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EFFECTS OF KRP-197, MUSCARINIC RECEPTOR ANTAGONIST ON K+ RELEASE OF THE SALIVERY GLAND AND BLADDER CONTRACTION IN THE RAT

Aims of Study

We previously reported that KRP-197/ONO-8025 (4-(2-methylimidazolyl)- 2,2- diphenylbutyramide) *in vitro* and i*n vivo* is a potent M1&M3 receptor subtype selective antagonist (27th Annual Meeting of ICS (1997)), and inhibits in the dose-dependent manner [^{3}H]-ACh outflow evoked by electrical field stimulation (EFS) in physostigmine-treated bladder muscle strips (30th Annual Meeting of ICS (2000)). The present study was undertaken to examine the effects of KRP-197 on potassium ion (K+) release of salivary gland (indicator for volume secretion *in vitro*) and contractile response induced by electrical field stimulation (EFS) in the bladder smooth muscle treated with physostigmine in the rat.

Methods

Bladder smooth muscle strips obtained from male rats were mounted in the thermostatically controlled organ bath for isometric tension developed by EFS applied usually at a frequency of 10 Hz using rectangular waves 40 V, 0.5msec. K^+ release from the rat submandibular strips induced by acetylcholine (ACh, $3x10^{-6}$ mol/L) and EFS was measured by means of a mV-meter, which monitored the voltage difference between a potassium selective electrode and a reference. Electrical stimulation consisted of 300 shocks applied at a frequency of 30 Hz using monophasic rectangular waves 40 V, 1msec. in salivary gland.

Results

EFS- and ACh-induced K^+ release from rat salivary gland strips were dose-dependently inhibited by the treatment with KRP-197, muscarinic receptor subtype selective and non-selective antagonists (pirenzepine, methoctramine, darifenacin and atropine) and antipollakiuric drugs (oxybutynin, tolterodine , propiverine). The inhibitory concentration (IC_{50}) of cholinergically evoked K^+ release was well correlated with the affinity for M3 subtype receptor, but not M1 and M2. Oxotremorine and McN-A-343 inhibited EFS–induced K^+ release, but these responses were not completely antagonised by the treatment with methoctramine. EFS–induced bladder twitch contractions in physostigmine-treated muscle were inhibited by the treatment with KRP-197 and pirenzepine at more than 10^{-10} and 10^{-8} mol/L, respectively. Furthermore, frequency contraction curve in detrusor muscle strips shifted to the left by physostigmine without affecting maximum contraction developed, and this shift to the left was reversed by pirenzepine.

Conclusions

These data suggest that K^+ release in salivary gland is produced through Ca^+ -dependent, neurogenic and cholinergic neurotransmissions, muscarinic receptor subtypes M2 and M3 possibly mediating ACh-release modulator (inhibitory) and volume secretion (excitatory), respectively, and that bladder contraction is primarily mediated by M3 muscarinic receptors, whereas M1 are involved in contractions by means of ACh release, probably prejunctionally. Therefore, drugs acting at M1 and M3 muscarinic receptors, e.g. KRP-197 might be expected to be more effective for the treatment of overactive bladder disorders with fewer effects in the salivary glands.