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Studies on the peripheral pharmacology of fenazoxine, a potential antidepressant drug

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1. The action of fenazoxine, a cyclized analogue of orphenadrine, has been studied on peripheral tissues innervated by adrenergic and cholinergic nerves. The hypothesis that cyclization of the alkyl amino chain of orphenadrine results in a molecule which retains the noradrenaline sensitizing action of orphenadrine but lacks the antimuscarinic activity has been investigated.

2. The antimuscarinic activity of fenazoxine, on guinea pig ileum, was approximately 1/30 that of orphenadrine.

3. Fenazoxine, desmethylimipramine and cocaine potentiated the response to noradrenaline and sympathetic nerve stimulation on the cat nictitating membrane, isolated rabbit ear artery and isolated driven atrial strip preparations.

4. On the driven atrial strip preparation, fenazoxine at concentrations of 5×10^{-7}M and 1×10^{-6}M produced a small potentiation of the inotropic response to isoprenaline. At 5×10^{-6}M and 5×10^{-5}M fenazoxine antagonized the inotropic response to tyramine.

5. Chronic denervation of the cat nictitating membrane abolished the potentiating action of fenazoxine.

6. The results presented suggest that fenazoxine inhibits the uptake of catecholamines in a manner similar to that reported for desmethylimipramine and cocaine.

7. Evidence is also presented which suggests that fenazoxine, like desmethylimipramine, possesses anti-noradrenaline activity at higher concentrations.

Fenazoxine, 1-phenyl-5-methyl-1, 3, 4, 6 tetrahydro 5-H-benzo (f)-2, 5-oxazocine hydrochloride (Klohs, M. W., Draper, M. D. & Petracek, F. J. ; unpublished), may be considered as a cyclized analogue of the anti-parkinsonian agent orphenadrine (Fig. 1).

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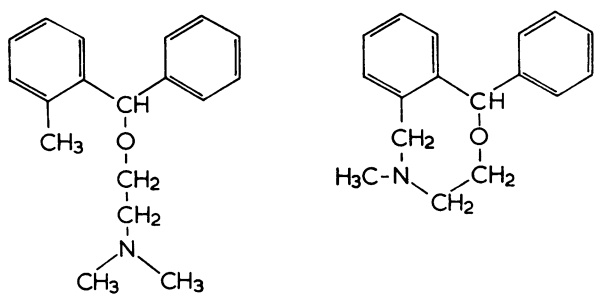
Orphenadrine has been shown to possess a number of pharmacological properties. Bijlsma, Harms, Funcke, Tersteege & Nauta (1956) have demonstrated that orphenadrine has antimuscarinic and antihistaminic activity. More recently, Bassett, Story & Cairncross (1968) showed that orphenadrine potentiated the positive inotropic actions of a number of sympathomimetic amines on the isolated guinea-pig atria preparation. These authors suggested that orphenadrine inhibited the neuronal uptake of certain sympathomimetic amines. In support of this hypothesis, Story & Story (1969) demonstrated that orphenadrine reduced the uptake of tritiated (±)-noradrenaline by isolated guinea-pig atria. It was of interest to note that orphenadrine has been used clinically for the treatment of depression (Bojanovksý & Chloupková, 1961 ; Šedivec & Vánã, 1961). The antidepressant effect of orphenadrine may possibly be related to its ability to inhibit the uptake of noradrenaline at certain synaptic sites.

Carlsson (1966) and Hillarp, Fuxe & Dahlström (1966) suggested that the antidepressants desmethylimipramine and protriptyline act at least partly by blocking the membrane transport of monoamines in neurones of the central nervous system. Glowinski & Axelrod (1966) also postulated that imipramine and other antidepressant drugs exert their effect by preventing the re-uptake of noradrenaline liberated from central neurones, thus increasing its availability. Imipramine and other tricyclic antidepressant agents also possess distinct atropine-like activity (Domenjoz & Theobald, 1959 ; Cairncross, Gershon & Gust, 1963 ; Rathbun & Slater, 1963).

The possibility was considered that altering the molecular configuration of orphenadrine through cyclization of the alkyl amino side chain would result in a molecule with reduced antimuscarinic activity while still retaining the noradrenaline sensitizing properties of the parent compound. Should this be the case, then fenazoxine could be a clinically useful antidepressant agent without the atropine-like side-effects associated with the tricyclic antidepressants (Jarvik, 1966).

The purpose of the present investigation was to evaluate the noradrenaline potentiating activity and the antimuscarinic activity of fenazoxine. The antimuscarinic activity was examined using the isolated guinea-pig ileum. The ability of fenazoxine to sensitize the action of noradrenaline and nerve stimulation was studied on three sympathetically innervated tissues. These preparations were the cat nictitating membrane, the guinea-pig atrial strip and the isolated rabbit ear artery ; the activity of fenazoxine was compared with that of cocaine and desmethylimipramine, two drugs known to block the neuronal uptake of noradrenaline (Iversen, 1965).



Orphenadrine Fenazoxine

FIG. 1. Structure of fenazoxine and orphenadrine.

Methods

Guinea-pig ileum

Antimuscarinic activity was determined by the method of Schild (1947) using a 2 min contact time.

Nictitating membrane of the cat

Cats of either sex, weighing 2·3–4·0 kg, were anaesthetized by the intraperitoneal administration of pentobarbitone, 35 mg/kg. The trachea and left femoral vein were cannulated. Drugs were injected via the femoral vein. Contractions of the nictitating membrane were recorded by an isotonic strain gauge transducer coupled to a Beckman Dynograph recorder, Parameters measured were the contraction amplitude and the recovery time of contraction. The recovery time, $t\frac{1}{2}$, was measured as the time taken for the contraction to decay to half its peak amplitude.

Contractions of the nictitating membrane were elicited by either preganglionic stimulation of the cervical sympathetic nerve or injection of noradrenaline via the femoral vein. Stimuli of 0·5 msec duration were applied to the preganglionic nerve with bipolar platinum electrodes at a frequency of 5 pulses/sec for 10 sec. The stimulus voltage was adjusted to elicit submaximal responses. The interval between responses was sufficient to allow the membrane to return to its normal resting level. Five constant submaximal responses to nerve stimulation or injected noradrenaline were obtained before and after injection of cocaine, desmethylimipramine (DMI), or fenazoxine. The dose of noradrenaline varied between 6·9 and 19·0 μg/kg in normal animals and 28 and 170 ng/kg in denervated animals.

The nictitating membranes of four cats were denervated by removal of the left superior cervical and nodose ganglia at least 3 weeks before conducting the experiment. The operation was carried out under aseptic conditions using pentobarbitone anaesthesia, 35 mg/kg intravenously, with atropine sulphate premedication, 300 μg/kg intravenously.

Electrically driven guinea-pig atrial strip

The technique used was similar to that described by Blinks (1966). Triangular strips, approximately 5 mm wide at the base and 7 mm in length, were cut from the left atrium. The base of the strip was clamped in the electrode assembly and the apex was connected to an isotonic strain gauge transducer. The tissue was immersed in Krebs-Henseleit solution maintained at 32·5° C and gassed with oxygen containing 5% carbon dioxide.

Driving stimuli of 1 msec duration were delivered with a punctate platinum electrode at a frequency of 1 pulse/sec. The voltage used, 1·5 to 3 V, was just sufficient to elicit contractions. Contraction amplitude was recorded on a Brush MK 240 recorder. To overcome any slight inotropic action of the test compound affecting the results the difference between the resting and maximum amplitudes of contraction was measured.

Log. dose-response curves were established by the cumulative method for noradrenaline, isoprenaline or tyramine before and during the presence of cocaine, DMI or fenazoxine. Regression lines were fitted to linear portions of the curves by the method of least squares. Each pair of lines was tested statistically for coincidence and parallelism and the potency ratio was calculated.

Isolated central artery of the rabbit ear

Segments of the central ear artery 2 cm in length were double-cannulated and perfused at a constant flow rate (4–6 ml./min) with Krebs-Henseleit solution maintained at 37° C, according to Method 1 described by de la Lande, Cannell & Waterson (1966). Perfusion pressure was measured with a Statham P23Db pressure transducer coupled to a potentiometric recorder, calibrated 0–200 mm Hg. Periarterial nerve stimulation was delivered by concentric bipolar platinum electrodes. Stimulus parameters were: 2 or 5 pulses/sec, 0·5 msec duration, delivered for 10 sec. The stimulus voltage was supramaximal.

The constrictor responses to exogenous noradrenaline were elicited by leaving the drug in the extraluminal bathing solution until a maximal rise in perfusion pressure was attained. This period varied from 2 to 5 min. Noradrenaline log. dose-response curves were fitted by eye to the responses recorded in triplicate at three dose levels given in latin square sequence.

Cocaine, DMI or fenazoxine was added to the extraluminal bathing fluid and after 15 min equilibration with the tissue the log. dose-response curve was redetermined. The distance between the two curves was measured graphically to give the potency ratio.

Krebs-Henseleit solution

The atrial strip preparation was bathed with physiological solution having the following composition: NaCl, 118 mM ; KCl, 4·7 mM ; $NaHCO_3$, 25·0 mM ; $MgSO_4$, 0·45 mM ; KH_2PO_4, 1·03 mM ; $CaCl_2$, 2·5 mM ; and D (+) glucose, 11·1 mM. The isolated ear artery was bathed with a solution of similar composition with the following differences: $MgSO_4$, 0·53 mM and KH_2PO_4, 1·18 mM.

Drugs used

Atropine sulphate (Knoll), cocaine hydrochloride (Macfarlan Smith), desmethylimipramine hydrochloride (Pertofran ; Geigy), (±) fenazoxine hydrochloride (Riker), (±) isoprenaline hydrochloride (Isuprel ; Winthrop), (−) noradrenaline hydrochloride (Sigma) or bitartrate (Levophed ; Winthrop), (±) orphenadrine citrate (Riker) and tyramine hydrochloride (Sigma). Doses quoted in g/kg refer to the base.

Results

Guinea-pig ileum preparation

The mean pA_2 values (±S.E.) for the acetylcholine response were found to be: fenazoxine, 5·5 ± 0·2 ; orphenadrine, 6·96 ± 0·09 ; atropine, 8·70 ± 0·08.

Cat nictitating membrane preparation

Effects of cocaine, desmethylimipramine and fenazoxine on contractions elicited by preganglionic nerve stimulation. Cocaine, DMI and fenazoxine potentiated the recovery time, and the amplitude of contractions elicited by preganglionic nerve stimulation. The mean percentage increases (±standard error) of the two parameters are shown in Table 1.

Cocaine potentiated both the recovery time and the contraction amplitude over the entire concentration range studied. In contrast, with DMI or fenazoxine, as the injected dose was increased from 1·5 to 4·5 μ-moles/kg, the potentiation of the contraction amplitude was reduced, although the potentiation of the recovery time was further increased.

Experiments with alternate stimulation of the pre- and post-ganglionic cervical sympathetic nerve showed that there was no difference between the actions of fenazoxine on the nictitating membrane contractions elicited by the two methods of stimulation. Similar findings were reported by Haefely, Hürlimann & Thoenen (1964) for the action of cocaine.

Effect of fenazoxine on contractions elicited by noradrenaline in normal and denervated animals. Responses elicited by injection of noradrenaline into normal animals were potentiated by fenazoxine. This potentiation was greater than that observed when contraction was elicited by nerve stimulation (compare Tables 1 and 2).

Denervation completely abolished the potentiation of the contraction amplitude produced by fenazoxine, and greatly reduced the potentiation of recovery time (Table 2). The sensitivity of the membrane to injected noradrenaline was markedly enhanced by denervation (110–470 fold). This is comparable with the enhanced sensitivity reported by Haefely *et al.* (1964).

Electrically driven guinea-pig atrial strip preparation

Effects of cocaine, desmethylimipramine and fenazoxine on the noradrenaline dose-response relationship. Cocaine, DMI and fenazoxine potentiated the inotropic

TABLE 1. *Effect of cocaine, desmethylimipramine and fenazoxine on the responses of the cat nictitating membrane to preganglionic nerve stimulation*

Drug	Dose μ-mole/kg	Mean (±S.E.) % increase in recovery time $t\frac{1}{2}$	Mean (±S.E.) % increase in contraction amplitude
Cocaine (2)	0·15	13± 4	8±3
	1·5	86±11	33±2
	4·5	206±37	59±4
Desmethylimipramine (3)	0·15	19± 1	7±1
	1·5	157± 7	18±2
	4·5	259±21	4±4
Fenazoxine (3)	0·15	11± 1	8±2
	1·5	121± 3	34±4
	4·5	209± 9	29±5

Numbers in brackets indicate the number of experiments.

TABLE 2. *Effect of denervation on the action of fenazoxine on the responses of the cat nictitating membrane elicited by exogenous noradrenaline*

Dose of fenazoxine μ-mole/kg	Mean* % increase in recovery time $t\frac{1}{2}$ (± S.E.)			Mean* % increase in contraction amplitude (± S.E.)		
	Control	Denervated	$P<$	Control	Denervated	$P<$
0·15	63± 8	46± 7	0·20	63± 8	2± 7	0·001
1·5	275±29	50± 7	0·001	195±29	−1± 8	0·001
4·5	461±90	101±17	0·001	242±37	3±11	0·001

* Mean values derived from pooled results of four experiments.

action of noradrenaline. The mean potency ratios obtained at several molar concentrations are shown in Fig. 2. At 5×10^{-8}M, only DMI showed significant potentiation.

At 5×10^{-6}M, however, both cocaine and fenazoxine were more potent than DMI. At 5×10^{-5}M all three compounds exhibited a reduced noradrenaline potentiation relative to that observed at 5×10^{-6}M.

Effect of fenazoxine on the isoprenaline dose-response relationship. Fenazoxine produced a small but significant potentiation of the isoprenaline response at concentrations of 5×10^{-7}M and 1×10^{-6}M. At 5×10^{-5}M fenazoxine reduced the isoprenaline response (see Fig. 2).

Effect of fenazoxine on the tyramine dose-response relationship. The inotropic response of the atrial strip to tyramine was reduced by fenazoxine. At 5×10^{-6}M fenazoxine, the potency ratio was $0{\cdot}05 \pm 0{\cdot}02$. At 5×10^{-5}M the potency ratio was reduced to $0{\cdot}020 \pm 0{\cdot}005$. The figures quoted are the means ($\pm$standard error) of the potency ratios obtained from three experiments.

Isolated central artery of the rabbit ear

Effect of cocaine, desmethylimipramine, and fenazoxine on the response elicited by periarterial stimulation. Addition of cocaine, DMI or fenazoxine to the extraluminal bathing fluid at a concentration of 2×10^{-8}M potentiated the increase in perfusion pressure elicited by periarterial nerve stimulation.

At 2×10^{-6}M, fenazoxine and DMI showed two actions dependent on the duration of contact of the drug with the artery. A marked potentiation of the response to nerve stimulation was seen 2 min after the addition of DMI or fenazoxine. This

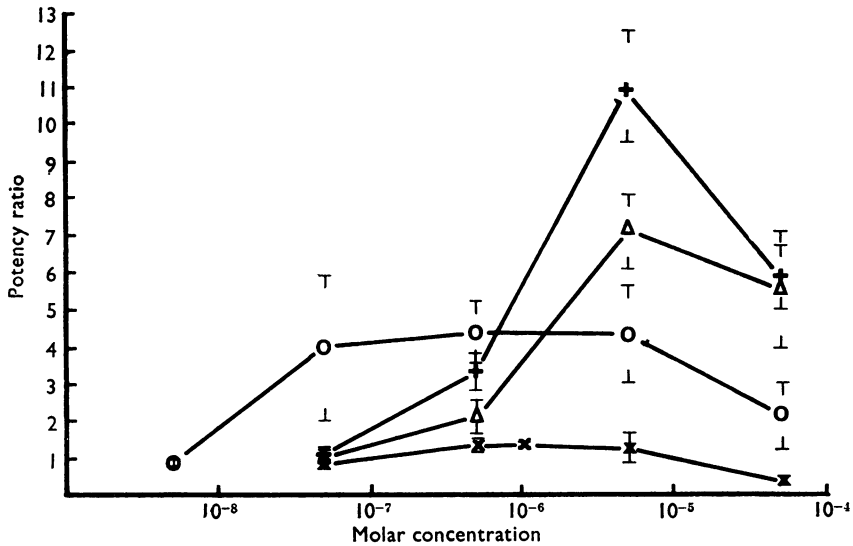


FIG. 2. Driven guinea-pig atrial strip preparation. The effect of various concentrations of cocaine (△——△), desmethylimipramine (○——○) and fenazoxine (+——+) on the noradrenaline potency ratio and of fenazoxine on the isoprenaline potency ratio (×——×). Potency ratios <1 represent antagonism. Vertical bars denote standard errors.

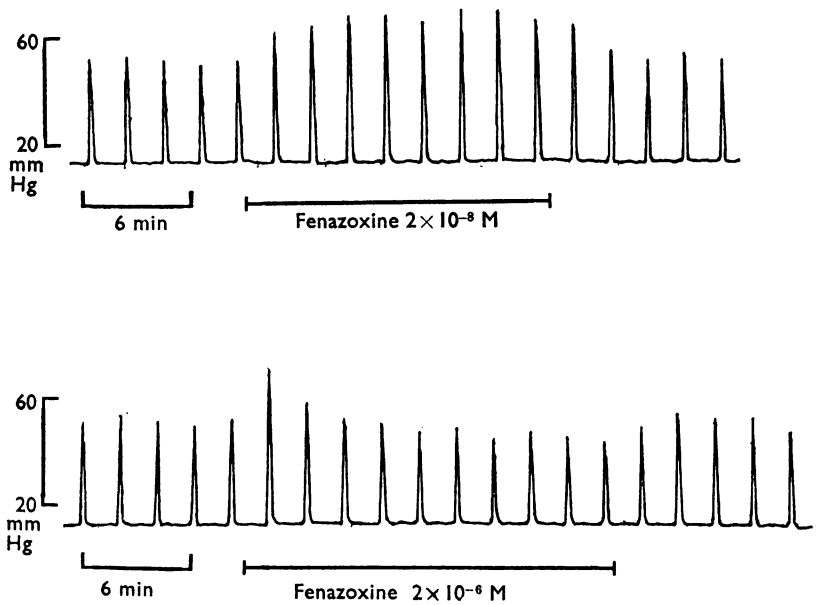


FIG. 3. Modification of the response of the isolated central artery of the rabbit ear to peri-arterial nerve stimulation by fenazoxine at two concentrations. Fenazoxine present in the extraluminal bathing fluid during the period indicated by the horizontal bars. Stimulus parameters: 2 pulses/sec, pulse width 0·5 msec, applied for 10 sec at supramaximal voltage. Abscissa, time: ordinate, pressure.

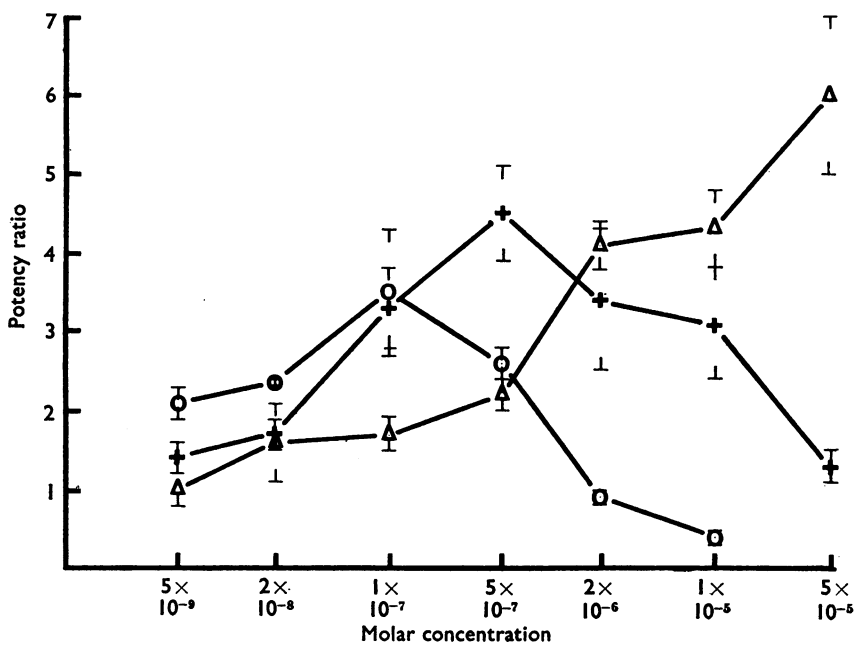


FIG. 4. Isolated rabbit ear artery preparation. The effect of concentration of cocaine (△——△), desmethylimipramine (○——○) and fenazoxine (+——+) on the noradrenaline potency ratio. Potency ratios <1 represent antagonism. Vertical bars denote standard errors.

effect was transient, however, and after incubation for 10–20 min an antagonism was produced. DMI exerted a greater antagonistic effect than fenazoxine. In contrast, cocaine at 2×10^{-6}M only potentiated the response.

Typical effects of fenazoxine at 2×10^{-8}M and 2×10^{-6}M are shown in Fig. 3.

Effects of cocaine, desmethylimipramine and fenazoxine on the responses elicited by exogenous noradrenaline. The results of these experiments are illustrated in Fig. 4.

Cocaine, DMI and fenazoxine potentiated the response to noradrenaline. DMI and fenazoxine produced maximal potentiation at 1×10^{-7}M and 5×10^{-7}M respectively ; the potentiation was reduced in magnitude at higher concentrations. In contrast, the potentiation by cocaine of the noradrenaline response increased with concentration over the entire concentration range investigated.

At concentrations of 2×10^{-6}M and higher, DMI and fenazoxine produced two distinct time-dependent actions on the noradrenaline response. These effects were similar to those previously described for periarterial nerve stimulation.

Discussion

The antimuscarinic activity of fenazoxine was considerably less than that of orphenadrine. It is probable that this reduction in antimuscarinic activity is due to the additional steric constraint imposed on the nitrogen atom of the fenazoxine molecule by ring closure.

Fenazoxine potentiated the responses to sympathetic nerve stimulation and exogenous noradrenaline on sympathetically innervated tissues. This action resembled that of cocaine and DMI. Cocaine (Haefely *et al.*, 1964 ; de la Lande, Frewin & Waterson, 1967) and DMI (Callingham, 1967) potentiate the action of noradrenaline on various peripheral tissues. Further, cocaine and DMI are potent inhibitors of the uptake of labelled noradrenaline into isolated perfused rat heart (Iversen, 1965), suggesting that these drugs potentiate the actions of noradrenaline by inhibiting its uptake into adrenergic nerve endings.

Bonaccorsi & Hrdina (1967) found that cocaine and DMI inhibited the tyramine pressor response of pithed rats. Cocaine also antagonized the tyramine response of rabbit aorta and guinea-pig atria (Furchgott, Kirkepar, Rieker & Schwab, 1963). The accumulation of ^{14}C-tyramine by mouse cortex slices was partially inhibited by cocaine and DMI (Ross & Renyi, 1966). These findings are consistent with the concept of tyramine acting " indirectly " by release of noradrenaline from neuronal stores (Fleckenstein & Burn, 1953 ; Burn & Rand, 1958). In the present studies, fenazoxine inhibited the action of tyramine on the guinea-pig atrial strip preparation.

Since fenazoxine potentiates the action of noradrenaline and antagonizes the action of tyramine, it is suggested that fenazoxine inhibits the neuronal uptake of noradrenaline and tyramine in a manner similar to that reported for cocaine and DMI.

Further evidence for the hypothesis that fenazoxine inhibits neuronal uptake of noradrenaline is obtained from the chronically denervated cat nictitating membrane experiments. Denervation abolished the potentiation of the contraction amplitude observed with fenazoxine, demonstrating that the potentiating action of fenazoxine is dependent on intact adrenergic nerve endings. Similar findings have been reported for cocaine (Haefely *et al.*, 1964).

Fenazoxine produced a small, although statistically significant, potentiation of the inotropic action of isoprenaline on the guinea-pig atrial strip. This potentiation is probably brought about by inhibition of $Uptake_2$, the process whereby isoprenaline is taken up in small amounts by rat heart (Callingham & Burgen, 1966). This possible action of fenazoxine would be in contrast to that of DMI. DMI affects the $Uptake_2$ process only at high concentrations (Iversen, 1965).

The results from the three adrenergically innervated preparations suggest that fenazoxine and DMI have an additional action not attributable to an inhibition of catecholamine uptake. As the concentration of fenazoxine or DMI was increased above a certain level, dependent on the tissue used, the noradrenaline potentiation was reduced. This effect was more marked with DMI, high concentrations of which antagonized the noradrenaline responses of the isolated artery. The inotropic action of isoprenaline on the isolated atrial strip was also antagonized by high concentrations of fenazoxine. Callingham (1967) showed that DMI antagonized the vasoconstrictor action of noradrenaline on the isolated rabbit ear. The suggestion of Bonaccorsi & Hrdina (1967) that DMI may possess "adrenolytic" activity is supported by the present data. These data also suggest that fenazoxine exhibits similar but less potent antagonistic activity. However, the present experiments do not give any indication of the specificity of this antagonism; non-specific depressant activity could account for the observed action. On the chronically denervated cat nictitating membrane, the depressant action of fenazoxine would be expected to antagonize the contraction amplitude. An antagonism was not observed, suggesting that either the nictitating membrane was incompletely denervated or that there had been partial regeneration of adrenergic nerve fibres. This suggestion is supported by the observation that potentiation of contraction recovery time of the nictitating membrane, although markedly diminished, was not abolished.

In the rabbit ear artery experiments, the potentiating and antagonistic actions of fenazoxine and DMI followed different time courses. The potentiating action was rapid in onset while the antagonism developed slowly.

The postulate that cyclization of the alkyl amino side-chain of orphenadrine would result in a compound with reduced antimuscarinic activity and which retained the noradrenaline sensitizing properties of orphenadrine has proved to be correct. The antimuscarinic activity of fenazoxine was only 1/30 of that of orphenadrine, whereas its ability to potentiate the actions of noradrenaline on peripheral tissues was comparable with that of DMI and cocaine. The significance of these findings in relation to the antidepressant activity of fenazoxine is being investigated.

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REFERENCES

BASSETT, J. R., STORY, M. & CAIRNCROSS, K. D. (1968). The influence of orphenadrine upon the actions of a series of sympathomimetic agents. *Eur. J. Pharmac.*, **4**, 198–204.

BIJLSMA, U. G., HARMS, A. F., FUNCKE, A. B. H., TERSTEEGE, H. M. & NAUTA, W. Th. (1956). The pharmacology of β-dimethylaminoethyl-2-methyl-benzhydrylether hydrochloride (BS5930). *Archs int. Pharmacodyn. Thér.*, **106**, 332–369.

BLINKS, J. R. (1966). Field stimulation as a means of effecting the graded release of autonomic transmitters in isolated heart muscle. *J. Pharmac. exp. Ther.*, **151**, 221–235.

BOJANOVSKÝ, J. & CHLOUPKOVÁ, K. (1961). The use of orphenadrine in depressive patients. *Activatis Nervosa Super.*, **3**, 230.

BONACCORSI, A. & HRDINA, P. (1967). Interactions between desipramine and sympathomimetic agents on the cardiovascular system. In *Proceedings of the First International Symposium on Antidepressant Drugs*, ed. Garrattini, S. & Dukes, M. N. G., pp. 149–157. Amsterdam: Excerpta Medica Foundation.

BURN, J. H. & RAND, M. J. (1958). The action of sympathomimetic amines in animals treated with reserpine. *J. Physiol. Lond.*, **144**, 314–336.

CAIRNCROSS, K. D., GERSHON, S. & GUST, I. D. (1963). Some aspects of the mode of action of imipramine. *J. Neuropsychiat.*, **4**, 224–231.

CALLINGHAM, B. A. (1967). The effects of imipramine and related compounds on the uptake of noradrenaline into sympathetic nerve endings. In *Proceedings of the First International Symposium on Antidepressant Drugs*, ed. Garrattini, S. & Dukes, M. N. G., pp. 35–43. Amsterdam: Excerpta Medica Foundation.

CALLINGHAM, B. A. & BURGEN, A. S. V. (1966). The uptake of isoprenaline and noradrenaline by perfused rat heart. *Mol. Pharmac.*, **2**, 37–42.

CARLSSON, A. (1966). Pharmacological depletion of catecholamine stores. *Pharmac. Rev.*, **18**, 541–549.

DE LA LANDE, I. S., CANNELL, V. A. & WATERSON, J. G. (1966). The interaction of serotonin and noradrenaline on the perfused artery. *Br. J. Pharmac. Chemother.*, **28**, 255–272.

DE LA LANDE, I. S., FREWIN, D. & WATERSON, J. G. (1967). The influence of sympathetic innervation on the vascular sensitivity to noradrenaline. *Br. J. Pharmac. Chemother.*, **31**, 82–93.

DOMENJOZ, R. & THEOBALD, W. (1959). Zur pharmakologie des Tofranil (N-(3-dimethyl-aminopropyl)-iminodibenzyl-hydrochlorid). *Archs int. pharmacodyn. Thér.*, **120**, 450–489.

FLECKENSTEIN, A. & BURN, J. H. (1953). The effect of denervation on the action of sympathomimetic amines on the nictitating membrane. *Br. J. Pharmac. Chemother.*, **8**, 69–78.

FURCHGOTT, R. F., KIRKEPAR, S. M., RIEKER, M. & SCHWAB, A. (1963). Actions and interactions of norepinephrine, tyramine and cocaine on aortic strips of rabbit and left atria of guinea-pig and cat. *J. Pharmac. exp. Ther.*, **142**, 39–58.

GLOWINSKI, J. & AXELROD, J. (1966). Effects of drugs on the disposition of H^3-norepinephrine in the rat brain. *Pharmac. Rev.*, **18**, 775–785.

HAEFELY, W., HÜRLIMANN, A. & THOENEN, H. (1964). A quantitative study of the effect of cocaine on the response of the cat nictitating membrane to nerve stimulation and to injected noradrenaline. *Br. J. Pharmac. Chemother.*, **22**, 5–21.

HILLARP, N. A., FUXE, K. & DAHLSTRÖM, A. (1966). Demonstration and mapping of central neurons containing dopamine, noradrenaline and 5-hydroxytryptamine and their reactions to psychopharmaca. *Pharmac. Rev.*, **18**, 727–741.

IVERSEN, L. L. (1965). The uptake of catechol amines at high perfusion concentrations in the rat isolated heart: A novel catechol amine uptake process. *Br. J. Pharmac. Chemother.*, **25**, 18–33.

JARVIK, M. E. (1966). Drugs used in the treatment of psychiatric disorders. In *The Pharmacological Basis of Therapeutics*, ed. Goodman, L. S. & Gilman, A., 3rd ed., pp. 159–214. New York: Macmillan.

RATHBUN, R. C. & SLATER, I. H. (1963). Amitriptyline and nortriptyline as antagonists of central and peripheral cholinergic activation. *Psychopharmac., Berlin*, **4**, 114–125.

ROSS, S. B. & RENYI, A. L. (1966). Uptake of some tritiated sympathomimetic amines by mouse brain cortex slices in vitro. *Acta pharmac. tox.*, **24**, 297–309.

SCHILD, H. O. (1947). pA, a new scale for the measurement of drug antagonism. *Br. J. Pharmac. Chemother.*, **2**, 189–206.

ŠEDIVEC, V. & VÁNÃ, J. (1961). The treatment of depressive states with orphenadrine. *Activatis Nervosa Super.*, **3**, 231.

STORY, D. F. & STORY, M. (1969). Inhibition by orphenadrine of ^{3}H-dl-noradrenaline and ^{14}C-tyramine uptake in atria. *Eur. J. Pharmac.*, **5**, 296–298.

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