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INCREASED α_1 ADRENERGIC BLADDER SUSCEPTIBILITY IN RATS WITH BLADDER OUTLET OBSTRUCTION - A POSSIBLE CAUSE FOR IRRITABILITY

AIMS OF STUDY: Irritative bladder symptoms are the most bothersome symptoms associated with bladder outlet obstruction (BOO) derived from benign prostatic hyperplasia. Unexpectedly, these irritative symptoms can remain even after effective surgical removal of the obstruction and normalization of urine flow. Importantly, irritative bladder symptoms are rapidly relieved by α_1 adrenergic receptor (AR) antagonists. One possibility for this relief is that α_1 ARs play an important, *de novo* role in bladder contractility in obstructed patients. In the current study, we examined expression of α_1 AR subtype mRNA and receptor proteins in detrusor of control and BOO rats.

METHODS: Female rats were partially obstructed by placing a ligature (1.1 mm opening) around the urethra. After six weeks the voiding behavior was studied with a computer assisted continuous micturition recording system (CMRS). Bladders (12 controls, 12 sham operated, 9 obstructed) were harvested and snap frozen in liquid nitrogen. After tissue RNA extraction with Trizol, α_1 AR mRNA was quantified using competitive RT-PCR. Specific artificial RNA competitors, containing either α_{1a}, α_{1b} or α_{1d} AR specific priming sites, were constructed and added in known amounts to tissue RNA extracts. PCR products were stained with ethidium bromide and quantified with fluorescence image analysis. Cellular membranes were prepared from whole bladders and the membrane protein concentration determined by a bicinchoninic acid assay. The total α_1 AR protein amount was quantified by [^{125}I]-HEAT saturation binding and compared to the total α_1 AR mRNA expression.

RESULTS: The surgically obstructed rats showed 6-fold increased bladder mass after six weeks. CMRS results indicated increased micturition frequency (2x) and decreased volume per micturition (30%) in the obstructed rats as compared to sham operated and unoperated control animals. In control and sham operated animals, 71% of the α_1 AR mRNA was of the α_{1a} subtype, while 24% was α_{1d} subtype and 5% α_{1b}. In obstructed animals the opposite relationship was seen with 75% of the α_1 AR mRNA being of the α_{1d} subtype and 23% of the α_{1a} subtype; α_{1b} remained low. This change in relative subtype expression was due to a 60% reduction in α_{1a} AR expression and a 3-5 fold increase in α_{1d} mRNA expression (based on pg mRNA/g wet tissue). However, the total α_1 AR mRNA expression per tissue mass increased only slightly (22%). The [^{125}I]-HEAT saturation binding shows comparable results for the protein translation of total α_1 ARs.

	α_{1a} AR mRNA	α_{1b} AR mRNA	α_{1d} AR mRNA	Total α_1 AR mRNA	Total α_1 AR protein
Control/Sham	98±82(71%)	7±3(5%)	33±52(24%)	139±138	30±8
Obstruction	38±52(23%)	3±3(2%)	128±112(75%)	170±168	39±4
% Change	-61%	-57%	+388%	+23%	+30%

Table

Subtype specific and total α_1 AR mRNA levels (pg mRNA/g wet tissue ± SD (%)) as well as total α_1 AR protein amount (fmol receptor/mg membrane protein ± SD) in normal and obstructed rat detrusor

CONCLUSIONS: Our findings indicate a remarkable increase of α_{1d} AR gene expression in the detrusor of obstructed rats that may contribute to the changes in their micturition behavior. Although the total α_1 AR mRNA expression and protein translation does not change remarkably due to decreased expression of α_{1a} AR mRNA, the receptor subtype shift itself is causing a 10-fold increase in alpha-adrenergic bladder susceptibility, since both physiological receptor agonists (norepinephrine and epinephrine) show a 10-fold higher affinity towards α_{1d} AR than towards α_{1a} or α_{1b} ARs. The [^{125}I]-

HEAT saturation binding supports an overall increase in α_1 ARs in obstructed detrusor. Competition experiments with subtype-selective antagonists are in progress to determine if the overall increase in α_1 ARs specifically correlates with the increased α_{1d} ARs mRNA. If these findings are confirmed in the human, targeting the α_{1d} AR may provide a new therapeutic approach to controlling bladder hyperactivity associated with BOO.

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Title (type in CAPITAL LETTERS, leave one blank line before the text):

EFFECT OF CO,POST-TREATMENT WITH TADENAN(TAD) ON THE MICTURITION CHARACTERISTICS OF THE RAT STIMULATED WITH DIHYDROTESTOSTERONE(DHT)

Aims of Study

Pre-treatment with oral TAD has been shown to possess a protective effect on bladder dysfunction. In the present paper, we evaluated the functional influence of co-treatment and post-treatment with oral TAD on the frequency/volume characteristics of micturition of conscious rats, stimulated with DHT to induce experimental prostate growth.

Methods

Studies were carried out over a period of 6 weeks using 40 SD male adult rats weighing 504±22g. Animals were divided into 4 equal groups and treated daily with DHT(1.25mg/Kg/s.c. dissolved in sesame oil(SO) as vehicle) and TAD (100mg/Kg/p.o. dissolved in peanut oil(PO) as vehicle). Groups were defined as follows: [1]:Vehicle only; SO during week 1-2, PO during week 1-6; [2]:DHT in SO during week 1-2, PO during week 1-6. [3]:DHT in SO+TAD in PO during week 1-2, TAD in PO during week 3-6. [4]:DHT in SO during week 1-2, TAD in PO during week 3-6. Micturition characteristics were monitored every 2 weeks for 24 hours while the conscious rats were housed in metabolic chambers. Data were analyzed in terms of frequency(F), and max. volume per micturition(V). At the conclusion of the 6 weeks period, rats were killed and wet prostate weight measured. Values are mean±SE.

Results

The effect of treatment in the four groups on F is given by Figure 1 which shows that DHT produces a significant increase in F attaining maximum effect by the 4th week of observation. As shown by Figure 1, co-treatment with TAD, as well as post-treatment with TAD significantly ($p<0.01$)suppresses the effect of DHT at the 4th week of observation.

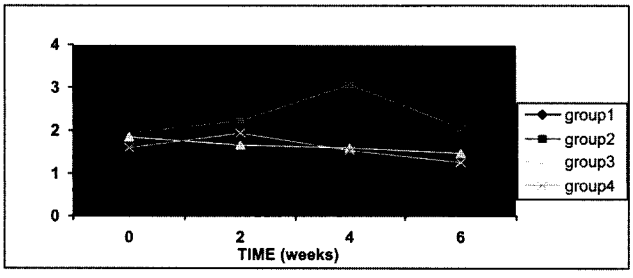

Figure 1 * $p<0.05$