of the drug administered in a dose of 3 ml/kg [7]. *Serenoa repens* extract was chosen as the reference preparation because it is most widely used in clinical practice for the treatment of PG [1]. *Serenoa repens* dose was chosen on the basis of published data [11].

One month after surgery, Impaza (3 ml/kg; n=6), *Serenoa repens* (Prostamol uno, Berlin-Chemie AG / Menarini Group, 50 mg/kg; n=6), or distilled water (controls, 5 ml/kg; n=8) were given for 1.5 months. Intact animals (n=7) served as baseline. Before and after the experiment, the animals were weighed; during the experiment, general condition and weight of animals were monitored. The animals were sacrificed by cervical dislocation 2.5 months after surgery. The ligated PG lobe was dissected. The gland was weighed, and the weight index was calculated as the ratio of organ weight (mg) to body weight (g); the volume of ligated PG lobe and its density was determined. In histological sections of PG stained with hematoxylin and eosin, epithelial and luminal areas were measured. The area of collagen fibers in the connective interlayers was evaluated in histological sections stained by the method of van Gieson. The percentage of the area was calculated as the ratio between the area of structural elements and standard section area.

RESULTS

Chronic inflammatory changes in PG that lead to deterioration of general condition and weight loss developed 2.5 months after ligation (Table 1). The weight of the anterior lobe of PG, its weight index, volume and density did not differ from the baseline values (Table 1). Morphologically, prostate was characterized by hyperemia, edema, lymphocyte and macrophage infiltration. Sclerosis was revealed in PG tissue as was seen from significant increase in the relative area of collagen fibers (by 82.3%) in comparison with that in intact animals (Table 2). The relative area of acinar epithelium significantly decreased in comparison with the baseline (by 34.3%) due to developing atrophic process. The relative area of the acinar lumens in the control group was by 44.2% below the baseline value (p<0.05; Table 2), which can be due both to morphological changes and reduced secretory activity of PG.

Body weight of animals treated with Impaza or *Serenoa repens* (but not controls) did not differ from that in intact rats by the end of the experiment (Table 1). It can be assumed that treatment with Impaza and reference drug neutralized the negative effects of surgery. The weight of lateral lobe of PG, its weight index, and volume were similar in all groups. The results obtained after administration *Serenoa repens* to rats agree with the data of previous clinical studies [3], which established that *Serenoa repens* does not significantly decrease PG volume in patients with chronic inflammation of PG.

Morphological analysis suggests that the signs of inflammation such as hyperemia, edema, and cell infiltration were less frequently observed in animals treated with Impaza or reference drug than in controls. In the rats treated with Impaza, the relative area of collagen fibers tended to decrease (by 16.3% in comparison with controls). However, this parameter significantly exceeded (by 52.5%) the baseline values. Administration of *Serenoa repens* significantly (by 51%) reduced the relative area of collagen fibers in comparison with the control (Table 2). Impaza increased the relative area of acinar epithelium in comparison with the baseline values. In rats receiving the reference drug, this parameter did not differ from that in the control rats and was by 28.5% below the baseline values. In the Impaza group, the relative area of the acinar lumens did not significantly differ from that in the control. Administration of *Serenoa repens* increased this indicator by 28.5% in comparison with that in controls, but it remained below the baseline va-

TABLE 1. Effect of Course Treatment with the Test Drugs on Rat Body Weight, Weight Index, Volume, and Density of PG (M±m)

Group	Body weight at the end of the experiment, g	PG weight index, mg/g	PG volume, cm^3	PG density, g/cm^3
Baseline	512.50±11.24	1.08±0.04	0.51±0.05	1.02±0.02
Controls	489.70±10.54*	1.25±0.12	0.54±0.07	0.95±0.03
Impaza	517.36±13.06	1.08±0.06	0.57±0.04	0.93±0.04
Serenoa repens	507.27±14.35	1.09±0.08	0.51±0.06	0.98±0.02

Note. *p<0.05 in comparison with baseline.

TABLE 2. Effect of Course Treatment with the Test Drugs on Relationships between Structural Elements in PG during CP (*M*±*m*; %)

Group	Relative area, %		
	collagen fibers	acinar epithelium	acinar lumens
Baseline	3.16±0.27	25.48±3.23	57.66±2.21
Controls	5.76±0.40^{+}	16.74±1.3^{+}	32.18±1.80^{+}
Impaza	4.82±0.35^{+}	20.00±1.68	32.38±2.00^{+}
Serenoa repens	2.82±0.45*	18.22±1.7^{+}	45.70±5.29*$^{+}$

Note. $p<0.05$ in comparison with *controls, $^{+}$baseline.

lues. Morphological analysis suggests that the increase in the relative area of the acinar lumens after treatment with *Serenoa repens* can be determined by inhibition of sclerotic processes, but stimulation of secretory activity in PG cannot also be excluded.

Analysis of these findings suggests that Impaza administered to rats with CAP decelerates the development of atrophic processes in PG. In contrast to Impaza, the reference drug *Serenoa repens* inhibits the formation of collagen fibers, *i.e*. produces an antisclerotic effect, but does not prevent the atrophy of acinar epithelium. This is consistent with data that Serenoa repens extract reduces the epithelial component in PG tissue and thereby aggravates atrophic changes [10].

Thus, we can assume that the therapeutic effects of Impaza and *Serenoa repens* are aimed at different stages of CAP pathogenesis. The data obtained indicate that Impaza is advisable to use in the complex therapy of CAP including CAP accompanied with erectile dysfunction.

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