

Sildenafil and doxazosin combination: relaxant effects in human corpus cavernosum

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OBJECTIVES

Erectile dysfunction (ED) and symptomatic benign prostatic hyperplasia (BPH) are highly prevalent in aging men. Both conditions are often associated. Phosphodiesterase 5 inhibitors (PDE5I) such as sildenafil are the first line treatment of ED. Alpha1-adrenoceptor antagonists (α -blockers) such as doxazosin are indicated for the treatment of patients with BPH associated lower urinary tract symptoms (LUTS). Combined use of PDE5I and α -blockers is becoming more common.

The study aim was to evaluate the relaxant effects of the combination of sildenafil with doxazosin on human corpus cavernosum

In vitro isometric tension studies using human corpus cavernosum tissue were performed to:

- evaluate the effect of sildenafil, doxazosin or a combination of both on norepinephrine (NE)-induced contractions
- evaluate the potential enhancing effect of doxazosin on the relaxation induced by sildenafil on NE precontracted strips.

MATERIALS & METHODS

Human cavernosal strip preparation

Human erectile tissues were obtained from the corpus cavernosum of patients (N=7) undergoing penile surgery for penile implant. Cavernosal strips were suspended in 5 ml organ chambers filled with Krebs-HEPES buffer containing 118 mM NaCl; 4.7 mM KCl; 1.2 mM MgSO₄; 1.2 mM KH₂PO₄; 2.5 mM CaCl₂; 4.2 mM NaHCO₃; 11.1 mM glucose, and 20.8 mM HEPES. Organ chambers were maintained at 37°C and continuously bubbled with 95% O₂ and 5% CO₂ to maintain a pH at 7.4.

In vitro contractile experiments

Following an equilibration period, the erectile tissues were primed by the addition to the organ bath of KCl (80 mM, 10 min) and after washings, by the addition of NE at 10⁻⁶ M during 5 min followed by acetylcholine (ACh) 10⁻⁵ M to check for endothelial integrity. After the priming period, the strips were washed by fresh Krebs-HEPES solution and allowed to re-equilibrate.

In a first set of experiments, the contraction of cavernosal strips to NE was described through concentration-response curves (CRC) to NE from 10⁻⁹ to 10⁻⁵ M obtained on each strip. Then after a 20-min incubation period with vehicle, or sildenafil (10⁻⁶ M), or doxazosin (10⁻⁷ M) or a combination of both (at these concentrations), CRC to NE was repeated.

In a second set of experiments, strips were preincubated during 20 min with doxazosin (10⁻⁹ or 10⁻⁸ M) or vehicle, then CRC to sildenafil (from 10⁻⁹ to 10⁻⁶ M) were constructed on NE (3.10⁻⁶ M) precontracted human cavernosal strips.

Data Analysis

For the first set of experiments, results of the 2nd CRC to NE were expressed in percentage of the maximal value obtained during the first CRC. For the second set of experiments, values were expressed in percentage of inhibition of the maximal response to NE. For each CRC, a pD₂ value and a mean maximal effect (Emax) were evaluated in each experimental condition. Data were expressed as mean ± SEM for N experiments corresponding to N = corpus cavernosum samples. Statistical analysis is performed with GraphPad Prism® 4.03 software.

CONCLUSIONS

- Sildenafil and doxazosin have additive relaxant effects on NE-induced contractions of isolated human corpus cavernosum.
- These experiments provide support for further clinical evaluation of the sildenafil and doxazosin combination in ED patients with BPH.

RESULTS

Effect of sildenafil, doxazosin or a combination of both on norepinephrine-induced contraction on human cavernosal strips

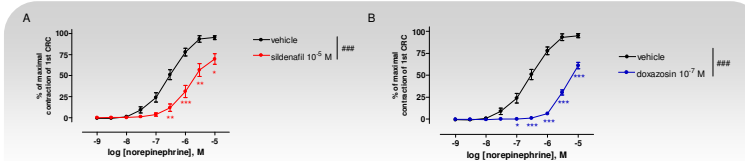


Figure 1: CRCs of NE were performed in presence of vehicle (N=8) or (A) sildenafil 10⁻⁶ M (N=7) or (B) doxazosin 10⁻⁷ M (N=7) on human cavernosal strips. The data are mean ± SEM. ###p<0.001, two-way ANOVA with interaction followed by a modified Student t test with the Bonferroni adjustment for multiple comparisons: *p<0.05, **p<0.01, ***p<0.001.

	pD ₂	Emax ^{max} (%)	N
vehicle	6.56±0.11	93.7±2.1	8
sildenafil 10 ⁻⁶ M	5.74±0.13	69.7±6.1 ^{†††}	7
doxazosin 10 ⁻⁷ M	N.D	61.0±3.6 ^{†††}	7
sildenafil 10 ⁻⁶ M+ doxazosin 10 ⁻⁷ M	N.D	29.3±8.9	8

Table 1: Potency (pD₂) and maximum response (Emax) values of NE-induced contractions on human cavernosal strips in presence of both sildenafil and doxazosin. ***p<0.001, one-way ANOVA followed by Newman-Keuls posttest, ^{†††}p<0.001 versus combination. ND: could not be determined.

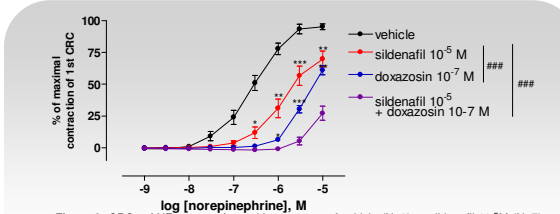


Figure 2: CRCs of NE were performed in presence of vehicle (N=8) or sildenafil 10⁻⁶ M (N=7) or doxazosin 10⁻⁷ M (N=7) or sildenafil 10⁻⁶ M+ doxazosin 10⁻⁷ M (N=8) on human cavernosal strips. The data are mean ± SEM. ###p<0.001, two-way ANOVA with interaction followed by a modified Student t test with the Bonferroni adjustment for multiple comparisons: *p<0.05, **p<0.01, ***p<0.001.

The combination of sildenafil and doxazosin exerted a greater inhibitory effect on norepinephrine-induced contractions of human cavernosal strips compared to each compound alone.

Effect of doxazosin on the relaxation induced by sildenafil on norepinephrine precontracted human cavernosal strips

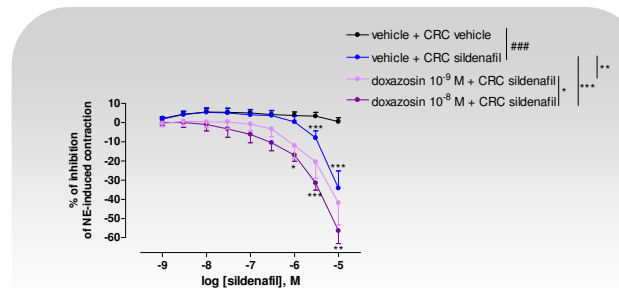


Figure 3: CRC of sildenafil or vehicle (N=7) were performed in presence of vehicle (N=7) or doxazosin 10⁻⁹ M (N=5) or doxazosin 10⁻⁸ M (N=7) on human corpus cavernosum. The data are mean ± SEM. ###p<0.001, two way with interaction followed by a modified Student test with the bonferroni adjustment for multiple comparison; *p<0.05, **p<0.01, ***p<0.001, two-way ANOVA followed by Bonferroni post-test; †p<0.05; **p<0.01; ***p<0.001.

	pD ₂ ^a	Emax (%) ^b	Pre-contraction values of NE (mg) ^c	N
vehicle+vehicle	ND	-0.41±2.0	2072.7±390.5	7
vehicle+sildenafil	5.4±0.1	-34.2±8.9	2322.0±485.8	7
doxazosin 10 ⁻⁹ M+sildenafil	5.6±0.1	-41.9±11.5	2036.4±393.7	5
doxazosin 10 ⁻⁸ M+sildenafil	5.7±0.1 [†]	-56.5±6.7	2333.2±293.7	7

Table 2: Potency (pD₂), maximum response (Emax) values of sildenafil-induced relaxation on norepinephrine-induced contractions with or without doxazosin preincubation on human cavernosal strips. ^ap<0.05, one-way ANOVA followed by Newman-Keuls posttest, ^bp<0.01 versus vehicle+sildenafil; ^csp=ns, one-way ANOVA.

Preincubation with doxazosin enhanced in a dose dependent manner the relaxant effect of sildenafil on norepinephrine-precontracted cavernosal strips