REFERENCES

BOWMAN, W. C., KHAN, H. H. & SAVAGE, A. O. (1977). *J. Pharm. Pharmac.*, **29**, 616–625.
BUTTERFIELD, J. L. & ELLIS, K. O. (1973). *Fedn Proc. Fedn Am. Socs exp. Biol.*, **32**, 772.
CRANEFIELD, P. F. (1975). *The conduction of the cardiac impulse.* Mount Kisco, New York: Futura Publishing Co.
ELLIS, R. H., SIMPSON, P., TATHAM, P., LEIGHTON, M. & WILLIAMS, J. (1975). *Anaesthesia*, **30**, 318–322.
ELLIS, K. O., CASTELLION, A. W., HONKOMP, L. J., WESSELS, F. L., CARPENTER, J. F. & HALLIDAY, R. P. (1973). *J. pharm. Sci.*, **62**, 948–951.
KOCH-WESER, J. & BLINKS, J. R. (1963). *Pharmac. Rev.*, **16**, 601–652.
MEYLER, M. J., WESSELING, H. & AGOSTON, S. (1976). *Eur. J. Pharmac.*, **39**, 127–131.
PINDER, R. M., BROGDEN, R. N., SPEIGHT, T. M. & AVERY, G. S. (1977). *Drugs*, **13**, 3–23.
TASHIRO, N. (1973). *Br. J. Pharmac.*, **48**, 121–127.

Effect of tazolol on β-adrenoceptors in isolated preparations of the guinea-pig and rat

S. J. CLARK†, R. H. POYSER*, *Beecham Pharmaceuticals, Research Division, Medicinal Research Centre, The Pinnacles, Harlow, Essex, CM19 5AD, U.K.*

The concept that β-adrenoceptors consist of two distinct sub-groups, designated β_1 and β_2 (Lands, Arnold & others, 1967) has been substantiated by reports in the literature describing agonists selective for β_2-adrenoceptors, e.g. salbutamol (Cullum, Farmer & others, 1969), and antagonists selective for either β_1-adrenoceptors, e.g. practolol (Dunlop & Shanks, 1968) and atenolol (Barrett, Carter & others, 1973), or β_2-adrenoceptors, e.g. butoxamine (Levy, 1966) and H35/25 (α,4-dimethyl-*N*-isopropylphenylethanolamine) (Levy, 1967). However, few if any compounds have been described which are selective agonists at β_1-adrenoceptors and we were therefore interested in the reports of Strosberg & Roszkowski (1972) and Lockwood & Lum (1974) which suggested that tazolol (1-isopropylamino-3-[thiazol-2-yloxy]propan-2-ol hydrochloride, formerly designated ITP) might fit this role. These groups of workers investigated the cardiovascular and metabolic effects of tazolol in anaesthetized animals (dogs and cats respectively) and concluded that the compound fitted into the category of a selective β_1-adrenoceptor stimulant. Subsequently, Strosberg & Buckley (1974) suggested that the cardiotonic properties of tazolol might be of value for the treatment of heart failure.

From the work described *in vivo* with tazolol, there is some uncertainty as to whether the compound is in fact a pure agonist. In view of these findings, we decided to investigate its activity on β-adrenoceptors using *in vitro* preparations. We chose isolated atrial and tracheal preparations from guinea-pigs (either sex, body weight range 320–570 g) and compared the β-adrenoceptor agonist activity of tazolol with isoprenaline and salbutamol on each tissue. The β_1-adrenoceptor stimulant activity was determined from increases in rate of spontaneously beating whole atria and from increases in the force of contraction of left atria electrically driven at a constant rate. The β_2-adrenoceptor stimulant activity was assessed from the inhibition of electrically-induced constrictions of the trachea.

Atrial preparations were suspended in McEwen's solution (1956) at 32°, aerated with 5% CO_2 in oxygen. A resting tension of 1 g was applied to each preparation and contractions recorded using an isometric force transducer (Dynamometer UF1). The contractions of spontaneously beating atria were used to trigger an instantaneous ratemeter. Stimulation of the isolated left atrium preparation consisted of square wave monophasic pulses of 0·5 ms duration and twice threshold voltage at a frequency of 2·25 Hz. The trachea was set up essentially as described by Farmer & Coleman (1970) to allow measurements of changes of intraluminal pressure in response to transmural electrical stimulation. This stimulation consisted of square wave monophasic pulses of 2 ms duration and supramaximal voltage applied to the tissue for 7 s periods at a frequency of 30 Hz. An interval of 2 min was allowed between each stimulation.

On each preparation, cumulative dose-response curves for the three drugs under investigation were obtained. The bathing fluid was changed between each drug and the order of addition of drugs was randomized between experiments. Since tazolol was suspected of having some antagonist activity, care was taken to make sure that the tissue washing procedure removed it from the bath so that the responses to any subsequent drugs administered were not impaired. This was achieved by administering an effective dose of isoprenaline before and afte tazolol. The responses to the drugs administered on each preparation were expressed as a percentage of the maximum response to isoprenaline and the results obtained are shown graphically in Fig. 1.

The maximum positive inotropic and chronotropic responses to tazolol on the guinea-pig atrial prepara-

* Correspondence.
† Present address: Wellcome Research Laboratories, Langley Court, Beckenham, Kent.

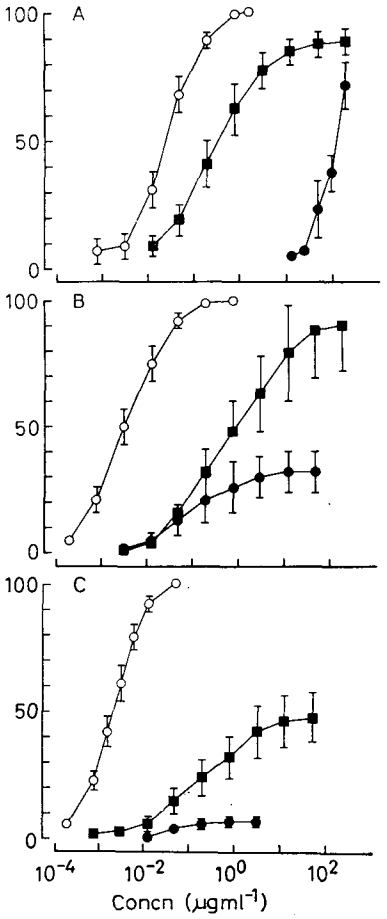

FIG. 1. Dose response curves for isoprenaline (○), salbutamol (■) and tazolol (●) A, in inhibiting increases in intraluminal pressure induced by transmural electrical stimulation of the guinea-pig isolated trachea (%); B, on the rate of the spontaneous beat of isolated whole atria of the guinea-pig (%); C, on the force of contraction of the electrically-driven isolated left atrium of the guinea-pig (%). Responses are expressed as a percentage of the maximum response to isoprenaline. Each point represents the mean ± standard error of at least four experiments.

tions were considerably less than those to isoprenaline indicating that the compound is a partial agonist. In fact, tazolol was less effective than salbutamol, being almost devoid of an agonistic effect on the β_1-adrenoceptors mediating the force of contraction. However, the apparent β_1-adrenoceptor selectivity of tazolol was confirmed in that lower concentrations were required to stimulate spontaneously-beating atria than the tracheal preparations. Isoprenaline also showed some selectivity to cardiac β_1-adrenoceptors, whereas salbutamol, as expected, tended to be more selective (particularly in relation to isoprenaline) to the tracheal β_2-adrenoceptors.

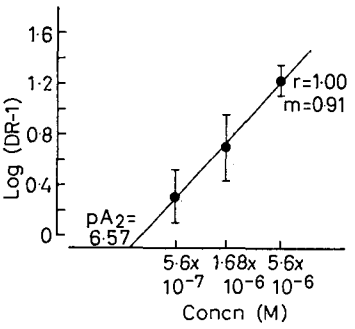

FIG. 2. Antagonism by tazolol of the positive inotropic response induced by isoprenaline in the rat isolated electrically-driven left atrium. Each preparation was exposed for 45 min to one of three concentrations of tazolol ($5{\cdot}6 \times 10^{-7}$, $1{\cdot}68 \times 10^{-6}$ and $5{\cdot}6 \times 10^{-6}$M) and the shift of the isoprenaline dose-response curve expressed as a dose ratio (DR). The calculated regression line for the log (DR-1) against the molar concentration of the antagonist (on a log scale) was plotted, each point representing the mean ± standard error of four preparations. Calculated regression coefficient (r) and slope (m) are shown alongside the calculated regression line. The calculated pA_2 value is shown at the intercept with the antagonist concentration axis.

The partial agonist activity of tazolol at cardiac β_1-adrenoceptors indicates that the compound may also possess antagonist activity at these receptors. Further experiments were therefore conducted to investigate potential β_1-adrenoceptor blocking activity of tazolol. Preliminary work indicated that rat atria rather than guinea-pig atria were more suitable for these experiments as the agonist effects of tazolol were less sustained on the former tissue. Thus electrically-driven left atria of rats (male Sprague-Dawley, body weight range 248–336 g) were used and the antagonist activity of tazolol was assessed from shifts in the cumulative dose-response curve to isoprenaline. It has been previously reported that the second and subsequent cumulative dose-response curves to isoprenaline on isolated atria are superimposable, but are dissimilar in slope to the first curve (Lumley & Broadley, 1975). As similar results were observed in our preliminary experiments, four curves to isoprenaline were obtained on each tissue in these antagonist studies. A single dose of tazolol (in the range $5{\cdot}6 \times 10^{-7}$-$5{\cdot}6 \times 10^{-6}$M) was incubated with the tissue for 45 min between the third and fourth cumulative dose-response curves. Comparisons were always made between the third and fourth curves, responses being expressed as a percentage of the maximum of each dose-response curve. The pA_2 value for tazolol antagonism of the inotropic response to isoprenaline was calculated by the method of Arunlakshana & Schild (1959) (Fig. 2). The calculated regression line intercepted the abscissa at the pA_2 value of 6·57. The calculated regression coefficient and slope both approached unity. These results indicate that tazolol has a β_1-adrenoceptor

blocking activity of the competitive type and it was interesting to note that the pA_2 value was similar to that reported for practolol (Lumley & Broadley, 1975).

Roszkowski, Strosberg & others (1972) observed a tachyphylaxis to tazolol in anaesthetized dogs and proposed an indirect mechanism for the cardiac stimulant effects of the compound. More recently Strosberg (1976) suggested that β-adrenoceptor blocking activity possessed by tazolol may also account for this phenomenon. The results of the present studies tend to support this contention.

In conclusion, this limited *in vitro* study has confirmed the selectivity of tazolol to β_1-adrenoceptors, but has shown that the compound is a partial agonist, and may act as an antagonist, at these receptors.

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REFERENCES

ARUNLAKSHANA, O. & SCHILD, H. O. (1959). *Br. J. Pharmac. Chemother.*, **14**, 48–58.
BARRETT, A. M., CARTER, J., FITZGERALD, J. D., HULL, L. & LE COUNT, D. (1973). *Br. J. Pharmac.*, **48**, 340P.
CULLUM, V. A., FARMER, J. B., JACK, D. & LEVY, G. P. (1969). *Ibid.*, **35**, 141–151.
DUNLOP, D. & SHANKS, R. G. (1968). *Ibid.*, **32**, 201–218.
FARMER, J. B. & COLEMAN, R. A. (1970). *J. Pharm. Pharmac.*, **22**, 46–50.
LANDS, A. M., ARNOLD, A., MCAULIFF, J. P., LUDUENA, F. P. & BROWN, T. G. (1967). *Nature*, **214**, 597–598.
LEVY, B. (1966). *J. Pharmac. exp. Ther.*, **151**, 413–422.
LEVY, B. (1967). *Ibid.*, **156**, 452–462.
LOCKWOOD, R. H. & LUM, B. K. B. (1974). *Life Sci.*, **14**, 73–81.
LUMLEY, P. & BROADLEY, K. J. (1975). *Eur. J. Pharmac.*, **34**, 207–217.
MCEWEN, L. M. (1956). *J. Physiol.*, **131**, 678–689.
ROSZKOWSKI, A. P., STROSBERG, A. M., MILLER, L. M., EDWARDS, J. A., BERKOZ, B., LEWIS, G. S., HALPERN, O. & FRIED, J. H. (1972). *Experientia*, **28**, 1336–1337.
STROSBERG, A. M. (1976). *Archs int. Pharmacodyn. Thér.*, **222**, 200–215.
STROSBERG, A. M. & BUCKLEY, H. T. (1974). *Fedn Proc. Fedn Am. Socs exp. Biol.*, **33**, 503.
STROSBERG, A. M. & ROSZKOWSKI, A. P. (1972). *Ibid.*, **31**, 567.

The roles of presynaptic function and hepatic drug metabolism in the hypothermic actions of two novel dopaminergic agonists

E. K. SILBERGELD*, H. ADLER, S. KENNEDY, D. B. CALNE, *The Experimental Therapeutics Branch, National Institute of Neurological and Communicative Disorders and Stroke, 9000 Rockville Pike, Building 36, Room 5A31, Bethesda, Maryland 20014, U.S.A.*

Hypothermia in rats acclimated to a cold environment has been attributed to stimulation of dopamine receptors in the cns (Yehuda & Wurtman, 1972). Accordingly, measurements of changes in core temperature of cold-acclimated rats has been used to assay for dopaminergic agonist activity of pharmacological compounds (Fuxe, Agnati & others, 1975; Calne, Claveria & Reid, 1975).

We report here the results of temperature studies on two new compounds proposed to have the properties of dopaminergic agonists—lergotrile [(+)-2-chloro-6-methylergoline-8-actonitrile] and 25–397 (9,10-didehydro-6-methyl-8β-(2-pyridylthiomethyl ergoline). These drugs were compared with bromocriptine, an ergot derivative successfully used for the treatment of parkinsonism (Calne, Teychenne & others, 1974). Although the three compounds have been described as dopamine agonists (Fuxe & others, 1975; Lew, Ohashi & Goldstein, 1976; Jaton, Loew & Vigouret, 1976) little is known about their mechanisms of action on dopaminergic pathways or the role of metabolism in their activation or inactivation. The results of some studies suggest that the integrity of presynaptic dopamine terminals is important for the activity of bromocriptine in inducing stereotypy and rotation in rats with unilateral lesions of the nigrostriatal pathway, while interruption of dopamine synthesis does not interfere with lergotrile-induced stereotypy (Pfeiffer & Silbergeld, 1976; Fuxe, Corrodi & others, 1974). Comparable studies have not been done on 25–397. This compound is reported to induce rotation analogously to bromocriptine and apomorphine; however, in contrast to other dopamine agonists, it does not induce stereotypy (Jaton & others, 1976; Silbergeld & Kennedy, unpublished observations).

The relatively long latent period between administration of bromocriptine and 25–397 and induction of their behavioural effects raises the possibility that their

* Correspondence.