

**SCA 128****NOVEL HUMAN ALPHA1A-ADRENERGIC RECEPTOR SINGLE NUCLEOTIDE POLYMORPHISMS ALTER RECEPTOR BIOLOGICAL FUNCTION**

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The  $\alpha_{1a}$ -adrenergic receptors ( $\alpha_{1a}$  ARs) are present in a wide variety of human tissues including brain, liver, prostate, vascular smooth muscle and myocardium. Nine single nucleotide polymorphisms (SNPs) in coding region of human  $\alpha_{1a}$  AR have been identified in our laboratory, 7 of which induce an amino acid change. We hypothesize that these SNPs could induce changes in receptor biological functions, which may influence variations in sympathetically-mediated diseases. To investigate the effect of these SNPs on receptor function, we initially introduced each SNP into the human  $\alpha_{1a}$  AR by site-directed mutagenesis and expressed these receptors at 1.5-2.4 pmol/mg in rat-1 fibroblasts. In the current study, we tested receptors with each SNP at lower, more

physiologic expression levels (0.21-0.44 pmol/mg) for ligand binding, receptor inositol phosphate (IP) signaling and cell growth. Compared with wild type receptors, 2 rare ( $P = 0.002$ ) SNPs in transmembrane region (TM) 4 and 7, respectively, caused a 3-fold decrease in binding affinity for agonists norepinephrine (NE), epinephrine and phenylephrine. While not influencing maximal IP activity, the SNP in TM7 resulted in a 3-fold reduction in NE potency stimulation of IP production (EC50:  $132.10 \pm 27.07$  vs  $47.10 \pm 2.58$   $\mu$ M). This same SNP (TM7) also caused a 3-fold increase in affinity for antagonist 5-methyurapidil while another SNP in TM5 ( $P = 0.005$ ) caused a 3-fold loss of affinity for antagonist phentolamine. Interestingly, a fourth SNP in the third intracellular loop ( $P = 0.003$ ) did not affect ligand binding, but significantly increased maximal receptor IP activity (2.2-fold) and stimulated cell proliferation (2.3-fold). In conclusion, at relatively, physiologic expression levels, 4 naturally occurring human  $\alpha_{1a}$  AR SNPs induce altered receptor biological activity; this finding has potentially important implications in many areas of medicine.