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DULOXETINE COUNTERACTS BLADDER OVERACTIVITY INDUCED BY INTRAVESICAL ACETIC ACID IN ANESTHETIZED FEMALE GUINEA-PIGS THROUGH ACTIVATION OF 5-HT RECEPTORS.

Hypothesis / aims of study

A similar innervation of urethral sphincter and bladder in guinea-pigs, cats and humans compared to rats (1) has been reported. Since in cats, overactive bladder induced by acetic acid was reversed by duloxetine (2), the aim of this study was to (i) evaluate the effects of duloxetine on cystometric parameters in overactive bladder induced by intravesical infusion of acetic acid (AA), (ii) to determine the mechanism of action of duloxetine in anesthetized female guinea-pigs.

Study design, materials and methods:

Female Dunkin-Hartley guinea-pigs were anesthetized with urethane (1.5 g/kg, i.p.). A catheter was implanted into the bladder through the dome and another one into the jugular vein for drug administration. A plastic tube was inserted into the trachea. Urinary bladder was continuously infused at the rate of 12 ml/hr with AA 0.2% or saline. In a first series of experiments, after a 30 min control period following the first micturition, duloxetine (0.1, 0.3, 1, 3 or 5 mg/kg), fluoxetine (10 mg/kg) or vehicle (saline) was intravenously (i.v.) infused during 5 min (n=6-8 per group). The effect of duloxetine on bladder pressure was observed during 1 hour after administration. Duloxetine (3 mg/kg) was also tested using the same protocol in guinea-pigs with intravesical infusion of saline.

In a second series of experiments, a 5-HT receptor antagonist, methiothepin (1 mg/kg, i.v., n=6), an α_1-adrenoreceptor antagonist, prazosin (1 mg/kg, i.v., n=7) or their vehicle (n=7) was administered 5 min before duloxetine (3 mg/kg, i.v.) in female guinea-pigs with AA 0.2% bladder infusion. The effects of duloxetine were observed during 1 hour after administration.

Each cystometric parameter, Intercontraction interval (ICI), Threshold Pressure (ThP), Amplitude of Micturition (AM) and Basal Pressure (BP), was measured and averaged per 30 min period post administration. For each group, the mean of ICI, ThP, AM and BP was compared to basal values (3 micturition cycles before drug administration) using a one way Anova with repeated measures followed by Newman-Keul's test. Basal values of each cystometric parameters for each group were compared using a one way Anova followed by Newman-Keul's test. A P value < 0.05 was considered to be statistically significant.

Results

Intravesical infusion of AA 0.2% elicited a significant and reproducible decrease in ICI in AA 0.2% groups. This decrease was closed to 50 % compared to basal values of ICI in saline group ($p<0.001$, table 1). No relevant effect on all others cystometric parameters was noticed. Intravenous administration of vehicle did not modify ICI values in guinea-pigs with AA 0.2% bladder infusion, whereas it induced an increase in ICI in guinea-pings with saline bladder infusion. Duloxetine reversed partially and significantly, in a dose dependent manner the decrease in ICI induced by AA bladder infusion, starting from the dose of 0.3 mg/kg with a maximal effect at the dose of 5 mg/kg, reaching -43 ± 8 % of the basal value ($p<0.05$, table 1). Fluoxetine at the dose of 10 mg/kg i.v. also increased ICI to values similar to those induced by duloxetine at 3 mg/kg ($p<0.001$, table 1) and was without effect on all others cystometric parameters. The effect of duloxetine on ICI was completely antagonized by methiothepin, but not by prazosin ($p<0.01$, table 1).

Table 1: Effects of vehicle, duloxetine, fluoxetine and combination of duloxetine with methiothepin or prazosin on ICI in anesthetized female guinea-pigs.

Intravesical infusion	Treatment (mg/kg, i.v.)		ICI (sec)		
			basal	0-30 min	30-60 min
Saline	Vehicle		540±112	656±92 *	712±115 **
AA 0.2%	Vehicle		206±28 +++	208±27	240±34
	Duloxetine 0.1		187±24 +++	214±36	209±21
	Duloxetine 0.3		202±18 +++	241±28 *	248±27 *
	Duloxetine 1		197±22 +++	255±20 ***	270±16 ***
	Duloxetine 3		235±26 +++	323±32 ***	300±22 ***
	Duloxetine 5		222±15 +++	273±14 ***	288±21 ***
	Fluoxetine 10		206±26 +++	263±15***	298±21***
	Vehicle	Duloxetine 3	271±27 +++	381±36 ***	351±32 ***
	Methiothepin 1		259±12 +++	208±12	215±12
	Prazosin 1		260±17 +++	372±36 **	360±33 **

+++ $p<0.001$ *versus* saline/vehicle group, one way Anova followed by Newman-Keul's test (n=6-8 per group)

*$p<0.05$, **$p<0.01$, ***$p<0.001$ *versus* basal values, one way Anova with repeated measures followed by Newman-Keul's test (n=6-8 per group)

Interpretation of results

In anesthetized female guinea-pigs, intravesical infusion of AA elicited bladder overactivity characterized by a significant decrease in ICI. As previously described in cats (2), duloxetine, a 5-HT and NE re-uptake inhibitor, counteracted in a dose dependent manner the effects on ICI induced by AA bladder infusion. Fluoxetine, a selective 5-HT reuptake inhibitor reproduced the effects of

duloxetine on ICI in female guinea-pigs with AA bladder infusion suggesting an involvement of 5-HT receptors in the mechanism of action of duloxetine. This hypothesis was confirmed by the inhibition of the duloxetine effect by methiothepine, an antagonist of 5-HT_{1-2} receptors and by the lack of effect of prazosin, an α_1-adrenoreceptor antagonist.

Concluding message

In anesthetized female guinea-pigs, duloxetine, a NE and 5-HT re-uptake inhibitor reverses bladder overactivity, via 5-HT1 and/or 5-HT2 receptors, α_1-adrenoreceptors seems not to be involved.

Because of urinary tract innervation in guinea-pigs is closed to humans, these results suggest that guinea-pig is a suitable experimental model alternative to cat to investigate the effects of drugs on bladder overactivity.

Since it has been reported that duloxetine increases urethral sphincter activity in cats (2), further investigations evaluating the effects of duloxetine on external urethral sphincter activation in guinea-pigs are undergoing.

References

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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?	**Yes**
Name of ethics committee	**French Ministry for Agriculture and Fisheries**