Is Muscarinic Receptor mRNA Expression in the Human Prostate Associated with Genotype for the M$_2$ Receptor?

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Muscarinic acetylcholine receptors are highly expressed in the human prostate (Eur Urol 54: 326, 2008) but little is known about their regulation. A CA repeat polymorphism has been described in the 5' untranslated region of the human M$_2$ receptor which is associated with a marked greater receptor expression in human airway smooth muscle cells (Am J Respir Cell Mol Biol 30: 678, 2004). Therefore, we have explored the expression of M$_2$ receptor mRNA in the human prostate relative to genotype for this polymorphism; mRNA for the other four human muscarinic receptor subtypes was used as an internal control.

Prostate and blood samples were obtained with informed patient consent and approval from the local ethical committee from 101 consecutive patients (age 68.6 ± 0.8 years, prostate volume 82.6 ± 4.5 ml) undergoing transurethral resection of the prostate for treatment of benign prostatic enlargement or obstructive prostatic carcinoma. DNA was isolated from the blood samples, and genotype for the CA repeat polymorphism of the M$_2$ receptor was determined by capillary gel electrophoresis. mRNA for all five muscarinic receptor subtypes was isolated from the prostate samples and quantified by real-time PCR. After testing several housekeeping genes for potential variability, RPLP0 was chosen as the reference gene for our study. Expression of a given receptor subtype mRNA was determined as ∆∆C$_t$ value relative to RPLP0 expression and the mean of the tested population. Patient age, pathological diagnosis (benign enlargement vs. carcinoma), prostate size and potentially interfering medications were explored as possible confounding factors. All data are means ± SEM. Statistical significance of differences between groups was tested by ANOVA, and significance of associations by linear regression analysis with p < 0.05 considered as significant.

Genotyping for the CA polymorphisms detected alleles with 6, 11, 12, 13, 14 and 15 repeats with overall allele frequency being similar to previously reported data. The three most frequent genotypes were 13/13, 13/14 and 11/13 in 28%, 24% and 13% from tested population, respectively.

mRNA expression of the M$_2$ receptor was not associated with age, pathological diagnosis, prostate volume or co-medication to a clinical relevant or statistically significant extent. None of the genotypes was associated with a meaningful or statistically significant difference in M$_2$ receptor expression (∆∆C$_t$ value 0.14 ± 0.35, 0.08 ± 0.41 and 0.12 ± 0.65 for 13/13, 13/14 and 11/13, respectively. Moreover, genotype for the M$_2$ receptor CA repeat polymorphism also was not associated with differences in expression of M$_1$, M$_3$, M$_4$ or M$_5$ muscarinic receptor mRNA.

We conclude that in human prostate, in possible contrast to human airways, genotype for the M$_2$ receptor CA repeat polymorphism is not associated with differences of expression of any of the muscarinic receptor subtypes.