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Comparative anticholinergic activities of 10 histamine H_1 receptor antagonists in two functional models

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Abstract

We determined the relative rank orders of anticholinergic potencies of 10 antihistamines in two functional bioassays: (1) an in vitro assay measuring inhibition of carbachol-induced contractions of isolated guinea pig *trachealis* muscle, (2) an in vivo bioassay comparing systemic hypotensive responses to bolus i.v. injections of acetylcholine before and after infusions of an antihistamine in anaesthetized rats. In vitro, the rank order of anticholinergic potencies of the antihistamines was cyproheptadine>promethazine>desloratadine>diphenhydramine>loratadine>chlorpheniramine>hydroxyzine>pyrilamine. The pA_2 values ranged from 8.2 ± 0.4 for cyproheptadine to 4.8 ± 0.4 for pyrilamine. Fexofenadine and cetirizine (up to 3×10^{-4} M) were inactive. In vivo, five antihistamines showed anticholinergic activity: cyproheptadine>promethazine>desloratadine>loratadine>diphenhydramine. The remaining antihistamines had no significant effect at i.v. infusion doses up to 50 ìmol/kg. Cetirizine and fexofenadine did not antagonize cholinergic responses in either model.

Keywords: Antihistamine; Anticholinergic; Carbachol; Histamine H_1 antagonist; Acetylcholine

1. Introduction

Since their discovery and early development in the 1940s, histamine H_1 receptor antagonists (antihistamines) have become one of the most widely used classes of medications for allergic disorders (Slater et al., 1999). Older "first-generation" antihistamines exhibit high binding affinity for H_1 receptors, but many of these drugs exhibit binding affinity for other classes of cellular receptors such as the muscarinic cholinergic subtypes (M_1–M_5) (Kubo et al., 1987). Anticholinergic properties of antihistamines have long been recognized (Sherrod et al., 1947; Reuse, 1948), and some clinical effects of antihistamines, such as dry mouth, constipation, urinary retention and tachycardia, are attributed to the antimuscarinic actions of these drugs (Simons, 1999). Another characteristic feature of the older antihistamines is that they gain access to the brain and bind to cellular receptors in the central nervous system (CNS), causing sedation and impaired psychomotor performance (Hindmarch and Shamsi, 1999; Shamsi and Hindmarch, 2000).

Newer "second-generation" antihistamines were developed as relatively more selective histamine H_1 receptor antagonists than the first-generation agents, with an aim of minimizing centrally mediated effects, such as sedation. However, it would appear that some of the newer antihistamines are capable of binding to muscarinic receptors, as well as to histamine H_1 receptors in the brain (Ter Laak et al., 1993).

Numerous functional models have been used to characterize the anticholinergic properties of antihistamines. For example, Niemegeers et al. (1982b) evaluated the central anticholinergic activity of several pharmacological classes of drugs by antagonism of physostigmine-induced lethality in rats. Among the antihistamines tested in this model, diphenhydramine and cyproheptadine were active. The central anticholinergic activity of diphenhydramine was also noted by its ability to suppress cholinomimetic-induced jaw

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tremors in a rat model (Carlson et al., 2000). Cardelús et al. (1999) measured ocular mydriatic responses in guinea pigs, and contractions of rabbit isolated iris muscle, reflecting anticholinergic activity; desloratadine was active and fexofenadine was inactive at the concentrations tested.

Thus, many studies have established that antihistamines can manifest anticholinergic effects to different degrees. A difficulty in gauging and comparing relative anticholinergic properties among antihistaminic drugs is that different model systems have been employed in comparative analyses. Also, in some reports, only a few antihistamines have been directly compared in a given test model.

Therefore, an aim of this study was to determine relative rank orders of anticholinergic potencies of a group of 10 antihistamines representing several different pharmacochemical classes, as quantified independently in two functional models of cholinergic antagonism: (1) an in vitro carbachol-induced muscle contraction model using guinea-pig tracheal segments; and (2) an in vivo model based on transient systemic arterial hypotensive responses to intravenous acetylcholine injections in urethane-anaesthetized rats. The 10 antihistamic drugs included six first-generation and four second-generation agents representing six pharmacochemical classes: substituted alkylamines, ethanolamines, ethylenediamines, phenothiazines, piperidines, and piperazines (Brown and Roberts, 2001).

2. Materials and methods

2.1. Animals

Male Hartley guinea pigs weighing 400–550 g were obtained from Hilltop Lab Animals (Scottsdale, PA, USA) and male Sprague–Dawley rats weighing 325–425 g were obtained from ACE Animals (Boyertown, PA, USA). Animals were acclimated and maintained in the vivarium at the University of the Sciences in Philadelphia (USP), Philadelphia, PA. All procedures and experimental protocols were reviewed and approved by the Institutional Animal Care and Use Committee at USP.

2.2. In vitro carbachol-induced tracheal contraction model

Guinea pigs were sacrificed by CO_2 narcosis and tracheae were excised and placed in modified Krebs–Henseleit buffer (37 °C and pH 7.4) containing 1 μM indomethacin. Tracheae were cleaned of fatty and connective tissue and cut into segments, two cartilage rings thick. Segments were bisected opposite the *trachealis* muscle and suspended in 10 ml tissue baths containing buffer aerated with a mixture of 95% O_2 and 5% CO_2. The integrity of the epithelium was verified by histological examination of several tracheal segments at the conclusion of an organ bath experiment.

Tissues were equilibrated for 60 min to a passive tension of 2 g, during which time the bath solution was refreshed every 15 min. *Trachealis* muscle contractions were measured via Grass FT03 force transducers on a Grass polygraph (Grass Instrument, Quincy, MA, USA). Tissues were primed and tested for viability with 10 μM carbachol. Following this challenge, tissues were washed with fresh buffer solution every 5 min for 20 min until tension returned to baseline. The tissues were then re-equilibrated at resting tension for 30 min, with fresh buffer added every 15 min.

A cumulative concentration–response curve was generated using carbachol 1×10^{-8} M through 3×10^{-5} M, in half log-unit increments. This served as the control response of the tissue in the absence of any antagonist compound. Tissues were re-equilibrated to resting tension following repeated washing. Tissues were then incubated with a single concentration of putative antagonist (or vehicle), following which, a second concentration–response curve to carbachol was generated in the presence of the antagonist (or vehicle). Each putative antagonist was tested, in different tissues, at several incremental bath concentrations ranging from low nanomolar up to a maximum of 0.3 mM, depending upon the occurrence of significant rightward shifts in tissue response curves to carbachol.

A group of 10 structurally diverse antihistamines was tested; six first-generation compounds (chlorpheniramine, cyproheptadine, diphenhydramine, hydroxyzine, promethazine and pyrilamine), and four second-generation compounds (cetirizine, desloratadine, fexofenadine and loratadine). Four reference antimuscarinic compounds were also tested: atropine, 4-diphenylacetoxy-*N*-methylpiperidine methiodide (4-DAMP), methoctramine and pirenzepine.

2.3. In vivo hypotensive response model

Rats were anaesthetized intraperitoneally with urethane (1.2 g/kg) and tracheotomized. The right jugular vein was isolated by manual dissection and cannulated. This cannula served as the drug delivery port for all subsequent i.v. injections.

For blood pressure measurements, the right carotid artery was exposed, separated from the vagus nerve and cannulated with polyethylene tubing connected to a Gould P23 pressure transducer (Gould, Oxnard, CA, USA). Systemic arterial blood pressure was recorded on a Grass polygraph. Following surgery, animals were allowed to stabilize for 10 min. Animals with erratic blood pressures or with a mean arterial pressure below 75 mm/Hg were not included in the analysis. For all experiments, the surgical preparation was identical and the i.v. drug injection volume was fixed at 0.1 ml.

The selection of appropriate i.v. doses of each of the 10 antihistamines was based on a series of preliminary experiments to establish the largest dose of each drug that could be infused without evoking more than a 25 mm Hg change in mean arterial blood pressure. This dose was

Table 1
Doses of histamine H_1 receptor antagonists used to evaluate anticholinergic activity

Antagonist	Dose (μmol/kg)[a]		
	(Low)	(Med)	(High)
Atropine sulfate[b]	0.2	1.0	2.0
Cetirizine hydrochloride[c]	2.0	20.0	50.0
(±) Chlorpheniramine maleate[b]	2.0	20.0	50.0
Cyproheptadine hydrochloride[b]	2.0	8.0	20.0
Desloratadine[c]	6.0	20.0	30.0
Diphenhydramine hydrochloride[b]	2.0	20.0	50.0
Fexofenadine hydrochloride[c]	2.0	20.0	50.0
Hydroxyzine dihydrochloride[b]	2.0	20.0	50.0
Loratadine[c]	2.0	20.0	50.0
Promethazine hydrochloride[b]	2.0	20.0	35.0
Pyrilamine maleate[b]	2.0	20.0	50.0

[a] Doses are calculated from the free base form.
[b] Sigma, St. Louis, MO, USA.
[c] Donated by Aventis Pharma, Bridgewater, NJ, USA.

arbitrarily designated as the 'high' dose. Two lower doses were also tested to enable construction of a three-point dose–response curve. Table 1 lists the three incremental doses of each compound studied. The experimental protocol was as follows: randomized i.v. bolus injections of acetylcholine (0.1, 1.0, and 10.0 μg/kg) were given in duplicate to obtain a series of graded, baseline hypotensive responses. The 'low' dose of antagonist was then infused over 5 min. Ten minutes later, the series of acetylcholine injections was repeated. The acetylcholine injections were repeated twice again, following 'medium' and 'high' infused doses of the antagonist. The vehicle for cyproheptadine, desloratadine, fexofenadine and loratadine was 95% ethanol; all other drugs were dissolved in 0.9% saline. Neither vehicle modified hypotensive responses to i.v. acetylcholine.

2.4. Statistical analyses

2.4.1. In vitro carbachol-induced tracheal contraction model

Data were analysed using the method of Arunlakshana and Schild (1959). Dose ratios were calculated for each concentration of antagonist according to the formula:

$$\text{Dose Ratio} = EC'_{50} / EC_{50}$$

In the above equation, EC'_{50} is the concentration of carbachol causing half the maximal response in the presence of antagonist, and EC_{50} is the concentration causing half the maximal response in the absence of antagonist. The concentration of the antagonist was plotted versus log (dose ratio−1). Linear regression was used to generate a best-fit line for antagonist data, where the pA_2 value of the antagonist is the x-intercept of the regression line. To test whether the Schild slope differed from unity, the regression line was compared with a hypothetical regression line with a slope of unity using an analysis of covariance. Slopes were considered to differ significantly from unity if $P<0.05$.

2.4.2. In vivo hypotension response model

Blood pressure responses after antihistamine infusions were compared with baseline responses by a repeated-measures analysis of variance, with Dunnett's post-hoc test. Responses were considered to be significantly different when $P<0.05$. For atropine and the antihistamines exhibiting significant anticholinergic activity, ratios of equally hypotensive doses of acetylcholine were calculated according to the formula:

$$\text{Ratio of Equi} - \text{Hypotensive Doses of Acetylcholine} = ED'_{40} / ED_{40}$$

In the above equation, ED'_{40} is the bolus i.v. dose of acetylcholine that evoked a 40% reduction in blood pressure (from the experimental maximum) in the presence of the putative antagonist, and ED_{40} is the bolus i.v. dose of acetylcholine that produced the same response in the absence of the putative antagonist. The experimental maximum was defined as the mean maximal decrease in blood pressure elicited by 10 μg/kg i.v. of acetylcholine for all rats (n=57), and was equal to 62±10.9 (S.D.) mm Hg.

3. Results

3.1. In vitro carbachol-induced tracheal contraction model

Eight of the antihistamines expressed anticholinergic activity of varying potencies, as evidenced by parallel rightward shifts of the concentration–response curves for carbachol. The rank order of potencies was: cyproheptadine> promethazine>desloratadine>diphenhydramine> loratadine>chlorpheniramine>hydroxyzine>pyrilamine (Table 2). The pA_2 values ranged from 8.2±0.4 for

Table 2
Antagonist potencies

Antagonist	pA_2 Value	Schild slope
Atropine	9.5±0.3	0.92±0.08
Cyproheptadine	8.2±0.4	0.95±0.15
Promethazine	7.8±0.3	0.98±0.10
Desloratadine	6.9±0.4	0.96±0.08
Diphenhydramine	6.3±0.3	1.19±0.03
Loratadine	5.6±0.3	0.63±0.04[a]
(±) Chlorpheniramine	5.4±0.4	1.31±0.10
Hydroxyzine	5.2±0.2	0.88±0.16
Pyrilamine	4.8±0.4	0.86±0.23
Cetirizine	Indeterminable	Indeterminable
Fexofenadine	Indeterminable	Indeterminable
Pirenzepine (M_1)	7.4±0.4	0.70±0.12[a]
Methoctramine (M_2)	5.4±0.2	1.2±0.30
4-DAMP (M_3)	9.2±0.3	0.81±0.11

Results are mean±S.D. (n=4–10).
[a] Significant slope difference from unity ($P<0.01$).

cyproheptadine to 4.8±0.4 for pyrilamine. The relative anticholinergic potencies of the four reference antimuscarinic compounds were: atropine>4-DAMP>pirenzepine> methoctramine (Table 2).

Representative concentration–response curves for six of the antihistaminic drugs are illustrated in Fig. 1. Diphenhydramine and chlorpheniramine (Fig. 1A and B), first-generation drugs, displayed parallel rightward shifts of the concentration–response curves to carbachol, as did desloratadine and loratadine (Fig. 1C and D), second-generation drugs. Fexofenadine and cetirizine, tested at tissue bath concentrations up to 3×10^{-4} M, did not produce observable cholinergic antagonism when compared to vehicle (Fig. 1E and F).

Calculated pA_2 values against carbachol, derived in this study of histamine H_1 antagonists, were compared with published pA_2 values of these compounds against histamine-induced smooth muscle contraction. Antihistaminic pA_2 values typically have been obtained using isolated guinea pig ileum. Such a comparison provided a means to estimate the relative antihistaminic/anticholinergic selectivity of the compounds in similar functional bioassay systems. Table 3 lists pA_2 values of histamine H_1 antagonists against carbachol from the present study and corresponding pA_2 values against histamine from several published studies. Selectivity ratios were calculated as the antilog of the difference between the pA_2 value against carbachol and the pA_2 value against histamine. Selectivity ratios varied greatly among the six histamine H_1 antagonists for which published pA_2 data were found: promethazine, desloratadine, diphenhydramine, and loratadine yielded selectivity ratios of 13 to 50, whereas chlorpheniramine and pyrilamine had much greater apparent selectivity, with ratios of 10,000 and 25,119, respectively.

3.2. In vivo hypotension response model

Only 5 of the 10 antihistamines that were tested significantly inhibited acetylcholine hypotensive responses to i.v. injections of acetylcholine compared with pre-

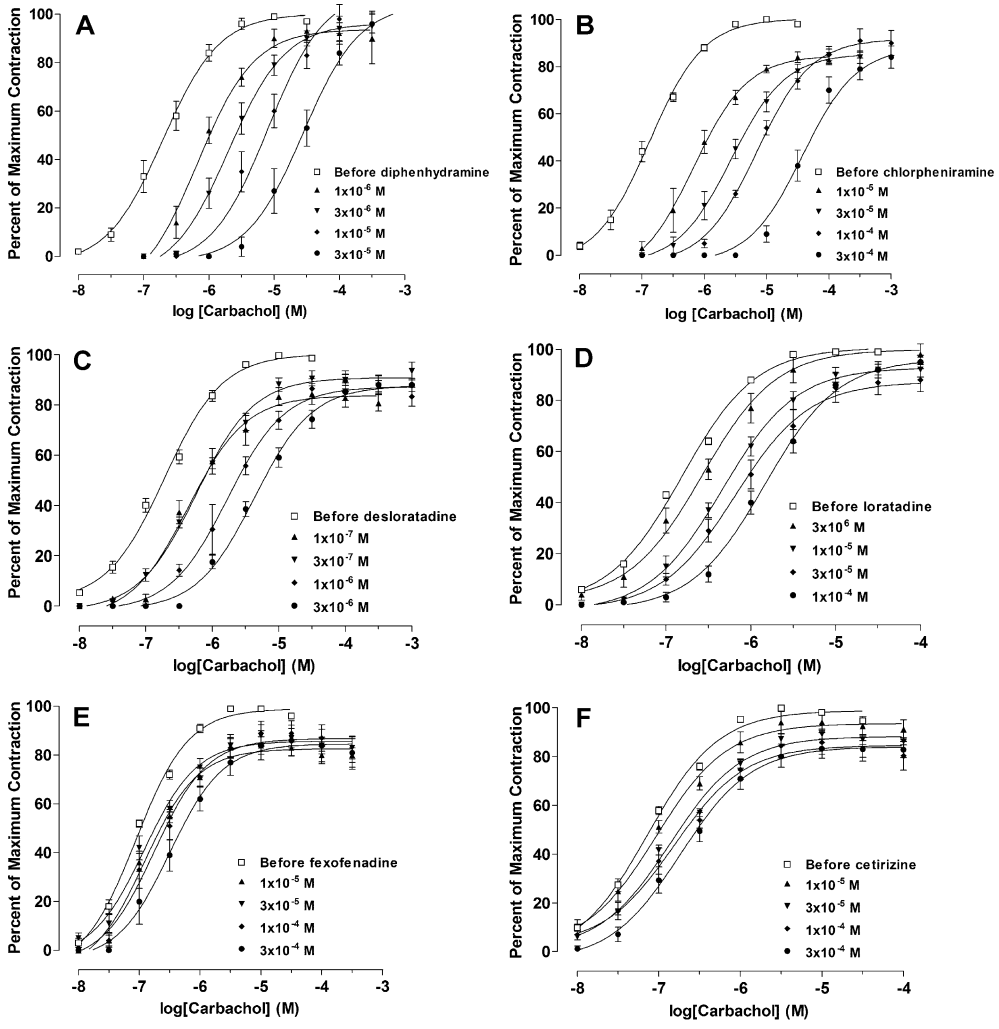

Fig. 1. Concentration–response curves for carbachol-induced contractions of guinea-pig tracheal smooth muscle in the absence and presence of: (A) diphenhydramine (n=5), (B) chlorpheniramine (n=5), (C) desloratadine (n=4), (D) loratadine (n=7), (E) fexofenadine (n=5), and (F) cetirizine (n=5). Results are mean±S.E.M.

Table 3
Anticholinergic and published antihistaminic pA_2 values for histamine H_1 antagonists

Antagonist	Anticholinergic pA_2 (g pig trachea)	Antihistaminic pA_2 (g pig ileum published data)*	Selectivity Ratio[a]	*Reference
Cyproheptadine	8.2±0.4	Not found		
Promethazine	7.8±0.3	8.9	13	Hill et al., 1981
Desloratadine	6.9±0.4	8.2	20	Kreutner et al., 2000
Diphenhydramine	6.3±0.3	7.80	32	Labrid et al., 1977
Loratadine	5.6±0.3	7.3	50	Kreutner et al., 2000
Chlorpheniramine	5.4±0.4	9.1, 9.6	10,000[b]	Hill et al., 1981; Kreutner et al., 2000
Hydroxyzine	5.2±0.2	Not found		
Pyrilamine	4.8±0.4	9.2	25,119	Hill et al., 1981; Labrid et al., 1977
Cetirizine	Indeterminable	6.3 (g pig trachea)		Abe et al., 1994
Fexofenadine	Indeterminable	Not found		

[a] Antilog[pA_2(histamine)−pA_2(carbachol)].
[b] Denotes ratio derived using the average of two pA_2 values.

antagonist responses: cyproheptadine, desloratadine, diphenhydramine, loratadine, and promethazine (Table 4). Atropine, the reference antimuscarinic compound, was by far the most potent and effective anticholinergic agent in this functional test model. Representative dose–response curves obtained with four of the second-generation antihistamines are shown in Fig. 2. Desloratadine and loratadine (panels A and B) significantly inhibited acetylcholine-induced hypotension, whereas fexofenadine and cetirizine (panels C and D) did not. The rank order of potencies of the five active

Table 4
Hypotensive responses to acetylcholine before and after i.v. infusion of various antihistamines and atropine at low, medium and high doses

Antagonist[a]	Acetylcholine dose (μg/kg)	Maximum % decrease in mean arterial blood pressure[b]			
		Pre-antagonist	Low dose	Medium dose	High dose
Atropine	0.1	29±3.0	0.8±0.8[a]	0.8±0.8[a]	0.8±0.8[a]
	1.0	45±3.4	26±5.9[a]	9±3.4[a]	2±1.2[a]
	10.0	61±3.7	46±5.1	33±3.0[a]	18±4.2[a]
Cyproheptadine	0.1	23±2.0	17±3.8	7±1.8[a]	2±0.8[a]
	1.0	40±2.7	37±2.3	28±2.5[a]	16±4.5[a]
	10.0	50±2.2	47±1.7	39±3.2[a]	24±3.8[a]
Promethazine	0.1	27±1.1	21±1.8[a]	8±1.1[a]	4±0.9[a]
	1.0	42±0.9	39±1.4	28±4.2[a]	19±1.4[a]
	10.0	57±0.9	51±3.4	41±1.0[a]	37±2.8[a]
Desloratadine	0.1	27±0.9	22±3.0	16±2.6[a]	11±2.5[a]
	1.0	40±1.4	37±1.3	33±1.3[a]	28±1.7[a]
	10.0	47±0.8	46±0.3	41±1.7[a]	40±2.8[a]
Lotatadine	0.1	26±2.2	23±2.1	23±3.1	12±1.7[a]
	1.0	43±1.7	41±2.5	35±1.7[a]	31±1.6[a]
	10.0	52±1.8	51±2.2	44±2.5[a]	41±1.9[a]
Diphenhydramine	0.1	25±1.7	23±2.0	14±3.9[a]	10±2.4[a]
	1.0	48±2.8	45±1.5	39±3.9	37±2.0
	10.0	57±2.5	53±2.2	53±2.1	52±1.1
Cetirizine	0.1	26±2.2	26±1.8	26±1.1	23±1.3
	1.0	40±2.6	37±1.2	38±2.7	37±1.5
	10.0	61±1.3	52±2.8	49±3.8	53±3.8
Hydroxyzine	0.1	25±3.1	23±2.2	20±4.1	27±2
	1.0	42±2.5	40±1.3	40±0.7	40±1.1
	10.0	52±1.8	49±2.5	51±1.1	49±1.3
Fexofenadine	0.1	27±1.7	26±1.8	24±1.2	26±3.1
	1.0	41±1.8	41±1.7	38±3.0	38±1.2
	10.0	49±1.5	49±2.1	46±2.4	45±2.8
Pyrilamine	0.1	29±1.7	26±2.0	27±4.0	31±4.4
	1.0	47±1.0	45±1.7	48±2.3	50±2.1
	10.0	57±2.2	51±2.9	57±2.5	57±3.1
Chlorpheniramine	0.1	33±1.2	31±2.1	29±2.9	25±1.6
	1.0	48±1.4	47±1.1	46±1.6	44±2.0
	10.0	61±3.2	57±2.3	59±0.9	55±1.7

[a] $P<0.05$ compared with pre-antagonist response to acetylcholine.
[b] Each antagonist was tested in four to six rats. Data are group means±S.E.M.

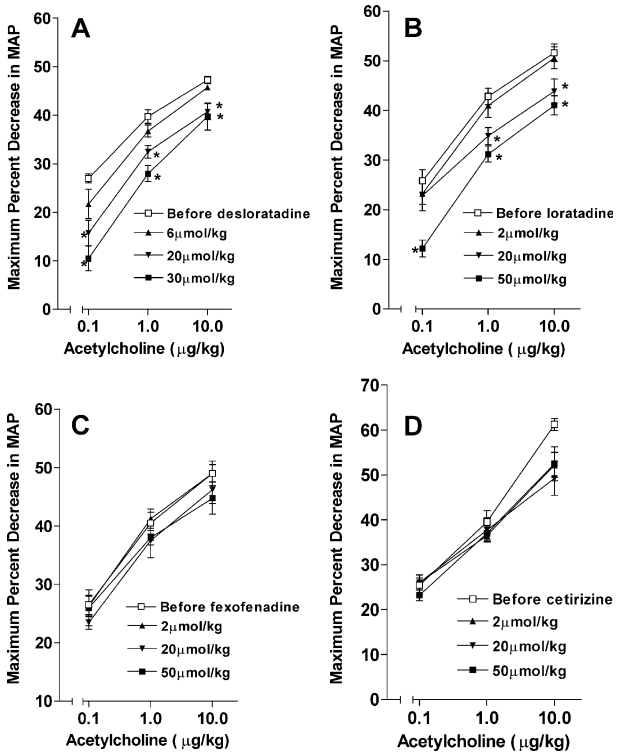

Fig. 2. Effects of (A) desloratadine (n=4), (B) loratadine (n=5), (C) fexofenadine (n=4), and (D) cetirizine (n=4), on acetylcholine-induced hypotensive responses. Results are expressed as mean±S.E.M.

compounds was: cyproheptadine>promethazine>desloratadine>loratadine>diphenhydramine, as reflected by the slopes and positions of the regression lines shown in Fig. 3. The remaining five antihistamines (cetirizine, chlorpheniramine, fexofenadine, hydroxyzine and pyrilamine), at intravenous infusion doses up to 50 μmol/kg, did not significantly alter hypotensive responses to acetylcholine.

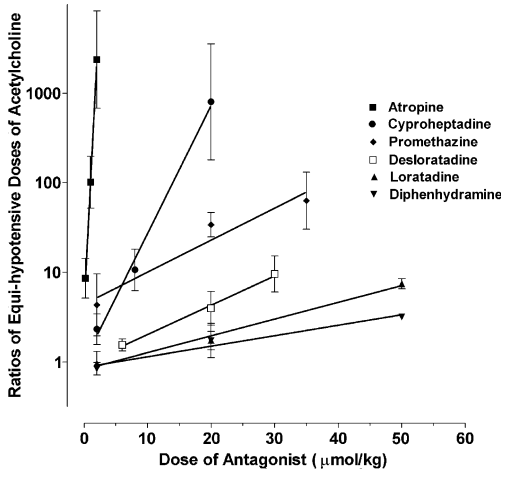

Fig. 3. Ratios of equi-hypotensive doses (ED_{40}) of acetylcholine in the presence of three increasing doses of atropine or each of five antihistamines. Results are expressed as mean±S.E.M. (n=4–6).

4. Discussion

As anticipated, the 10 structurally diverse antihistamines tested in this study showed a broad range of anticholinergic potency and effectiveness, in both the in vitro guinea-pig *trachealis* and the in vivo rat blood pressure models of cholinergic functional antagonism. Fig. 4 illustrates the significant correlation between anticholinergic pA_2 values in the in vitro *trachealis* model and regression slope values of the ratios of equi-hypotensive doses of acetylcholine in vivo for the five antihistamines which displayed anticholinergic activity in both model systems (Spearman correlation, r=0.94, P<0.05).

First-generation antihistamines, such as cyproheptadine, diphenhydramine and promethazine, are recognized as having clinically significant anticholinergic properties (Brown and Roberts, 2001). Two of the second-generation compounds tested also had significant anticholinergic properties in both of our functional test models, and were interspersed with first-generation compounds within the rank order of anticholinergic potencies. Desloratadine (pA_2=6.9±0.4) showed more than 10-fold higher potency than its parent compound loratadine (pA_2=5.6±0.3) in the *trachealis* test model. Similarly, Cardelús et al. (1999) reported a pA_2 value of 6.67±0.09 for desloratadine against carbachol-induced contractions of isolated iris smooth muscle from rabbits, a model of muscarinic M_3 receptor-mediated function. These two second-generation drugs also appear to have antihistaminic/anticholinergic selectivity in vitro comparable to diphenhydramine and promethazine (Table 3).

Under our in vivo assay conditions, three first-generation antihistamines did not have a significant inhibitory effect on acetylcholine-induced hypotensive responses: pyrilamine, chlorpheniramine, and hydroxyzine. Of the second-generation compounds, cetirizine and fexofenadine had no significant inhibitory effect, whereas loratadine and desloratadine significantly suppressed acetylcholine-induced hypotension, with desloratadine showing somewhat greater activity than its parent compound.

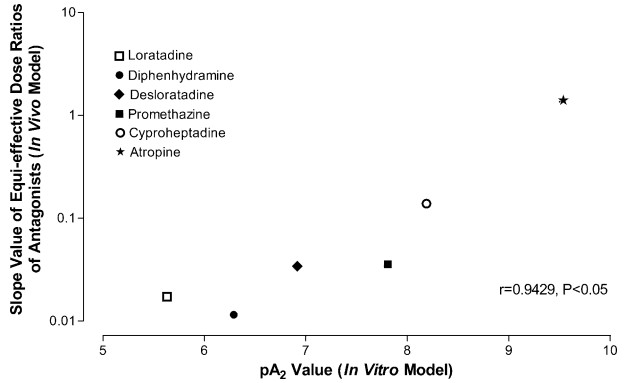

Fig. 4. Correlation between in vivo and in vitro anticholinergic activities of five antihistamines showing activity in both model systems, and atropine.

The observed rank order of potency of muscarinic receptor subtype-selective antagonist compounds in the guinea pig *trachealis* model, 4-DAMP>pirenzepine>methoctramine, is consistent with results of other investigators showing that the cholinergic contractile response of tracheal smooth muscle of guinea pigs is mediated mainly via muscarinic M_3-receptors (Haddad et al., 1991; Eglen et al., 1996). There is some evidence that antihistaminic compounds with measurable anticholinergic activity may have affinity for muscarinic subtypes other than M_3 receptors. Ellis and Seidenberg (2001) have reported high in vitro binding affinities of loratadine and desloratadine for other muscarinic subtypes, particularly M_1 and M_2.

Muscarinic receptor subtypes involved in mediating vasodilation are less fully characterized than those involved in the contractile response of *trachealis* muscle of guinea pigs. However, evidence suggests that for rat mesenteric (Hendriks et al., 1992) and renal (Eltze et al., 1993) resistance blood vessels, as well as the pulmonary artery (McCormack et al., 1988; Walch et al., 1999), the receptors are primarily of the muscarinic M_3 subtype. Interestingly, different subtypes appear to be involved in different species; in humans the muscarinic M_3 subtype is also implicated, as well as M_1, in the control of vascular tone in isolated pulmonary arteries (Walch et al., 1999; Norel et al., 1996).

Since both bioassay systems would appear to constitute functional examples of mainly muscarinic M_3 receptor-mediated responses, consistency in relative rank orders of activity of the antihistamines was not surprising. However, the in vitro *trachealis* contraction model was more 'sensitive' in distinguishing among the individual antihistamines than the in vivo arterial blood pressure model, possibly due to greater physiological complexity underlying regulation of systemic arterial blood pressure in the intact organism versus contraction of the isolated tracheal muscle preparation.

Chlorpheniramine, cetirizine, fexofenadine, and hydroxyzine are chiral compounds. Racemic mixtures were tested in the present study because they are present in commercial drug product formulations. Investigation of the anticholinergic activity of the enantiomers of each of these compounds is warranted, but was not addressed in this study.

In summary, the rank orders of anticholinergic activity of 10 structurally diverse antihistamines were quantified in two bioassay models of muscarinic M_3 receptor functional antagonism. The findings generally confirm and expand upon previously published reports on the anticholinergic properties of first-generation antihistaminic drugs studied in several different functional bioassay systems (Niemegeers et al., 1982a,b; Carlson et al., 2000; Remy et al., 1977). Cyproheptadine, promethazine and diphenhydramine showed measurable activity in both of the test models. Among the newer-generation antihistamines, loratadine and its active metabolite desloratadine were observed to have anticholinergic activity in both bioassay models, whereas cetirizine and fexofenadine did not.

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