J. Pharm. Pharmacol. 1990, 42: 102–107
Received April 14, 1989

Effects of Cularine and Other Isoquinoline Alkaloids on Guinea-pig Trachea and Human Bronchus

M. L. CANDENAS, E. NALINE, M. P. D'OCÓN*, D. CORTES* AND C. ADVENIER

*Laboratoire de Pharmacologie, Faculté de Médecine Paris-Ouest et Institut Biomédical des Cordeliers, 15 rue de l'Ecole de Médecine, F-75270 Paris Cédex 06, France, *Laboratorio de Farmacognosia y Farmacodinamia, Departamento de Farmacologia y Farmacotécnia, Facultad de Farmacia, Avda. Blasco Ibañez 13, 46010-Valencia, España*

Abstract—The relaxant effects of the isoquinoline alkaloids, cularine, antioquine, obaberine and 6, 7-dimethoxy-1, 2, 3, 4-tetrahydroisoquinoline, with structures related to that of papaverine, have been studied on the guinea-pig isolated trachea and human bronchus against contractions induced by acetylcholine (ACh), histamine, neurokinin A (NKA) and KCl. These effects were compared with those of papaverine and theophylline. Among the alkaloids tested, the most potent was cularine, the relaxant activity of which, in terms of pD_2, was between those of papaverine and theophylline. The results showed that for the guinea-pig isolated trachea and the human bronchus, the cularine concentrations required to inhibit K^+-induced contractions were close to those necessary to counteract ACh-, histamine- and NKA-induced contractions. The relaxant activity of cularine was not modified when it was tested on tracheal preparations under resting tone or after epithelium removal. In addition, cularine was able to inhibit Ca^{2+}-induced contractions in a Ca^{2+}-free, K^+-enriched solution at the same concentrations as those which inhibited the action of the different contractile agents in normal Krebs solution. All these data show that cularine displays non-specific antispasmogenic activity on guinea-pig and human airways. The lower relaxant activity (pD_2) of cularine compared with papaverine suggests the importance of free rotation of the isoquinoline-benzyl ring linkage as well as of the degree of hydrogenation of the heterocyclic ring in the spasmolytic effects of isoquinoline compounds.

Isoquinoline alkaloids are often found in nature, associated mainly with the Magnoliales and Ranunculales orders. Pharmacologically, one of the most representative compounds of this group is the benzylisoquinoline, papaverine, which has non-specific relaxant activity, although its mechanism of action remains unclear (Fujioka 1984; Calixto & Loch 1985; Zhang & Zhou 1988). However, little is known about the potentially therapeutic value of other isoquinoline alkaloids as smooth muscle relaxants.

In the present study, we have examined the smooth muscle relaxant activity of several isoquinoline structures on the guinea-pig isolated trachea and the human bronchus, comparing them with those of papaverine and theophylline. Two of the alkaloids, antioquine and obaberine, are bisbenzylisoquinolines and have been isolated from the stem bark of *Pseudoxandra sclerocarpa*, antioquine being the major alkaloid obtained from this Columbian plant (Cortes et al 1985). Cularine is an isoquinoline alkaloid isolated from plants belonging mainly to the Fumariaceae family (Castedo 1985). We have also tested 6, 7-dimethoxy-1, 2, 3, 4-tetrahydroisoquinoline, in an attempt to determine the influence of loss of the benzyl ring on the relaxant activity of these compounds. Their structure is shown in Fig. 1.

Materials and Methods

Guinea-pig isolated trachea

Male guinea-pigs (250-350 g) were killed by a blow on the head and exsanguinated. The trachea was removed and placed in Krebs solution (composition mM: NaCl 118, KCl 5·4, $CaCl_2$ 2·5, KH_2PO_4 1·2, $MgSO_4$ 0·6, $NaHCO_3$ 25 and glucose 11·7). Following removal of adhering fat and connective tissue, the trachea was slit along its longitudinal axis, directly opposite the smooth muscle. Strips consisting of three adjacent cartilage rings were prepared according to Emmerson & Mackay (1979).

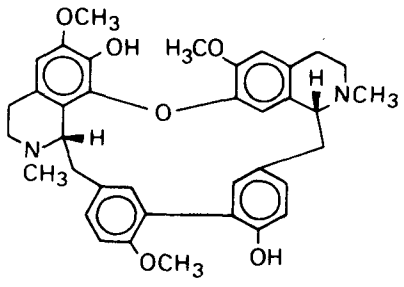
In some experiments, the epithelium of one of these strips was removed by gently rubbing the luminal surface (over both the smooth muscle and cartilage areas) with a cotton-tipped applicator (Hay et al 1986a, b; Raeburn et al 1986; Tschirhart & Landry 1986; Devillier et al 1988); the other strip served as a paired control.

The strips were then suspended in 10 mL organ baths containing Krebs solution at 37°C, gassed with 95% O_2 and 5% CO_2 and equilibrated under an initial tension of 1·80 g. After equilibration for 1·25 h, the resting tension was between 0·6 and 1·4 g. Under these conditions, responses to agonists were reproducible. Tension was measured isometrically with Celaster strain gauges and Celaster amplifiers and displayed on Linsee recorders.

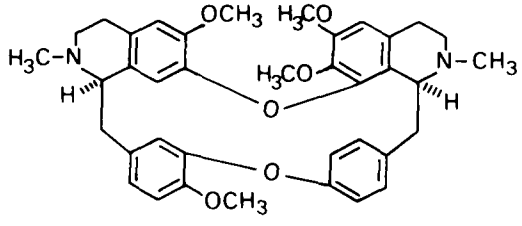
Human bronchus

Human bronchial tissue (usually with an inner diameter of 4–6 mm) from patients undergoing surgery for lung cancer, but taken at a distance from the malignancy, was dissected free of parenchyma and transported to the laboratory in ice-cold Krebs solution previously aerated with a mixture of 95% O_2 and 5% CO_2. The tissue was stored overnight at 4°C, and the experiment was carried out the following day. Published data have shown that overnight storage of tissue does not alter its reactivity (Brink et al 1980; Ghelani et al 1980; Guillot et al

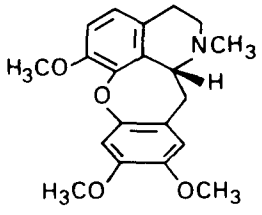
Correspondence to: M. L. Candenas, Laboratoire de Pharmacologie, Faculté de Médecine Paris-Ouest et Institut Biomédical des Cordeliers, 15 rue de l'Ecole de Médecine, F-75270 Paris Cédex 06, France.



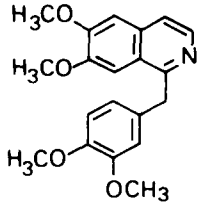
ANTIOQUINE



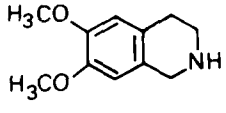
OBABERINE



CULARINE



PAPAVERINE



6,7-DIMETHOXY-1,2,3,4-TETRAHYDROISOQUINOLINE

FIG. 1. Structures of isoquinoline compounds tested.

1984; Vincenc et al 1984). Transversally cut rings from a segmental bronchus were suspended in Krebs solution under an initial tension of 2·0 g and treated in every respect as the guinea-pig isolated tracheal strips.

Guinea-pig trachea

In all experiments, preparations were first tested for maximal tension with acetylcholine (ACh, 10^{-3} M), then allowed to rest for at least 1 h during which washing was performed every 15 min.

In a first series of experiments, the tracheal strips were contracted to 70–90% of maximal contraction with ACh (10^{-4} M), histamine (10^{-5} M), neurokinin A (NKA, 10^{-8} M) and KCl (10^{-2} M). When a stable contraction was obtained, cumulative concentration-response curves to papaverine (10^{-7} to 10^{-4} M), cularine (10^{-6} to 3×10^{-4} M), antioquine (10^{-6} to 10^{-4} M), obaberine (10^{-6} to 3×10^{-4} M), 6, 7-dimethoxy-1, 2, 3, 4-tetrahydroisoquinoline (10^{-6} to 10^{-3} M) or theophylline (10^{-5} to 3×10^{-3} M) were obtained by adding increasing concentrations of the drugs at 10–15 min intervals. Once the concentration-response curve was completed, theophylline (3×10^{-3} M) was added to the bath to determine maximal relaxation. The results were expressed as percentage of the effect induced by theophylline.

In a second series of experiments performed on parallel segments of trachea, the concentration-response curves to cularine, papaverine and theophylline were established on the preparations under resting tone or precontracted with histamine (10^{-5} M) and in the presence or in the absence of epithelium. The results were expressed as a percentage of maximal relaxation induced by theophylline (3×10^{-3} M).

In a third series of experiments Ca^{2+} dose-response curves were established according to Godfraind et al (1968). Tracheal strips were incubated for 1 h in a solution similar to the one just described but without $CaCl_2$, then for 15 min in $CaCl_2$-free solution in the presence of ethylenediaminetetraacetic acid (EDTA, 10^{-3} M). The preparations were washed at intervals of 15 min. In a second stage, the strips were incubated in a calcium-free solution with additional K^+. The composition of the potassium-enriched solution was (mM): NaCl 109, KCl 30, $MgCl_2$ 0·49, $NaHCO_3$ 11·9, Na_2HPO_4 0·4 and glucose 5·5 (pH 7·46). After incubation, the dose-response curves of Ca^{2+} (10^{-5} to 3×10^{-3} M) were determined on parallel segments of trachea by cumulative addition. The drugs tested were added to the bath 15 min before addition of Ca^{2+}, except in one of the tracheal strips, which served as a paired control. Responses were expressed as a percentage of maximal contraction induced with calcium in the control concentration-response curve.

In a fourth series of experiments, dose-response curves to adenosine were obtained by increasing the concentration of adenosine at 5–10 min intervals, the tracheal strip being precontracted with histamine (10^{-5} M). After the concentration-response curve to adenosine was completed, theophylline (3×10^{-3} M) was added to the bath to determine maximal relaxation. Other tracheal strips were pretreated with dipyridamole (10^{-5} M) or with dipyridamole (10^{-5} M) and cularine (10^{-5} or 3×10^{-5} M) or papaverine (3×10^{-7}, 10^{-6}, or 3×10^{-6} M) 10 min before addition of histamine. The adenosine-induced relaxation was expressed as a percentage of the maximal effect of theophylline.

Owing to the development of tachyphylaxis, only one series of concentration-response curves was obtained from each tracheal preparation in each series of experiments.

Human bronchus

Isolated human bronchi were first contracted to maximal tension with ACh (10^{-3} M). Afterwards, the effects of cularine, papaverine and theophylline were tested against contractions induced by ACh, histamine and KCl at the concentrations stated above and NKA (10^{-7} M). After the concentration-response curves to the different drugs were completed, theophylline (3×10^{-3} M) was added to the bath. Relaxations were expressed as a percentage of the effect induced by theophylline, and $-\log$ EC50 values were calculated from the log concentration-effect curves.

Statistical analysis

Statistical analysis of the results obtained was performed using Student's *t*-test. All values in the test and tables are expressed as mean ± s.e. mean. *P* values below 0·05 were considered to be significant.

Drugs

The drugs used were: antioquine and obaberine (Laboratoire de Matière Medicale, Faculté de Pharmacie, Chatenay-Malabry, France), cularine (Departamento de Quimica Orgánica, Facultad de Ciencias Quimicas, Santiago de Compostela, España), 6,7-dimethoxy-1, 2, 3, 4-tetrahydroisoquinoline, adenosine (Aldrich Chemie, FRG-Steinheim), papaverine (Laboratoires Salvoxyl Wander, Paris), acetylcholine dihydrochloride (Lematte et Boinot, Paris), dipyridamole (Boehringer-Ingelheim, F-Reims), histamine hydrochloride (Sigma, St. Louis, USA), neurokinin A (School of Medicine, Sherbrooke, Canada) and theophylline (Lab. Bruneau et Cie., Paris). All other reagents were of analytical grade.

All drugs were dissolved in distilled water except for dipyridamole, which was dissolved in ethanol. All dilutions were made with Krebs solution.

Results

Relaxant effects of isoquinoline alkaloids on guinea-pig trachea and human bronchus

Fig. 2 shows the concentration-response curves obtained with antioquine, obaberine, 6, 7-dimethoxy-1, 2, 3, 4-tetrahydroisoquinoline, cularine, papaverine and theophylline on the plateaus of contraction caused by the various contracting agents used in the guinea-pig isolated trachea and human bronchus preparations. Maximal relaxations obtained with antioquine (10^{-4} M) and obaberine (10^{-4} M) against histamine in guinea-pig trachea, expressed as a percentage of maximal response to theophylline (3×10^{-3} M), were 32·5 ± 3·9% and 36·6 ± 0·7%, respectively, and partial effects were also observed with the other contracting agents. Maximal relaxations induced by 6, 7-dimethoxy-1, 2, 3, 4-tetrahydroisoquinoline (10^{-3} M) against histamine-induced contractions were 72·4 ± 11·7%, but relaxations of 52·8 ± 10·6%, 39·0 ± 0·5% and 9·2 ± 4·7% were obtained when ACh, NKA and KCl were used as contractile agents.

Table 1. −log EC50 values of cularine, papaverine and theophylline on contraction induced by different agents in guinea-pig trachea and human bronchus.

Concn (M)	n	Cularine −log EC50	n	Papaverine −log EC50	n	Theophylline −log EC50
Isolated guinea-pig trachea						
Hist (10^{-5})	6	4·57 ± 0·12	6	6·00 ± 0·03	5	4·09 ± 0·02
ACh (10^{-4})	6	4·16 ± 0·06	5	5·56 ± 0·06	6	3·47 ± 0·06
NKA (10^{-8})	5	4·39 ± 0·09	4	6·09 ± 0·02	5	4·22 ± 0·02
KCl (10^{-2})	6	4·33 ± 0·03	6	6·00 ± 0·03	5	4·20 ± 0·02
Ca^{2+} (3×10^{-4})	5	4·31 ± 0·08	4	5·90 ± 0·10	—	—
Human bronchus						
Hist (10^{-5})	5	4·22 ± 0·03	5	5·52 ± 0·11	4	3·95 ± 0·12
ACh (10^{-4})	5	3·86 ± 0·10	5	5·22 ± 0·05	4	3·56 ± 0·13
NKA (10^{-7})	5	4·07 ± 0·10	5	5·54 ± 0·09	4	3·79 ± 0·15
KCl (10^{-2})	5	4·14 ± 0·07	4	5·41 ± 0·09	4	3·90 ± 0·14

For this reason, values of −log EC50 were not calculated for these three products.

On the other hand, maximal relaxations induced by cularine were not significantly different from those produced by papaverine (10^{-4} M) and theophylline (3×10^{-3} M), whatever the agonist used (Fig. 2). Table 1 shows that the −log EC50 values obtained for each of these three drugs, cularine, papaverine and theophylline, were similar with both human bronchi and guinea-pig trachea, when histamine, NKA or KCl were used to contract the preparation. Table 1 also shows that the −log EC50 values obtained for each relaxant drug against ACh-induced contractions were lower than those obtained for these three drugs when contractions were induced by any of the other agonists, i.e. significantly more of each non-specific drug is required to functionally antagonize ACh-induced contractions in both preparations, compared with the other agonists (see also minimum effective, agonist doses in Fig. 2).

Influence of epithelium and precontraction of preparation on cularine concentration response curves in the guinea-pig isolated trachea

Table 2 shows that removal of the epithelium did not result in any significant changes of E_{max} or −log EC50 values, i.e. there were no significant shifts of the concentration-response curves to cularine, papaverine and theophylline, irrespective

Table 2. Influence of epithelium removal on −log EC50 and E_{max} values of cularine, papaverine and theophylline in the isolated guinea-pig trachea. Values are mean ± s.e. mean. The number of experiments is given in parentheses

	Cularine −log EC50		Papaverine −log EC50		Theophylline −log EC50	
Pretreatment	With epithelium	Without epithelium	With epithelium	Without epithelium	With epithelium	Without epithelium
Spontaneous tone	4·27 ± 0·07 (5)	4·48 ± 0·12 (5)	6·12 ± 0·20 (4)	5·87 ± 0·12 (4)	4·01 ± 0·19 (4)	3·69 ± 0·18 (4)
Histamine (10^{-5} M)	4·38 ± 0·05 (5)	4·60 ± 0·10 (5)	6·08 ± 0·17 (5)	6·16 ± 0·09 (5)	3·93 ± 0·15 (5)	3·82 ± 0·08 (5)
	E_{max} (g)		E_{max} (g)		E_{max} (g)	
Spontaneous tone	1·31 ± 0·22	1·32 ± 0·31	1·31 ± 0·41	1·51 ± 0·41	1·37 ± 0·61	1·17 ± 0·48
Histamine (10^{-5} M)	2·15 ± 0·46	1·57 ± 0·33	1·71 ± 0·32	2·11 ± 0·20	1·85 ± 0·35	1·10 ± 0·33

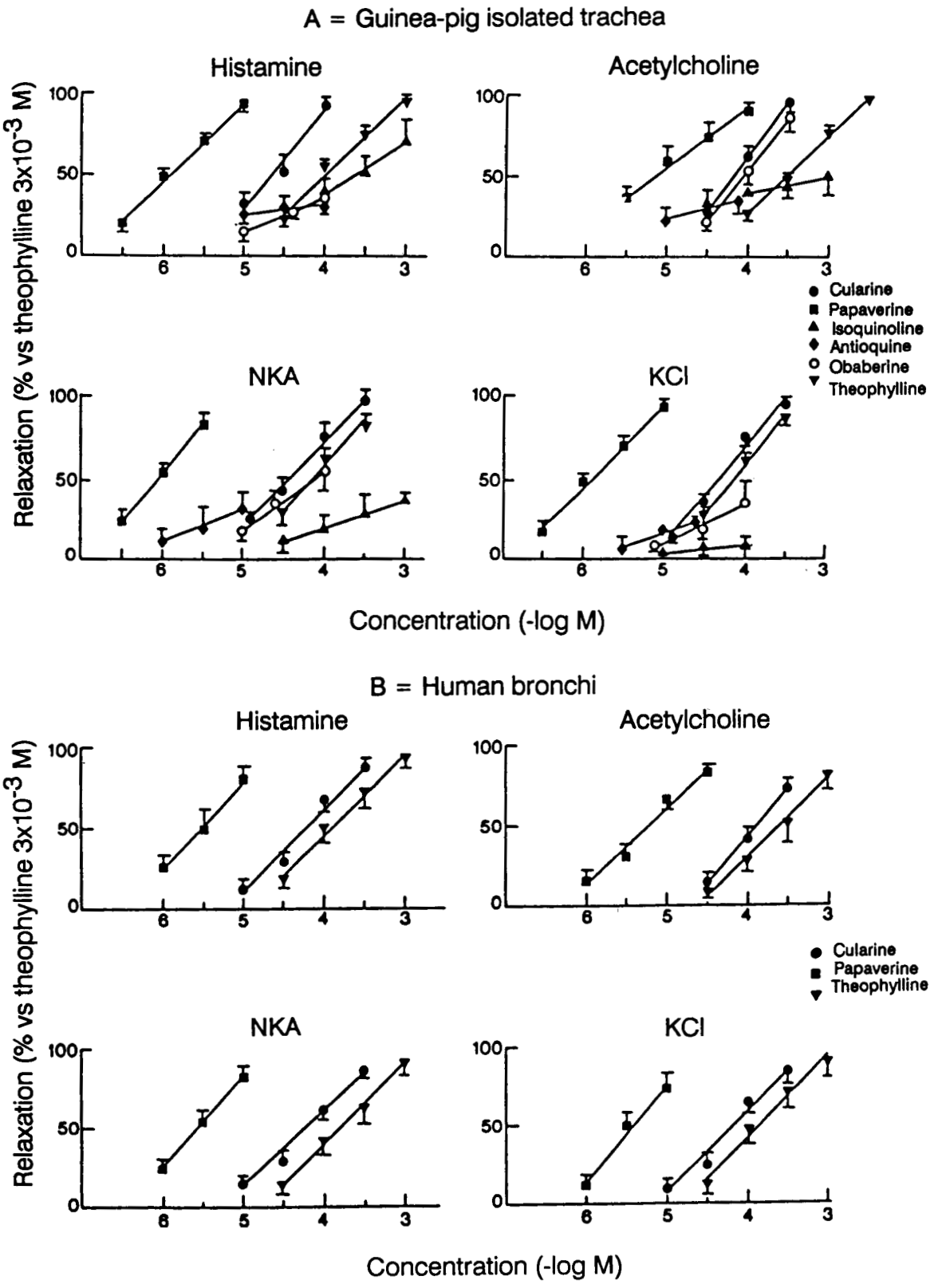


FIG. 2. Effects of cularine (●), 6, 7-dimethoxy-1, 2, 3, 4-tetrahydroisoquinoline (isoquinoline) (▲), antioquine (◆), obaberine (○), papaverine (■) and theophylline (▼) on the contraction of the guinea-pig isolated trachea (A) and human bronchus (B) induced by acetylcholine, histamine, neurokinin A and KCl. Vertical bars indicate s.e. mean of 4 to 6 experiments.

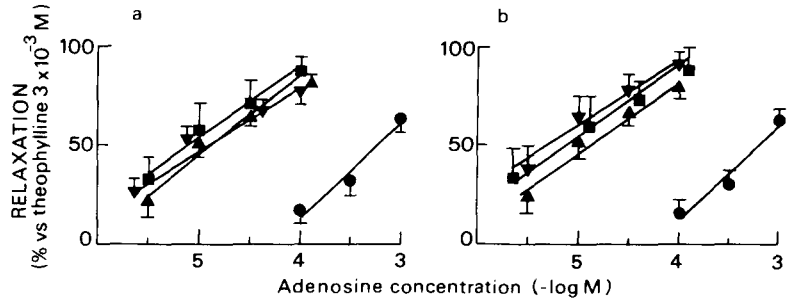


FIG. 3. Effect of papaverine (a) and culatine (b) in the adenosine concentration-response curves on the guinea-pig isolated trachea in the presence of dipyridamole 10^{-5} M. (●) control; (■) dipyridamole 10^{-5} M and dipyridamole 10^{-5} M+(▼) papaverine 3×10^{-7} M or (▲) papaverine 3×10^{-6} M; dipyridamole 10^{-5} M+ (▼) culatine 10^{-5} M and (▲) culatine 3×10^{-5} M. Vertical bars represent s.e. mean of 4 to 5 experiments.

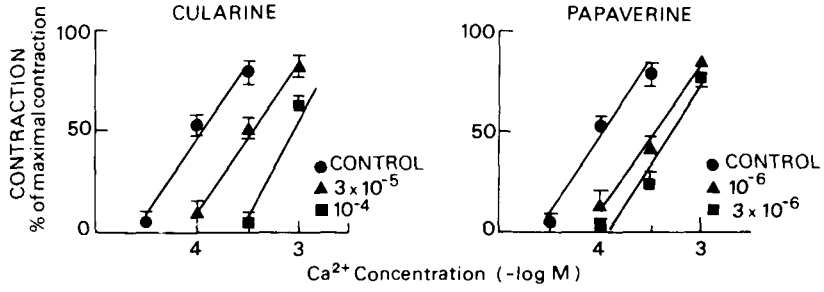


FIG. 4. Ca^{2+} concentration-response curves obtained in guinea-pig isolated trachea in the absence and in the presence of culatine (3×10^{-5} or 10^{-4} M) and papaverine 10^{-6} or 3×10^{-6} M. Vertical bars indicate s.e. mean of 4 to 5 experiments.

of whether the tracheas were under resting tone or precontracted with histamine.

Influence of culatine and papaverine on adenosine concentration-response curves

As has been previously described (Advenier et al 1988), with the guinea-pig isolated trachea precontracted with histamine (10^{-5} M), adenosine exerted a relaxant effect which was potentiated in the presence of dipyridamole (10^{-5} M), the $-\log$ EC50 value obtained in our study being $5{\cdot}06 \pm 0{\cdot}13$ ($n = 7$).

When concentration-response curves to adenosine were constructed in the presence of dipyridamole (10^{-5} M) and papaverine (3×10^{-7} to 3×10^{-6} M) or culatine (10^{-5} to 3×10^{-5} M), no antagonistic activity of culatine or papaverine against adenosine was observed (Fig. 3). The $-\log$ EC50 values obtained for adenosine in the presence of dipyridamole (10^{-5} M) and papaverine (3×10^{-7}, 10^{-6} M or 3×10^{-6} M) were $5{\cdot}08 \pm 0{\cdot}25$ ($n = 5$), $5{\cdot}06 \pm 0{\cdot}10$ ($n = 5$) and $5{\cdot}05 \pm 0{\cdot}09$ ($n = 4$), respectively. In the presence of culatine (10^{-5} and 3×10^{-5} M), the $-\log$ EC50 values obtained were $4{\cdot}97 \pm 0{\cdot}41$ ($n = 4$) and $5{\cdot}30 \pm 2{\cdot}26$ ($n = 4$), respectively. These results showed that papaverine and culatine did not affect the sensitivity of the guinea-pig isolated trachea to adenosine.

Influence of culatine and papaverine on the Ca^{2+} dose-response curves

The effects of culatine and papaverine on the calcium dose-response curves in a potassium-enriched medium are shown in Fig. 4, and the corresponding $-\log$ EC50 values for 3×10^{-4} M Ca^{2+} are given in Table 1. Both alkaloids displaced the calcium dose-response curves to the right, and $-\log$ EC50 values obtained were similar to those calculated for the contractile action of the other agents.

Discussion

Among the isoquinoline alkaloids tested in the present study, culatine was the most potent relaxant. The data obtained showed that in both guinea-pig trachea and human bronchus culatine was as effective as papaverine and theophylline in terms of maximal effect (E_{max}) whereas culatine was more active than theophylline but less active than papaverine in terms of apparent affinity (pD_2) (Table 1).

In addition, the concentrations of culatine needed to inhibit the contractile actions of ACh, histamine or NKA were similar to those which inhibited contractions induced by the depolarizing agent KCl. In bronchial smooth muscle it has been shown that contractile responses to ACh, histamine and NKA are mediated predominantly by Ca^{2+} derived from intracellular stores, whereas contractions induced by the depolarizing agent KCl are closely dependent on an influx of extracellular calcium, the latter being specifically inhibited by calcium antagonists (Cerrina et al 1983; Advenier et al 1984, 1986; Allen et al 1985; Matran et al 1988). Thus, all our results suggest that, in bronchial tissues, the behaviour of culatine is close to that of the other spasmolytic agents tested, papaverine and theophylline, and clearly differs from the behaviour of calcium antagonists in these preparations. The fact that culatine antagonized calcium at concentrations which inhibited the contractile actions of the other agents, also indicates that it is a non-specific antispasmogenic agent.

Thus, cularine appears to be quite different from antioquine, which was shown to present calcium-antagonist activity on the rat uterus (D'Ocón et al 1989).

It may be observed that the relaxant action of cularine and papaverine resembles that of theophylline in several respects, but since theophylline displays adenosine antagonism (Persson 1987; Advenier et al 1988), which may explain many of its side effects, we have studied the possible antagonism of adenosine effects by cularine and papaverine. By showing that the adenosine concentration-response curves were not displaced by either papaverine or cularine, our results support previous suggestions that the adenosine antagonism exerted by theophylline is unrelated to the bronchodilator action of this drug (Persson 1987).

Our results show finally that the $-\log EC50$ and E_{max} values of cularine, papaverine or theophylline were not modified after epithelium removal, either under spontaneous tone or after histamine-induced contraction, as observed with papaverine (Farmer et al 1986). Conversely, Goldie et al (1986) and Lundblad & Persson (1988) observed that epithelium removal does not modify the EC50 of theophylline, but reduces its E_{max}.

Comparing the chemical structures and the inhibitory effects on bronchial smooth muscle of the isoquinoline alkaloids tested, it may be concluded that: (1) the two bisbenzylisoquinoline structures tested, antioquine and obaberine, and 6, 7-dimethoxy-1, 2, 3, 4-tetrahydroisoquinoline had lower relaxant activities than papaverine, and (2) cularine was the most potent of the alkaloids tested but also showed a lower spasmolytic activity than papaverine. All these data indicate the apparent importance of the benzyl ring and of the oxidation state of the pyridine ring in the smooth muscle relaxant effects of isoquinoline compounds. In this sense, the loss of free rotation of the benzyl ring and the hydrogenation of the pyridine ring (cularine) produce a reduction in relaxant activity whereas the elimination of the benzyl ring (6, 7-dimethoxy-1, 2, 3, 4-tetrahydroisoquinoline) causes a virtually total loss of activity.

Acknowledgements

The authors are grateful to Dr M. C. Villaverde and Dr L. Castedo for the generous gift of cularine.

References

Advenier, C., Cerrina, J., Duroux, P., Floch, A., Renier, A. (1984) Effects of five different organic calcium antagonists on guinea-pig isolated trachea. Br. J. Pharmacol. 82: 727–733

Advenier, C., Naline, E., Renier, A. (1986) Effects of Bay k 8644 on contraction of the human isolated bronchus and guinea-pig isolated trachea. Ibid. 88: 33–39

Advenier, C., Devillier, P., Matran, R., Naline, E. (1988) Influence of epithelium on the responsiveness of guinea-pig isolated trachea to adenosine. Ibid. 93: 295–302

Allen, S. L., Foster, R. W., Small, R. C., Towart, R. (1985) The effects of the dihydropyridine Bay k 8644 in guinea-pig isolated trachealis. Ibid. 86: 171–180

Brink, G., Grimaud, C., Guillot, C., Orehek, J. (1980) The interaction between indomethacin and contractile agents on human isolated airway muscle. Ibid. 69: 383–388

Calixto, J. B., Loch, S. (1985) Ketamine-inhibition of calcium-induced contractions in depolarized rat uterus: comparison with other calcium antagonists. Ibid. 85: 189–195

Castedo, L. (1985) The Chemistry and Pharmacology of cularine alkaloids. In: Phillipson, J. D., Roberts, M. F., Zenk, M. H. (eds). The Chemistry and Biology of Isoquinoline Alkaloids. Springer-Verlag, Berlin-Heidelberg, pp 102–125

Cerrina, A., Advenier, C., Renier, A., Floch, A., Duroux, P. (1983) Effects of diltiazem and other Ca^{2+} antagonists on guinea-pig tracheal muscle. Eur. J. Pharmacol. 94: 241–249

Cortes, D., Saez, J., Hocquemiller, R., Cavé, A. (1985) Alcaloïdes des annonacées, LIII. Alcaloïdes du Pseudoxandra aff. lucida. Etude de l'antioquine et de ses dérivés. J. Nat. Prod. 48: 76–85

Devillier, P., Advenier, C., Drapeau, G., Marsac, J., Regoli, D. (1988) Comparison of the effects of epithelium removal and of an enkephalinase inhibitor on the neurokinin-induced contractions of guinea-pig isolated trachea. Br. J. Pharmacol. 94: 675–684

D'Ocón, M. P., Candenas, M. L., Anselmi, E., Zafra-Polo, M. C., Cortes, D. (1989) Antioquine: a new bisbenzylisoquinoline alkaloid with calcium antagonist activity. Arch. Int. Pharmacodyn. Ther. 297: 205–216

Emmerson, J., Mackay, D. (1979) The zig-zag tracheal strip. J. Pharm. Pharmacol. 31: 798

Farmer, S. G., Fedan, J. S., Hay, D. W. P., Raeburn, D. (1986) The effects of epithelium removal on the sensitivity of guinea-pig isolated trachealis to bronchodilator drugs. Br. J. Pharmacol. 89: 407–414

Fujioka, M. (1984) Lack of a causal relationship between the vasodilator effect of papaverine and cyclic AMP production in the dog basilar artery. Ibid. 83: 113–124

Ghelani, A. M., Holroyde, M. C., Sheard, P. (1980) Response of human isolated bronchial and lung parenchymal strips to SRS-A and other mediators of asthmatic bronchospasm. Ibid. 71: 107–112

Godfraind, T., Kaba, A., Polster, P. (1968) Differences in sensitivity of arterial smooth muscles to inhibition of their contractile response to depolarization by potassium. Arch. Int. Pharmacodyn. Ther. 172: 235–239

Goldie, R. G., Papadimitriou, J. M., Paterson, J. W., Rigby, P. J., Self, H. M., Spina, D. (1986) Influence of the epithelium on responsiveness of guinea-pig isolated trachea to contractile and relaxant agonists. Br. J. Pharmacol. 87: 5–14

Guillot, C., Fornaris, M., Badier, M., Orehek, J. (1984) Spontaneous and provoked resistance to isoproterenol in isolated human bronchi. J. Allergy Clin. Immunol. 74: 713–718

Hay, D. W. P., Farmer, S. G., Muccitelli, R. M., Raeburn, D., Fedan, J. S. (1986a) Influence of the epithelium on relaxation responses of the guinea-pig trachea. Fed. Proc. 45: 985

Hay, D. W. P., Raeburn, D., Farmer, S. G., Fleming, W. N., Fedan, J. S. (1986b) Epithelium modulates the reactivity of ovalbumin-sensitized guinea-pig airway smooth muscle. Life Sci., 38: 2461–2468

Lundblad, K. A. L., Persson, C. G. A. (1988) The epithelium and the pharmacology of guinea-pig tracheal tone in vitro. Br. J. Pharmacol. 93: 909–917

Matran, R., Naline, E., Advenier, C., Lockhart, A., Tricot, J. F., Regoli, D. (1988) Role of extracellular calcium in the effects of substance P and neurokinin A on guinea-pig trachea and human bronchus. Fundam. Clin. Pharmacol. 2: 47–55

Persson, C. G. A. (1987) The pharmacology of anti-asthmatic xanthines and the role of adenosine. Asthma Rev. 1: 61–94

Raeburn, D., Hay, D. W. P., Farmer, S. G., Fedan, J. S. (1986) Epithelium removal increases the reactivity of human isolated tracheal muscle to methacoline and reduces the effect of verapamil. Eur. J. Pharmacol. 123: 451–454

Tschirhart, E., Landry, Y. (1986) Airway epithelium does release a relaxant factor: demonstration with substance P. Ibid. 132: 103–104

Vincenc, K., Hwang, O., Van Breemen, C. (1984) Relaxation and contraction responses to histamine in the human lung parenchymal strip. Ibid. 98: 201–210

Zhang, M.-S., Zhou, E.-F. (1988) Methylflavonolamine hydrochloride inhibits contractions induced by noradrenaline, calcium and potassium in rabbit isolated aortic strips. Br. J. Pharmacol. 94: 1184–1188