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CORRELATION BETWEEN THE EXPRESSION LEVEL OF ALPHA1-ADRENOCEPTOR SUBTYPE MRNA AND PATIENT AGE OR PROSTATE VOLUME IN BENIGN PROSTATIC HYPERPLASIA PATIENTS

Hypothesis / aims of study

The expression of alpha1-AR mRNA and proteins has been quantitatively and pharmacologically characterized in selected human tissue, and all three alpha1-AR subtypes are present in the prostate. Previous quantification of alpha1-AR mRNA expression within human prostatic tissue has revealed that alpha1a-AR predominates, followed by alpha1d-AR and alpha1b-AR. Consistent with this finding, agonist binding at alpha1a-ARs mediates human prostatic smooth muscle contraction, which normally results from sympathetic nervous system stimulation producing some of the symptoms of LUTS. However, a relatively alpha1d-AR-subtype-selective antagonist, naftopidil, has shown good efficacy in decreasing symptoms and increasing urinary flow rates in patients with BPH. This shows that not only alpha1a-AR but also alpha1d-AR may play an important role in the alpha1-AR-mediated smooth muscle contraction in the prostate. Real-time RT-PCR is becoming a widely used method to quantify the gene expression from cells, tissues, or tissue biopsies Compared to the previously used methods, the real-time RT-PCR method we used is considered to be a very rapid, sensitive, reliable, accurate and reproducible method for determining mRNA levels in tissue or cells than other methods, and appears particularly suitable to quantify gene expression in large series of samples.In order to confirm the expression level of each alpha1-AR subtype in a large number of BPH patients, we used a real-time RT-PCR method for routine quantification of each alpha1-AR subtype expression level. The specific role of each alpha1-AR subtype in regulating prostatic smooth muscle function has not been established, and it is unknown whether they are affected by age and prostate volume. We also examined whether age and prostate volume influence human prostate alpha1-AR subtype expression.

Study design, materials and methods

Enrolled in our study were 75 men who were 50 years old or older (mean age; 69.0+/-6.9 years) with LUTS secondary to untreated BPH diagnosed at the outpatient urology clinic of our institution during 2000 to 2006. Diagnostic transperineal ultrasound prostate needle biopsies were performed, and pathologically no malignancies were found. Excepting diagnostic biopsy specimens for malignancy or benign, four biopsy specimens were obtained from the transition zone to examine the expression level of alpha1a-, alpha1b- and alpha1d-AR mRNA by real-time (TaqMan) RT-PCR. TaqMan PCR for each alpha1-AR subtype was performed by an Applied Biosystems PRISM 7700 sequence detection system using prostate biopsy specimens to quantify the expression level of each alpha1-AR subtype. To quantify the results obtained by TaqMan RT-PCR, we used the standard curve method. Amplification plots for alpha1-AR subtypes and housekeeping genes, beta-actin, were constructed from fluorescence emission data collected during PCR amplification. The copy number of each alpha1-AR subtype and beta-actin mRNA in an unknown sample was quantified by preparing a standard curve using dilutions of a known amount of the respective cDNA plasmid. Significance of differences of median expression levels among each alpha1-AR subtype was determined by the Kruskal-Wallis test and Scheffe's method. The correlation between the expression levels of each alpha1-AR subtype and each patient age or prostate volume was evaluated with the Pearson correlation test.

Results

The median expression level (interquartile range) was 1.24 (0.66-2.32), 0.16 (0.10-0.33) and 1.11 (0.75-2.27) copies/beta-actin for alpha1a-, alpha1b- and alpha1d-AR mRNA, respectively. Although there were statistically significant differences between the median expression levels of alpha1a-AR or alpha1d-AR mRNA and alpha1b-AR mRNA ($p < 0.0001$), there were no significant differences between the median expression levels of alpha1a-AR and alpha1d-AR mRNA. However, the expression levels were different from patient to patient.The patients were divided into those who showed predominance of alpha1a-AR mRNA expression levels (n=36) and those who showed predominance of alpha1d-AR mRNA expression levels (n=39). There were no patients who expressed predominantly the alpha1b-AR subtype. The expression levels of alpha1a-AR, alpha1d-AR and total alpha1-AR mRNA showed a significant positive correlation with patient age ($p < 0.01$, r=0.32, 0.31 0.33, respectively. However, the correlation of the expression levels of alpha1a-AR, alpha1b-AR, alpha1d-AR and total alpha1-AR mRNA with prostate volume was insignificant (r=-0.19, -0.12, -0.09 and -0.12, respectively).

Interpretation of results

We demonstrated that there were patients with a subtype predominantly alpha1a-AR and those with a subtype predominantly alpha1d-AR. Our results showed that, in the prostate, the expression levels of alpha1a-AR, alpha1d-AR and total alpha1-AR mRNA correlated significantly in a positive way with patient age, but did not correlate with prostate volume.

Concluding message

The difference in the expression of the alpha1-AR subtype with the patient may be the cause of the difference in the effectiveness of several subtype selective alpha1-AR antagonists from patient to patient. The increase of expression levels alpha1a-AR, alpha1d-AR and total alpha1-AR mRNA with age may lead to increase of prostatic urethral pressure and LUTS in elderly men, and may be an important factor in the pathogenesis of clinically significant BPH. This may be one of the causes that LUTS associated with BPH does not always depend on prostate volume.