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FUNCTIONAL BLADDER SELECTIVITY OF SVT-40776: A COMPARATIVE STUDY.

Hypothesis / aims of study

SVT-40776 is a novel quinuclidine derivative highly selective for human M3 over M2 receptors [1,2,3], that is currently on Phase II clinical testing for overactive bladder treatment. Meanwhile M2 receptors regulate cardiac function, M3 subtype plays a predominant role in bladder smooth muscle (BSM) contraction through the activation of phosphoinositide breakdown. The salivary glands (SG) are known to be abundant in M3 receptors, whose activation is crucial for salivary secretion. Thus, dry mouth is one of the most common adverse reactions induced by muscarinic antagonists.

The aim of this study was to comparatively investigate the effect of SVT-40776, solifenacin, oxybutynin and tolterodine on the inositol phosphate accumulation in response to muscarinic receptor activation in BSM and SG.

Study design, materials and methods

The concentration of inositol-4-monophosphate (Ins4P), as a measure of phosphoinositide breakdown, was determined as follows: mice SG and BSM suspensions were labelled with [^{3}H]-*myo*-inositol for 2 h at 37ºC. Tissues were preincubated for 20 min with antagonists, before the addition of carbachol (CCh, 10 µM), and then further incubated for 1 h at 37ºC in the presence of lithium chloride (10 mM). The reaction was stopped with a mixture of chloroform/methanol, and the phases were separated with chloroform and water. After centrifugation, the organic layer was taken for radioactivity measurement. The aqueous layer was applied to Bio-Rad AG 1x8 columns and radioactivity in the last eluate was determined.

Results

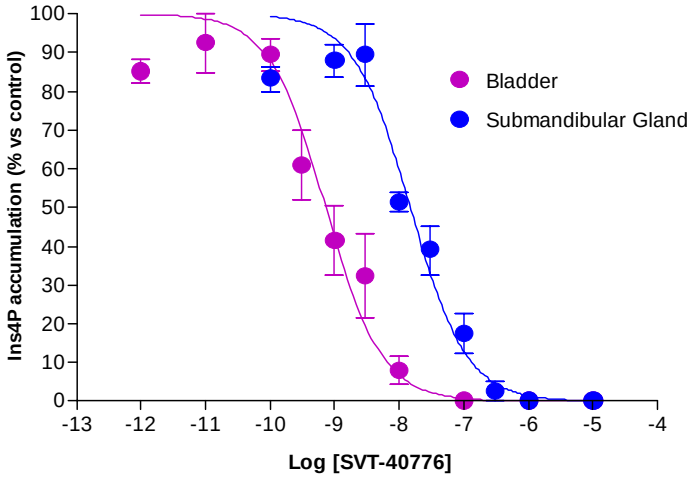
CCh stimulates Ins4P production on SG and BSM with EC50s of 4.1 and 3.6 µM respectively. The obtained results, compared with previously published binding affinities, are summarized in the following Table and Figure:

Table. Affinity constants (Ki, nM) on CCh-stimulated [3H]-Ins4P accumulation.

Compound	Binding hM3	**SG**	**BSM**	**Ratio GS/BSM**
SVT-40776	0.18	**4.3 (3.2-5.8)**	**0.19 (0.13-0.27)**	**23**
Solifenacin	7.3	**3.4 (2.2-5.1)**	**3.3 (2.4-4.8)**	**1.0**
Tolterodine	4.13	**12.5 (7.2-21.7)**	**3.4 (2.4-4.8)**	**3.7**
Oxybutynin	1.6	**38.8 (27.5-54.6)**	**16.1 (11.0-23.5)**	**2.4**

Each value denotes the mean of n=4-8 experiments. Confidence intervals are expressed in parentheses. The in vitro binding to human M3 receptor expressed in CHO cells is included as a measure of comparison

Figure. Inhibition of CCh (10 µM)-mediated functional responses in mice BSM and SG by SVT-40776



Data are expressed as percent maximum response (mean±SEM, n=4-8)

Interpretation of results

- CCh caused a time- and concentration-dependent accumulation of Ins4P in mice SG and BSM.
- SVT-40776 inhibited the CCh-induced Ins4P accumulation in BSM, exhibiting an excellent correlation between Ins4P inhibition and its human M3 receptor affinity.
- SVT-40776 inhibited less markedly this functional response in SG than in BSM, indicating a very favourable selectivity index between both tissues (23-fold).
- Ratios showed by solifenacin (1.0), tolterodine (3.7) and oxybutynin (2.4) suggest a lack of tissue selectivity with these agents.
- Further studies are required to explore the mechanism underlying the selective profile of SVT-40776.

Concluding message

SVT-40776, but not solifenacin, tolterodine or oxybutynin, shows high and relevant bladder selectivity in a functional assay comparing muscarinic blockade on submandibular gland vs. bladder muscle. This particular profile of functional response may be indicative of the influence of cell/tissue background upon the pharmacology of the M3 receptor.

These findings strongly suggest a promising therapeutic profile of SVT-40776 based its reduced dry mouth potential.

References

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