Br. J. clin. Pharmac. (1988), **26**, 703–708

Comparative effects of rilmenidine and clonidine on bronchial responses to histamine in asthmatic subjects

A. T. DINH XUAN[1], R. MATRAN[1], J. REGNARD[1], P. VITOU[2], C. ADVENIER[3] & A. LOCKHART[1]
[1]Laboratoire de Physiologie et d'Explorations Fonctionnelles Respiratoires, Hôpital Cochin, Paris, [2]Institut de Recherches Internationales Servier, Neuilly sur Seine and [3]Laboratoire de Pharmacologie, Faculté de Médecine Paris Ouest, Paris, France

1 The effects of pretreatment with clonidine and rilmenidine, a new α_2-adrenoceptor agonist, on the bronchial responses to inhaled histamine were studied on 3 different days in a controlled, double-blind, randomized study in 12 asymptomatic asthmatic subjects. Clonidine and rilmenidine were orally administered as single and equipotent doses of 150 μg and 1 mg, respectively. All the subjects were non-smokers with normal lung function tests (forced expiratory volume in one second (FEV_1) = 97 ± 10% predicted FEV_1).
2 Histamine (first dose = 543 nmol) was delivered by a breath activated dosimeter (DeVilbiss no. 646 nebulizer) every 5 min; FEV_1 was measured in triplicate after each dose and the largest value was analysed. The three dose-response curves were compared by analysis of variance.
3 Both clonidine and rilmenidine decreased arterial blood pressure in all subjects. There was no difference in baseline values and pre-challenge values of FEV_1 after placebo, clonidine and rilmenidine on the 3 study days. Compared with placebo, both rilmenidine and clonidine significantly increased the bronchial responses to histamine ($P < 0.05$ and $P < 0.01$ respectively) an effect which was significantly more marked with clonidine than rilmenidine ($P < 0.05$).
4 We suggest that the enhancement of bronchial responsiveness to histamine by clonidine and rilmenidine may result from their effects on both central and peripheral α_2-adrenoceptors, and that the lesser aggravation of histamine-induced bronchial obstruction in asthmatic subjects on rilmenidine might be explained by its lesser central and/or greater peripheral effects than clonidine.

Keywords asthma α_2-adrenoceptors bronchial hyperreactivity clonidine neural control of the airways rilmenidine

Introduction

α_2-adrenoceptors are widely distributed and are present on central and peripheral neurons, on vascular smooth muscle and on endocrine tissue (Brown, 1988). Stimulation of central α_2-adrenoceptors increases parasympathetic outflow and reduces sympathetic outflow from the central nervous system (Schmitt, 1977) and thus causes a decrease in blood pressure and heart rate. The parasympathetic system increases bronchial tone and the sympathetic system reduces it (Boushey *et al.*, 1980; Nadel & Barnes, 1984).

Correspondence: Dr A. T. Dinh Xuan, Laboratoire d'Explorations Fonctionnelles, Hôpital Cochin, 27 rue du Faubourg St Jacques, 75014 Paris, France

There are few available data on the effects of α_2-adrenoceptor agonists on human bronchi *in vivo*, and, to our knowledge, only clonidine has been studied (Lindgren *et al.*, 1986; Dinh Xuan *et al.*, 1988). In asthmatic subjects, clonidine (75 μg) as an aerosol caused a slight improvement in resting bronchial obstruction and a marked reduction in the magnitude of the early bronchial response to inhaled antigen (Lindgren *et al.*, 1986) whereas intravenous clonidine (75–150 μg) caused no fall in peak expiratory flow rate (Salorinne & Poppius, quoted by Lindgren, 1987). Conversely, we have found, with a single dose of oral clonidine, a slight though significant aggravation of the bronchial response to histamine in mild asthmatic patients (Dinh Xuan *et al.*, 1988). Furthermore, a single case of acute asthma possibly related to the oral intake of clonidine has been reported in a child (Ashkenazi *et al.*, 1984).

Results of studies in animals are also controversial. Intravenous administration of clonidine aggravated in a dose-dependent manner the bronchial obstruction caused in guinea pigs by histamine, serotonin and acetylcholine *in vivo* (Advenier *et al.*, 1983). Conversely, clonidine as an aerosol reduced the acute bronchial obstruction caused by ovalbumin in sensitized guinea pigs and the bronchospasm caused by vagal stimulation in anaesthetized guinea pigs, an effect which was antagonized by yohimbine (Andersson *et al.*, 1986).

Rilmenidine (S 3341) is a recently developed oxazoline derivative which is somewhat more selective for α_2-adrenoceptor than clonidine (Van Zwieten, 1988) and whose antihypertensive activity may not be exclusively centrally mediated (Laubie *et al.*, 1985; Van Zwieten *et al.*, 1986; Koenig-Berard *et al.*, 1988). Rilmenidine causes a lesser enhancement of histamine-induced bronchial obstruction than clonidine in guinea pigs (Macquin-Mavier *et al.*, 1988). Therefore, we compared the effect of rilmenidine with that of clonidine on the bronchial response to histamine in asthmatic subjects.

Methods

Subjects

We studied 12 asthmatic, non-smoking subjects (six males and six females), aged 25 (±3) years old (mean ± s.d.) and whose group average forced expiratory volume in one second (FEV_1) was 97 ± 10% predicted FEV_1 (mean ± s.d.). All suffered from mild asthma and had a personal and/or familial history of atopy. The diagnosis of personal atopy was based upon a history of either rhinitis, conjunctivitis or eczema and was confirmed by positive prick-tests with common inhaled allergens. All had normal resting lung function that did not vary during the study. None of the subjects was receiving anti-asthma medication on a regular basis. Ten of them had required occasional use of inhaled β_2-adrenoceptor bronchodilators and/or cromoglycate during the past year. All therapy was suspended 72 h before the study day according to the recommendations of the American Thoracic Society (1980). The study was approved by the Ethics Committee of Cochin Medical School (Paris V University) and all participants volunteered for the study after giving informed consent.

Outline of the study

Each subject was studied on 3 different days at least 1 week apart with an interval between the first and the last study day of less than 1 month. In all subjects dose-response curves for the effects on FEV_1 of serially doubling doses of histamine after pretreatment of either placebo, rilmenidine or clonidine were obtained at the same time of the day. The study was double-blind and randomized according to a latin square design. All subjects abstained from drinking tea and coffee between the preceding evening and the end of each trial.

Each trial was carried out in the morning and started with the measurement of flow-volume curves. Immediately after, the subjects ingested either placebo, clonidine or rilmenidine administered as single dose of 150 μg and 1 mg, respectively, which are the recommended doses in the treatment of arterial hypertension. They remained comfortably seated throughout the study. Heart rate and blood pressure were then measured every 30 min. Again at 2 h, we measured flow-volume curves which were followed by the histamine challenge which consisted of measuring FEV_1 as a function of stepwise doubling doses of histamine. Thus, we established the dose-response curve between 130 and about 150 min after ingestion of clonidine and rilmenidine when their blood concentration is near maximal (Schmitt, 1977; Genissel *et al.*, 1988).

Technical details

Spirometric measurements In order to avoid any operator bias, we used an automated electronic spirometer (Autospiro AS 500, Minato) to measure FEV_1 at baseline, 2 h after dosing and during the bronchial challenge. At all time

points, three determinations of FEV_1 were obtained and the highest value was retained.

Bronchial challenge The inhalation tests were performed according to standard recommendations (Eiser *et al.*, 1983). Histamine dichloride (molecular weight = 184 g), diluted in saline at a concentration (weight/volume) of 10 mg ml^{-1} was delivered by breath activated dosimeter (Rosenthal-French, model D-2A) and nebulizer (DeVilbiss no. 646, Laboratory for Applied Immunology, Inc., Baltimore, MD). The aerosol was produced by an oxygen flow of 1.38 kPa and inhaled during tidal breathing with the patient's nose occluded. Each activation of the dosimeter delivered a preset quantity of nebulized solution which was determined beforehand by weighing (Mettler balance, Inc., Zurich). The first dose of histamine was 543 nmol. Thereafter 1 to 4 successive doubling doses of histamine were administered at intervals of 5 min in order to obtain a fall in $FEV_1 \geqslant 20\%$ of initial FEV_1 on the first study day and the same doses were used on the next two study days.

Individual non-cumulative dose-response curves were plotted by hand and the log dose of histamine causing a 20% fall ($\log PD_{20}$) from the pre-challenge FEV_1 was determined by interpolation.

Statistical analysis

Homogeneity of basal FEV_1 on the 3 study days was tested by a two-way analysis of variance (latin square design). Pre-challenge values obtained 2 h after each pretreatment were compared with baseline values by a two-way analysis of variance (latin square design). Dose-response curves were fitted by linear regression (least-squares method) applied to variation of FEV_1 as a function of increasing doses of histamine and the regression lines on the 3 study days compared by conventional methods. PD_{20} were compared by the non-parametric Friedman test. All results were expressed as mean ± s.d.

Results

There was no modification in resting FEV_1 after placebo, rilmenidine and clonidine since baseline values and pre-challenge values of FEV_1 did not change significantly on the 3 study days (Figure 1). Compared with placebo clonidine caused in all subjects some aggravation of the histamine-induced bronchial obstruction ($P < 0.01$) (Table 1 and Figure 1). Rilmenidine reduced the histamine-induced bronchial obstruction in one subject (number 12), but histamine caused a greater fall in FEV_1 on rilmenidine than on placebo ($P < 0.05$) in the remaining 11 subjects (Table 1 and Figure 1). Clonidine enhanced the bronchial response to histamine more than rilmenidine. In 10 subjects the dose-response curve on clonidine was shifted to the left compared with that on rilmenidine whereas the two curves were superimposable in the remaining two subjects (numbers 2 and 5) (Figure 1). On average, the fall in FEV_1 was greater and the PD_{20} smaller on clonidine than rilmenidine and on rilmenidine than placebo (Table 1). The maximal change in FEV_1 after the highest dose of histamine used was 44.9 ± 15.5, 34.4 ± 15.4 and 23.3 ± 10.5% (mean ± s.d.) of initial FEV_1 on clonidine, rilmenidine and placebo, respectively (Table 1).

Although the above mentioned differences were consistent and significant ($P < 0.05$), they only reflected small changes in bronchial responsiveness to histamine on clonidine and, even more, on rilmenidine. The maximal fall in FEV_1 (% of initial FEV_1) was 1.42 ± 0.46 fold (mean ± s.d.) greater on clonidine than rilmenidine and was also greater by 2.13 ± 0.71 fold and 1.57 ± 0.47 fold (mean ± s.d) on clonidine and rilmenidine than placebo, respectively.

Rilmenidine and clonidine caused a fall in arterial blood pressure in all subjects ($P < 0.01$) with a decrease in systolic and diastolic blood pressure of 18 ± 2 mm Hg and 5 ± 2 mm Hg, respectively, on rilmenidine and of 22 ± 4 mm Hg and 9 ± 3 mm Hg, respectively, on clonidine. Both systolic and diastolic pressure did not significantly change on placebo, decreasing by 6 ± 3 and 1 ± 3 mm Hg, respectively. A slight reduction of heart rate during the 2 h following oral intake of both clonidine and rilmenidine took place in all subjects. However the overall change in resting heart rate was not significant. At the onset of the bronchial challenge, i.e. 2 h after dosing, the fall in systolic and diastolic blood pressure was the same on rilmenidine and clonidine.

Although the study was double-blind, some of the subjects were aware that they had taken an active compound because of side-effects such as thirst, dryness of the mouth and above all, drowsiness. None of these side-effects could be specifically ascribed to clonidine or rilmenidine. However, answering to a questionnaire at the end of each study day, six and four subjects complained of marked and moderate drowsiness, respectively, on clonidine, whereas only two and seven complained of marked and mild drowsiness, respectively, on rilmenidine. A very slight somnolence was reported by two subjects on placebo.

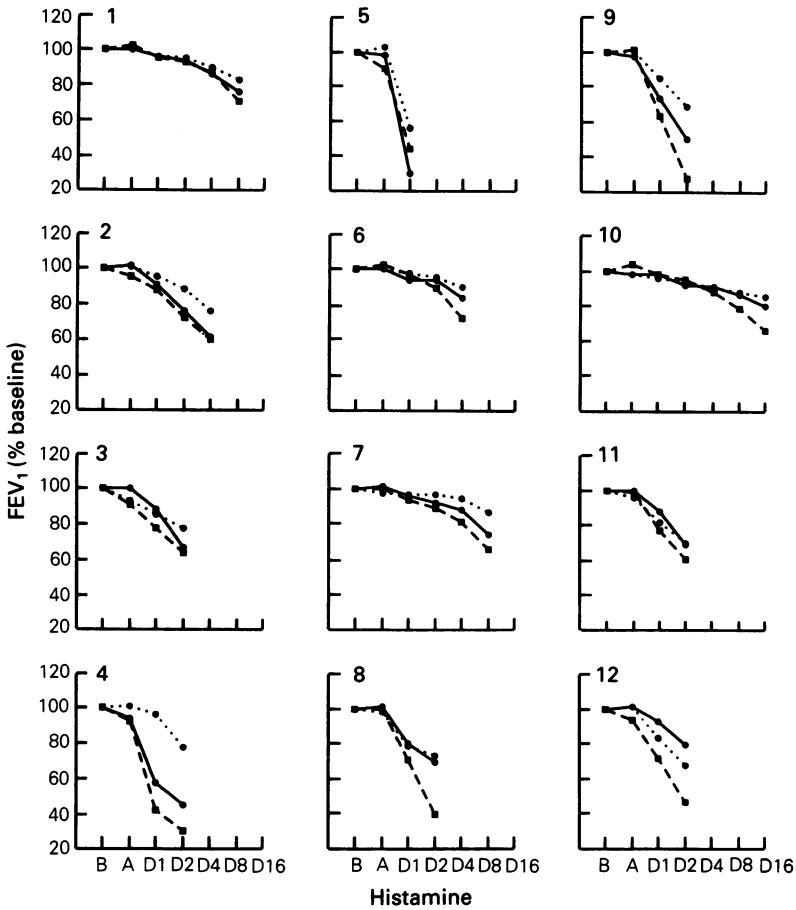

Figure 1 Individual dose-response curves for the effects on FEV_1 (% baseline FEV_1) of successive doubling doses of histamine (D1 = 543 nmol, D16 = 8688 nmol) after pretreatment of placebo (. . . . .), rilmenidine (——) or clonidine (----). B and A: FEV_1 before and 120 min after oral intake of pretreatment.

Table 1 Fall in forced expiratory volume in one second (FEV_1) as % of predicted and as % of baseline FEV_1 and doses of histamine causing a 20% fall from prechallenge FEV_1 (PD_{20}) on placebo (P), rilmenidine (R) and clonidine (C). Results are expressed as mean ± s.d. *P* values were obtained by analysis of variance

	Placebo	*Rilmenidine*	*Clonidine*	P *P–R*	*P–C*	*R–C*
FEV_1 (% predicted)	21.6 ± 9.4	30.8 ± 11.9	40.2 ± 14.3	*	**	*
FEV_1 (% baseline)	23.3 ± 10.5	34.4 ± 15.4	44.9 ± 15.5	*	**	*
log PD_{20}	3.35 ± 0.79	3.03 ± 0.58	2.83 ± 0.45	*	**	*

* $P < 0.05$; ** $P < 0.01$.

Discussion

The main result of the present study is that orally administered α_2-adrenoceptor agonists, clonidine and rilmenidine do not modify the basal tone of the airways and enhance the bronchial response to histamine, an effect that is significantly more marked with clonidine than rilmenidine.

The use of FEV_1 to assess the bronchial response may lead to underestimation of the bronchial obstruction caused by histamine (Fish *et al.*, 1981). Since the subject acts as his own control, this does not invalidate our results.

The time schedule of the study was chosen according to available data on bioavailability of clonidine (Schmitt, 1977) and rilmenidine (Genissel *et al.*, 1988). A previous study has shown that the absolute bioavailability of rilmenidine is close to 1 and that the maximal plasma concentration of both rilmenidine (Genissel *et al.*, 1988) and clonidine (Schmitt, 1977) is attained within 2 h of oral intake.

Radioligand binding studies have shown a greater affinity for α_2-adrenoceptors of rilmenidine compared with clonidine (Van Zwieten, 1988). However, the overall pharmacologic properties of both drugs are similar although the sedative effect was less with rilmenidine than clonidine (Van Zwieten *et al.*, 1986; Van Zwieten, 1988).

There is some evidence that orally administered clonidine and rilmenidine diffuse into the central nervous system (Schmitt, 1977; Laubie *et al.*, 1985) and are capable of stimulating central α_2-adrenoceptors which causes a concomitant enhancement of the parasympathetic outflow and reduction of the sympathetic one (Schmitt, 1977). Our finding that rilmenidine and clonidine aggravated the bronchial response to histamine is consistent with these central effects of α_2-adrenoceptor agonists since histamine-induced bronchial obstruction in asthmatic subjects is at least in part vagally mediated (Boushey *et al.*, 1980; Holtzman *et al.*, 1980; Nadel & Barnes, 1984).

There is an alternative explanation for the increase in bronchial tone or responsiveness on α-adrenoceptor agonists. Clonidine is capable of contracting isolated tracheal strips through a peripheral action on post-junctional α_2-adrenoceptors in the dog (Barnes *et al.*, 1983) or α_1-adrenoceptors in guinea pigs (Floch & Advenier, 1985), an effect which has not been confirmed with human airways (Matran *et al.*, 1986; Lindgren, 1987).

Conversely, aerosolised clonidine prevented allergen-induced bronchoconstriction in 10 asthmatic subjects (Lindgren *et al.*, 1986). Furthermore, a relaxant effect of clonidine on the airways has been found with isolated bronchi of guinea pigs (Grundström *et al.*, 1981) and human subjects (Grundström & Andersson, 1985) and attributed to activation of prejunctional α_2-adrenoceptors on the cholinergic neurons or to inhibition of release of neuropeptides by afferent C fibres (Grundström *et al.*, 1984) and mediators by mast cells or basophil polymorphonuclears (Andersson *et al.*, 1978).

According to the only *in vivo* study we found in the literature rilmenidine has a less marked effect on the aggravation of histamine-induced bronchial obstruction in guinea pigs than clonidine (Macquin-Mavier *et al.*, 1988). These results are consistent with our observations in man. It is tempting to speculate that the lesser reinforcement of histamine-induced bronchial obstruction by rilmenidine than clonidine is also attributable to a greater stimulation of inhibitory prejunctional α_2-adrenoceptors carried by postganglionic parasympathetic neurons innervating the bronchi (Grundström *et al.*, 1981; 1984; Grundström & Andersson, 1985). An alternative interpretation of our data is that rilmenidine has lesser central and/or greater peripheral effects than clonidine (Koenig-Berard *et al.*, 1988) as suggested by the lesser incidence of sedation on rilmenidine than clonidine (Van Zwieten *et al.*, 1986; Van Zwieten, 1988). Indeed, the precise role of central and peripheral α_2-adrenoceptors involved in the antihypertensive activity of rilmenidine is still debated (Laubie *et al.*, 1985; Van Zwieten *et al.*, 1986; Van Zwieten, 1988). Studies in pithed rats (Laubie *et al.*, 1985; Van Zwieten *et al.*, 1986) and isolated blood vessels (Verbeuren *et al.*, 1986) suggest that rilmenidine is possessed of a greater relaxant effect on smooth muscle than clonidine.

In conclusion, our observation of a reinforcement of the bronchial response to histamine with the two α_2-adrenoceptor agonists studied supports the hypothesis that α_2-adrenoceptors are involved in the nervous control of airways calibre in asthmatic subjects.

The authors express their gratitude to E. Ochsenbein-Mignée, M.D., for her invaluable help with statistical analysis of the data, to E. Millon for typing the manuscript and to M. Dacunha for preparing the figures.

This work has been supported by a grant from Institut de Recherches Internationales Servier.

References

Advenier, C., Floch, A. & Mallard, B. (1983). Bronchopulmonary effects of clonidine on the bronchomotor responses of the guinea pigs. *Eur. J. Pharmac.*, **89**, 85–94.

American Thoracic Society (1980). Guidelines for bronchial inhalation challenges with pharmacologic and antigenic agents, p. 11, *ATS News*, New York.

Andersson, R. G. G., Lindgren, B. R. & Colldahl, H. (1978). Inhibitory effects of imidazolines on histamine liberation from human leucocytes and on tracheal smooth muscle tone. *Acta Pharmac. Tox.*, **42**, 381–387.

Andersson, R. G. G., Fugner, A., Lindgren, B. R. & Mucacevic, G. (1986). Inhibitory effects of clonidine on bronchospasm induced by vagal stimulation or antigen challenge in guinea pigs. *Eur. J. Pharmac.*, **123**, 181–185.

Ashkenazi, S., Mimouni, M., Laron, Z. & Varsano, I. (1984). Asthmatic attack associated with oral clonidine test. *Eur. J. Pediatr.*, **142**, 125.

Barnes, P. J., Skoogh, B. E., Brown, J. K. & Nadel, J. A. (1983). Activation of alpha-adrenergic response in tracheal smooth muscle: a post receptor mechanism. *J. appl. Physiol.: Respirat. Environ. Exercise Physiol.*, **54**, 1469–1476.

Boushey, H. A., Holtzman, M. J., Sheller, J. R. & Nadel, J. A. (1980). State of the art: Bronchial hyperreactivity. *Am. Rev. resp. Dis.*, **121**, 389–413.

Brown, M. J. (1988). Sites of action of alpha$_2$ agonists and antagonists. *Am. J. Cardiol.*, **61**, 18D–21D.

Dinh Xuan, A. T., Regnard, J., Matran, R., Mantrand P., Advenier C. & Lockhart, A. (1988). Effects of clonidine on bronchial responses to histamine in normal and asthmatic subjects. *Eur. resp. J.*, **1**, 345–350.

Eiser, N. M., Kerrebijn, K. F. & Quanjer, P. H. (1983). Guidelines for standardization of bronchial challenges with (nonspecific) bronchoconstricting agents. *Bull. Eur. Physiopath. Resp.*, **19**, 495–514.

Fish, J. E., Ankin, M. G., Kelly, J. F. & Peterman, V.I. (1981). Regulation of bronchomotor tone by lung inflation in asthmatic and non asthmatic subjects. *J. appl. Physiol.: Respirat. Environ. Exercise Physiol.*, **50**, 1079–1086.

Floch, A. & Advenier, C. (1985). Interaction between clonidine and histamine on the guinea pig isolated trachea. *J. Pharm. Pharmac.*, **37**, 913–915.

Genissel, P., Bromet, N., Fourtillan, J. B., Mignot, A. & Albin, H. (1988). Pharmacokinetics of rilmenidine in healthy subjects. *Am. J. Cardiol.*, **61**, 47D–53D.

Grundström, N. & Andersson, R. G. G. (1985). Inhibition of the cholinergic neurotransmission in human airways via prejunctional alpha–2 adrenoceptors. *Acta Physiol. Scand.*, **125**, 513–517.

Grundström, N., Andersson, R. G. G. & Wikberg, J. E. S. (1981). Prejunctional alpha-2-adrenoceptors inhibit contraction of tracheal smooth muscle by inhibiting cholinergic neurotransmission. *Life Sci.*, **28**, 2981–2986.

Grundström, N., Andersson, R. G. G. & Wikberg, J. E. S. (1984). Inhibition of the excitatory non-adrenergic, non-cholinergic neurotransmission in the guinea pig tracheo-bronchial tree mediated by alpha-2 adrenoceptors. *Acta Pharmac. Tox.*, **54**, 8–14.

Holtzman, M. J., Sheller, J. R., Di Meo, M., Nadel, J. A. & Boushey, H. A. (1980). Effect of ganglionic blockade on bronchial reactivity in atopic subjects. *Am. Rev. resp. Dis.*, **122**, 17–25.

Koenig–Berard, E., Tierney, C., Beau, B., Delbarre, G., Lhoste, F. & Labrid, C. Cardiovascular and central nervous system effects of rilmenidine (S 3341) in rats. *Am. J. Cardiol.*, **61**, 22D–31D.

Laubie, M., Poignant, J. C., Scuvee–Moreau, J., Dabire, H., Dresse, A. & Schmitt, H. (1985). Pharmacological properties of (N-dicyclopropylmethyl) amino-2-oxazoline (S 3341) an alpha-2 adrenoceptor agonist. *J. Pharmac. (Paris)*, **16**, 259–278.

Lindgren, B. R. (1987). New aspects of the treatment of inflammatory reactions and broncho-obstruction in bronchial asthma. *Eur. J. resp. Dis.*, **70**, Suppl. 148, 1–72.

Lindgren, B. R., Ekström, T. & Andersson, R. G. G. (1986). The effect of inhaled clonidine in patients with asthma. *Am. Rev. resp. Dis.*, **134**, 266–269.

Macquin-Mavier, I., Clerici, C. & Harf, A. (1988). Effects of rilmenidine on the bronchomotor responses of the guinea pig. *Am. J. Cardiol.*, **61**, 45D–46D.

Matran, R., Naline, E., Advenier, C. & Lockhart, A. (1986). Effects of clonidine and three other alpha-2 adrenergic agonists on in vitro contraction of human bronchi. *Bull. Eur. Physiopath. Respir.*, **22**, Suppl. 8, 154s (abstract).

Nadel, J. A. & Barnes, P. J. (1984). Autonomic regulation of the airways. *Ann. Rev. Med.*, **35**, 451–467.

Schmitt, H. (1977). The pharmacology of clonidine and related products. In *Handbook of experimental pharmacology*, ed. Gross, F., pp. 299–396. Berlin, Heidelberg, New York: Springer Verlag.

Van Zwieten, P. A., Thoolen, M. J., Jonkman, F. A., Wilffert, B., de Jonge, A. & Timmermans, P. B. (1986). Central and peripheral effects of S 3341 [(*N*-dicyclopropylmethyl)-amino-2-oxazoline] in animal models. *Arch. int. Pharmacodyn. Ther.*, **279**, 1, 130–149.

Van Zwieten, P. A. (1988). Pharmacology of the alpha-2 adrenoceptors agonist rilmenidine. *Am. J. Cardiol.*, **61**, 6D–14D.

Verbeuren, T. J., Jordaens, F. H., Zonnekeyn, L. L. & Herman, A. G. (1986). Effect of the amino-oxazoline derivative (S 3341) on pre- and post-junctional alpha-adrenoceptors in isolated blood vessels. *Arch. int. Pharmacodyn. Ther.*, **284**, 1, 38–52.

(Received 7 January 1988, accepted 2 August 1988)