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THE EFFECTS OF PILOCARPINE, AN M3-MUSCARINIC RECEPTOR AGONIST, ON CONTRACTION OF THE PORCINE URINARY BLADDER IN VITRO

Hypothesis / aims of study
Contraction of urinary bladder is mediated by activation of muscarinic receptors, with M3-muscarinic receptors predominate. Cholinergic drugs such as bethanechol chloride have been considered to enhance detrusor contractility and promote bladder emptying in patients with underactive bladder. However, the use of cholinergic drugs has not been standardized due to the efficacy and serious side effects. Recently, pilocarpine, a M3-muscarinic receptor agonist has been reported to be effective for the treatment of dryness of eyes or salivation disorders[1]. This study examines the effects of pilocarpine on contraction of porcine urinary bladder.
Study design, materials and methods
Pig urinary bladder was collected from the abattoir and strips of tissues were mounted in 10ml organ baths containing Krebs solution (composition in mM: NaCl 118.4, KCl 4.7, CaCl2 1.9, NaHCO3 24.9, MgSO4 1.15, KH2PO4 1.15, glucose 11.7) which was maintained at 37oC and continuously gassed with 95% O2 and 5%CO2. The tissues were subjected to a resting tension of 1 g and allowed to equilibrate for 60 minutes. Cumulative concentration-response curves (CRCs) to pilocarpine were obtained, with Krebs solution containing in the presence of darifenacin, 4-DAMP (M3 selective antagonist), pirenzepine (M1 selective antagonist), methoctramine (M2 selective antagonist), or in the presence of vehicle. These muscarinic receptor antagonists were treated for 30 minutes before the addition of pilocarpine. Each tissue was used only to construct one CRC to pilocarpine with or without one concentration of a muscarinic antagonist, and affinity values for each antagonist were calculated.
Results
Pilocarpine induced contractions of smooth muscle of the detrusor in a concentration-dependent manner, with maximum contraction relative to 80 mM KCl of 134.4±22.3% and pEC50 values of 5.28±0.26. Darifenacin, 4-DAMP, pirenzepine, and methoctramine caused surmountable antagonism of responses to pilocarpine, with slopes of Schild plot of 1.37±0.20, 0.80±0.54, 1.05±0.30, and 0.91±0.35, respectively. The rank order of mean pA2 values was as follows: 4-DAMP (8.79±0.27) = darifenacin (8.73±0.06) > pirenzepine (6.72±0.12) > methoctramine (6.58±0.16).
Interpretation of results
These data suggest that pilocarpine produced contraction of the pig bladder, and that the affinity of M3-muscarinic receptor subtype antagonist was the highest on CRCs to pilocarpine in the porcine detrusor muscle.
Concluding message
Pilocarpine appears to produce contraction of the pig bladder through activation of M3-muscarinic receptor.
References
1. JAMA. 2010; 304(4): 452-60

Specify source of funding or grant	**None**
Is this a clinical trial?	**No**
What were the subjects in the study?	**ANIMAL**
Were guidelines for care and use of laboratory animals followed or ethical committee approval obtained?	**No**
Statement that no ethical approval was needed	**Tissue was taken from dead animal**