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1. Sanofi-Synthelabo Recherche

ACTIVATION OF 5-HT7 RECEPTORS IS LIKELY TO BE RESPONSIBLE FOR 5-HT EFFECTS IN THE RAT URINARY BLADDER

Aims of Study

It is known that 5-HT produces potentiation of the neurogenic response of the rat isolated detrusor muscle and bladder contraction in the anesthetized rat (1, 2). We thus decided to characterize pharmacologically the 5-HT receptors involved.

Methods

Female Wistar rats (250-350g) were used. *In vitro*: detrusor muscle strips were mounted between two platinum electrodes in 5 ml organ bath filled with a modified Krebs solution (1 µM propranolol, 37°C, 95% O_2 / 5% CO_2). Tissues responses were measured using isometric strain gauges connected to a data acquisition system. After 1 hour of equilibration and a contraction to 80 mM KCl, strips were submitted to electrical field stimulation (EFS, 5 Hz, 50 V, 0.3ms pulse duration, trains of 10 s every min). After stabilization of the EFS-induced contractions, tissues were incubated for 30 min with antagonist or vehicle, then a cumulative 5-HT concentration-effect curve (CRC) was obtained. CRC obtained following antagonist incubation was compared with CRC obtained in vehicle-treated strips from the same animal. 5-HT effects were expressed as percentage of the contractile response to 80 mM KCl.
In vivo: Rats were anesthetized with pentobarbital (30 mg/kg ip). The ureters and urethra were ligated, the bladder catheterized through the dome and infused with saline to obtain an intra-vesical pressure (IVP) between 10-20 cmH_2O. In preliminary experiments we found that 5-HT at 3, 10, 30 and 100 µg/kg i.v. dose-dependently increased IVP. 5-HT 30 µg/Kg i.v. was administered every 10 min three times (control value), then one dose of antagonist was perfused during 5 min and, after further 5 min, 30 µg/kg iv 5-HT was tested again. This cycle was repeated 4 times using increasing doses of the same antagonist. Results are expressed as % inhibition of IVP in comparison to control values (means of three consecutive responses to 30 µg/kg i.v. of 5-HT).

Results

In vitro: In basal conditions, 5-HT (0.01-30µM) did not induce any contractile effect. EFS produced contractile responses totally abolished by 1µM TTX. 5-HT (0.01-100 µM) induced a concentration-dependent enhancement of this neurogenic response. The effect of selective antagonists for several 5-HT receptor subtypes on the 5-HT-induced potentiating effect is illustrated in the annexed Table.
In vivo: In the presence of the antagonist vehicle, the increases in IVP induced by repeated doses of 30 µg/kg 5-HT were reproducible. R(+) Lisuride (LISU) tested in the range 3-100 µg/kg iv, dose-dependently inhibited 5-HT induced increase of IVP. At the maximal dose tested, LISU (n=5) almost totally inhibited the 5-HT effect (-95±2.4%); the calculated antagonist dose needed to inhibit 50% of the response to 30 µg/Kg 5-HT was 9.2±2.1 µg/kg i.v.

Conclusions

These results suggest the involvement of 5-HT_7 receptors in stimulated rat isolated detrusor muscle and in the bladder of anesthetized rats for the following reasons: a) The selective 5-HT_7 receptor antagonist SB 258741 blocks the 5-HT potentiating effect with the expected potency (3); b) pA_2 values obtained for LISU and mesulergine correlate with their potencies on 5-HT_7 receptors (4,5) but not with the potencies on 5-HT_{2a} /5-HT_{2c} receptors (6); c) *in vivo* results show that LISU inhibits 5-HT effect on IVP at doses known to abolish vasodilatation mediated by 5-HT_7 receptors in anesthetized Wistar rats (7).
In conclusion, we have found that 5-HT_7 receptor antagonists are active against increased bladder contractility in *in vitro* and *in vivo* models, and thus could be useful in the treatment of detrusor overactivity.

Table: pD_2 values of 5-HT in electrically-stimulated rat detrusor muscle strips and effects of several selective 5-HT receptor antagonists

pD_2 5-HT	*Emax (%80mM KCl)*	Antagonist tested	*Receptor subtype*	*Concentration tested (µM)*	*pA_2 value*
6.38±0.08 (n=3)	30.7± 3.6	1-[4-(1-Adamantane carboxamido)butyl]-4-(2-methoxyphenyl)piperazine	5-HT_{1A}	0.1	No effect
6.09 ± 0.06 (n=4)	45.2± 3.6	**Mesulergine**	5-HT_2 / 5-HT_7	0.3	**7.27±0.12** (n=4)
6.05 ± 0.27 (n=4)	36.7± 13	Ondansetron	5-HT_3	0.1	No effect
6.23 ± 0.13 (n=5)	27.1± 5.0	SB204070	5-HT_4	0.03	No effect
5.93 ± 0.11 (n=4)	37.4± 2.1	**R(+) Lisuride**	5-HT_7	0.3	**7.73± 0.45** (n=4)
6.27 ± 0.12 (n=8)	36.1± 4.4	**SB258741**	5-HT_7	0.3	**7.32± 0.22** (n=8)

References

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