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THE EFFECT OF PHENOXYBENZAMINE (DIBENYLINE) ON THE VASCULAR RESPONSE TO SYMPATHOMIMETIC AMINES IN THE FOREARM

BY M. J. ALLWOOD AND JEAN GINSBURG

From The Sherrington School of Physiology, and Department of Medicine, St Thomas's Hospital Medical School, London, S.E. 1

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The fact that adrenaline can cause vasoconstriction or vasodilatation in skeletal muscle has been known since the work of Dale (1906) on ergotoxine. Both these actions are due to a direct effect of adrenaline on the vessels, and do not depend upon an intact nerve supply (Elliott, 1905; Dale & Richards, 1927). Other substances which inhibit the constrictor response, such as chlorpromazine and phenoxybenzamine, also cause a reversal of the pressor action of the amine in animals (Nickerson, Henry & Nomaguchi, 1951; Huidobro, 1954; Green, Denison, Williams, Garvey & Tabor, 1954). In man chlorpromazine and phenoxybenzamine have been shown to reduce the constrictor effects of adrenaline and noradrenaline in the hand (Ginsburg & Duff, 1956, 1958; Duff & Ginsburg, 1957). The present investigation is a quantitative study of the effect of phenoxybenzamine (Dibenyline) on the response to adrenaline, noradrenaline and *iso*propylnoradrenaline in the human forearm. Results have previously been reported in brief (Allwood & Ginsburg, 1959). Our results with adrenaline confirm those obtained in a similar study by de la Lande & Whelan (1959).

METHODS

Thirty-four experiments were performed on twenty-eight healthy medical students of both sexes between the ages of 19 and 26 years. They lay on a comfortable couch, wearing normal indoor attire, in a room maintained at $21 \pm 1°$ C. Blood flow was measured simultaneously in both forearms at intervals of 0·5 min by venous occlusion plethysmography (Barcroft & Swan, 1953). A continuous infusion of 0·9 % NaCl solution was maintained through an indwelling needle inserted into the brachial artery of the test forearm; phenoxybenzamine and the amine under study (adrenaline tartrate, British Drug Houses; L-noradrenaline bitartrate, Bayer and Co.; *iso*propylnoradrenaline HCl, Winthrop) were given intra-arterially through this needle. In some experiments a similar infusion was maintained through a needle inserted into an antecubital vein of the opposite arm, so that the amine could be given intravenously. Ascorbic acid (0·1 mg) was added to each pint (0·57 l.) of saline to protect solutions of amines from oxidation (Gaddum, Peart & Vogt, 1949).

After initial recordings of resting blood flow, a control infusion of the amines under study was given intra-arterially or intravenously for a period of 10 min, except with intra-

arterial noradrenaline which was infused for 5 min. After the blood flow had returned to the resting value phenoxybenzamine hydrochloride (Dibenyline; Smith, Kline and French) was given intra-arterially into the test forearm in a total dose of 3 mg at the rate of 500 μg/min for 6 min. This caused a moderate increase in flow in the treated limb, with no change on the opposite (control) side. After a further control period the intra-arterial or intravenous infusion of the particular amine was then repeated, and blood flow compared with that recorded during the infusion before giving phenoxybenzamine.

Blood flow is expressed in ml. blood/100 ml. tissue/min. In experiments where the amine was given intravenously the blood flow in the test limb has been directly compared with that in the control limb. When the amine was given intra-arterially changes in blood flow have been compared in the same (test) limb before and after receiving phenoxybenzamine, after allowing for phasic 'spontaneous' variations in flow as measured in the contralateral limb (Duff, 1952; Ginsburg & Duff, 1956).

RESULTS

The responses of the forearm blood vessels to intra-arterial and intravenous infusions of sympathomimetic amines, both before and after phenoxybenzamine, are compared in Table 1.

TABLE 1. Effect of sympathomimetic amines on mean forearm blood flow (ml./100 ml./min) before and after phenoxybenzamine

	Intra-arterial infusion					
	Before phenoxybenzamine			After phenoxybenzamine		
	Pre-infusion	During infusion	Difference (%)*	Pre-infusion	During infusion	Difference (%)*
Adrenaline (2 μg/min)	3·2	1·3	− 54	8·1	21·4	+173
Noradrenaline (2 μg/min)	3·5	0·7	− 77	6·3	4·8	− 15
*Iso*propylnoradrenaline (0·1 μg/min)	3·1	11·4	+260	5·1	13·0	+194
	Intravenous infusion, after phenoxybenzamine					
	Control forearm			Test forearm		
	Pre-infusion	During infusion	Difference (%)*	Pre-infusion	During infusion	Difference (%)*
Adrenaline (10 μg/min)	2·7	4·0	+ 51	5·3	14·1	+226
Noradrenaline (10 μg/min)	3·7	2·7	− 23	6·7	7·3	+ 12
*Iso*propylnoradrenaline (4 μg/min)	3·6	5·5	+ 66	5·5	12·4	+175

* These are mean differences and *not* the difference between means. In calculating these values for intra-arterial infusions allowance was made for 'spontaneous' flow variations measured in the control limb (Duff, 1952; Ginsburg & Duff, 1956).

Effect of phenoxybenzamine on intra-arterial adrenaline

This was studied in six subjects; the results are summarized in Fig. 1 (*a*). In the first minute of an intra-arterial infusion of 2 μg/min adrenaline there was an initial transient vasodilatation, followed immediately by a vasoconstriction which lasted until the end of the 10 min infusion period. An 'after-dilatation' was then observed, reaching its maximum about 2 min after the end of infusion.

Intra-arterial phenoxybenzamine (500 μg/min for 6 min) caused a moderate increase in forearm blood flow, so that thereafter resting values in the test limb were about double those before giving phenoxybenzamine. Infusion of intra-arterial adrenaline in the same dosage as before now caused a more marked initial dilatation, with blood flow subsequently

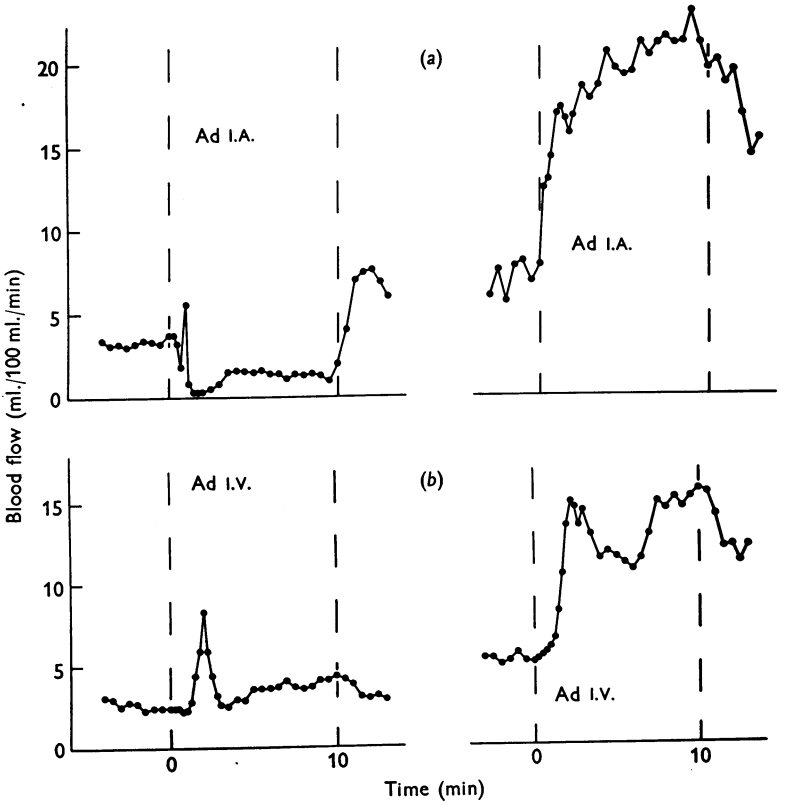

Fig. 1. Forearm blood flow during adrenaline infusions before and after phenoxybenzamine. (*a*) averaged results from six subjects given intra-arterial adrenaline (2 μg/min). (*b*) averaged results from six subjects given intravenous adrenaline (10 μg/min). Between the two parts of the experiments the subjects received intra-arterial phenoxybenzamine (3 mg during 6 min).

remaining at this high level throughout, or increasing further. There was no 'after-dilatation' following the end of this infusion, forearm flow remaining elevated for several minutes before returning slowly towards the resting level. Blood flow in the contralateral (control) forearm remained relatively constant throughout.

Forearm blood flow in the test limb decreased from an average of 3·2 ml./100 ml./min (resting) to 1·3 ml. during the control infusion; after

phenoxybenzamine it increased during administration of adrenaline from 8·1 (resting) to 21·4 ml. Mean changes from the 'expected' flow during adrenaline infusion in the treated limb were − 54 % before and + 173 % after giving phenoxybenzamine, a difference which is significant ($t = 4{\cdot}32$, $P < 0{\cdot}01$).

Intravenous adrenaline. This was studied in six subjects (Fig. 1(*b*)). Changes in the two forearms during the control infusion of 10 μg adrenaline/min for 10 min were similar, and consisted of a transient initial vasodilatation, followed by a decrease in flow and a smaller sustained vasodilatation during the remainder of the infusion.

After phenoxybenzamine the resting flow was about doubled in the test forearm, although unchanged in the control. During the subsequent intravenous infusion of adrenaline the blood flow in the test limb was markedly elevated compared with that in the control infusion, although maintaining a similar pattern of transient and sustained phases. Changes in the contralateral limb were similar to those during the control infusion.

Mean resting blood flow in the control and test forearms before giving phenoxybenzamine was 2·5 and 2·6 ml., increasing to 4·9 and 3·9 ml. respectively during the sustained dilatation. After giving phenoxybenzamine the mean resting blood flow in the control and test forearms was 2·7 and 5·3 ml., increasing respectively during adrenaline infusion to 4·0 and 14·1 ml. Before giving phenoxybenzamine there was no significant difference between the increase in flow on the control and test sides during intravenous adrenaline infusion.

After administration of phenoxybenzamine the increase during adrenaline infusion was unchanged on the control side, but on the treated side the increase of 226 % was significantly greater than the 51 % increase at the corresponding time in the contralateral (control) limb ($t = 2{\cdot}83$, $P < 0{\cdot}02$).

Effect of phenoxybenzamine on intra-arterial noradrenaline

The results obtained on five subjects are summarized in Fig. 2 (*a*). Resting blood flow in the test limb was 3·5 ml./100 ml./min, decreasing to 0·7 ml. during the infusion of noradrenaline 2 μg/min for 5 min. After phenoxybenzamine resting flow increased to 6·3 ml., decreasing to 4·8 ml. during subsequent noradrenaline infusion. Average blood flow in the contralateral (control) limb remained between 3·3 and 3·8 ml. throughout. The mean change from the 'expected' flow in the test forearm during noradrenaline infusion was a decrease of 77 % before and of 15 % after giving phenoxybenzamine, a difference which is significant ($t = 3{\cdot}94$, $P < 0{\cdot}01$).

Intravenous noradrenaline. This was studied in six subjects (Fig. 2(*b*)). Resting blood flow averaged 4·0 ml./100 ml./min in both the control and test forearms, decreasing to 3·0 and 3·6 ml. respectively during intravenous infusion of noradrenaline 10 μg/min for 10 min. After phenoxybenzamine the resting flow in the test limb increased to 6·7 ml. During the subsequent noradrenaline infusion the blood flow on the test side showed

a further slight increase to 7·3 ml., while that in the control forearm decreased from 3·7 to 2·7 ml. Before phenoxybenzamine the changes in blood flow during noradrenaline infusion were similar in the two forearms. After phenoxybenzamine the average change in flow in the test arm was an increase of 12 %, compared with a 23 % decrease in the control forearm.

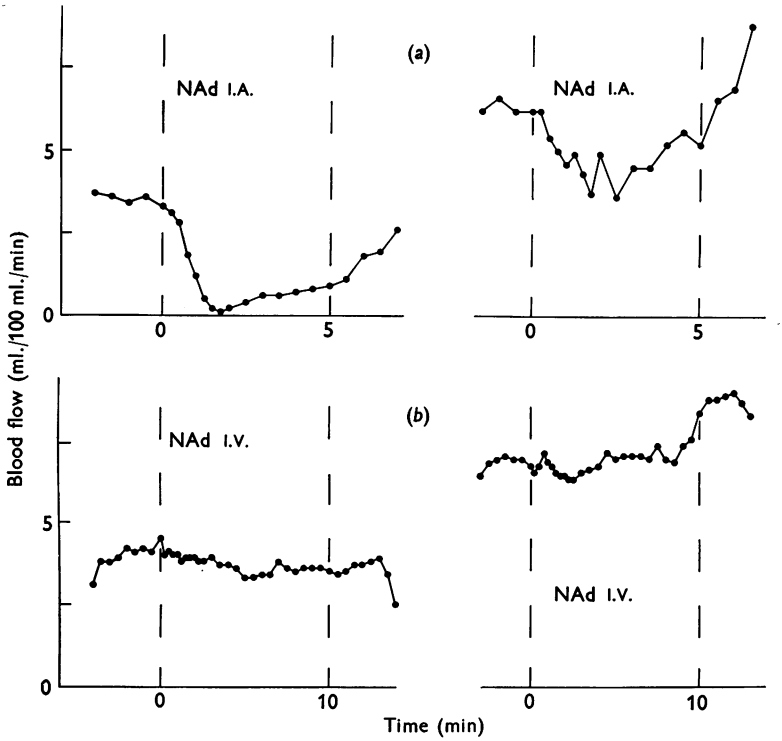

Fig. 2. Forearm blood flow during noradrenaline infusions before and after phenoxybenzamine. (*a*) averaged results from five subjects given intra-arterial noradrenaline (2 μg/min). (*b*) averaged results from six subjects given intravenous noradrenaline (10 μg/min). Between the two parts of the experiments the subjects received intra-arterial phenoxybenzamine (3 mg during 6 min). Note different time scales of (*a*) and (*b*).

This difference is not statistically significant ($t = 2{\cdot}02$, $0{\cdot}1 > P > 0{\cdot}05$). Changes in blood flow in the control forearm during noradrenaline infusion before and after phenoxybenzamine were similar, namely decreases of 21 and 23 % respectively.

Effect of phenoxybenzamine on intra-arterial isopropylnoradrenaline

The results obtained on five subjects are summarized in Fig. 3(*a*). Resting blood flow in the test forearm averaged 3·1 ml./100 ml./min, increasing to 11·4 ml. during intra-arterial infusion of *iso*propylnoradrenaline

0·1 μg/min. After intra-arterial phenoxybenzamine the resting blood flow on the treated side was 5·1 ml., increasing to 13·0 ml. during the subsequent infusion of *iso*propylnoradrenaline. Blood flow in the contralateral (control) forearm was between 2·4 and 3·3 ml. throughout. The increase in blood flow in the test forearm above the expected flow during *iso*propylnoradrenaline infusion was 260 % before and 194 % after giving phenoxybenzamine, a difference which is not significant ($t = 1{\cdot}07$, $P > 0{\cdot}1$).

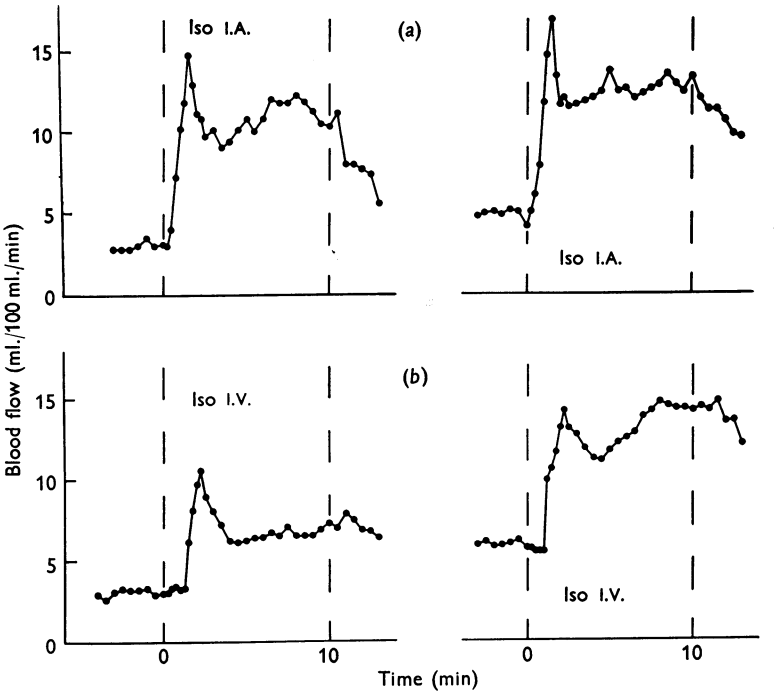

Fig. 3. Forearm blood flow during *iso*propylnoradrenaline infusions before and after phenoxybenzamine. (*a*) averaged results from five subjects given intra-arterial *iso*propylnoradrenaline (0·1 μg/min). (*b*) averaged results from six subjects given intravenous *iso*propylnoradrenaline (4 μg/min). Between the two parts of the experiments the subjects received intra-arterial phenoxybenzamine (3 mg during 6 min).

Intravenous isopropylnoradrenaline. This was studied in six subjects (Fig. 3(*b*)). Average resting blood flow in both forearms before Dibenyline was 2·7 ml./100 ml./min, increasing during intravenous infusion of *iso*propylnoradrenaline 4 μg/min to 4·9 and 5·7 ml. on the control and test sides respectively. After intra-arterial phenoxybenzamine the resting blood flow was 3·6 ml. in the control forearm, but increased to 5·5 ml. on

the test side. When the infusion of *iso*propylnoradrenaline was subsequently repeated, blood flow in the control forearm increased to 5·5 ml., that in the test forearm to 12·4 ml. During *iso*propylnoradrenaline infusion before phenoxybenzamine there was no significant difference between the increase in blood flow in the two forearms. After phenoxybenzamine the average increase in forearm blood flow on the treated side during *iso*propylnoradrenaline infusion was 175 %, compared with a 66 % increase on the control side, a difference which is significant ($t = 2{\cdot}74$, $P < 0{\cdot}05$). In the control limb there was no significant difference between the increase in blood flow during *iso*propylnoradrenaline infusion before and after phenoxybenzamine.

DISCUSSION

Intra-arterial infusion of phenoxybenzamine to a total dosage of 3 mg increased blood flow in the test forearm by an average of 90 %, while having no demonstrable effect on the blood flow in the opposite forearm. This vasodilatation is of the same order as that produced by blocking the deep nerves to the forearm (Barcroft, Bonnar, Edholm & Effron, 1943). The increase in forearm blood flow could therefore have been due to the blocking action of phenoxybenzamine on sympathetic excitatory activity. Other mechanisms may be involved, however, for Duff (1956) has shown phenoxybenzamine to have a direct vasodilator action, for it increased blood flow in recently sympathectomized limbs. An initial vasoconstrictor response to intra-arterial phenoxybenzamine did not occur in the present experiments, although this has been described in animals (Green *et al.* 1954).

While in animals very large doses of phenoxybenzamine have been shown to block vasodilator as well as vasoconstrictor responses (Nickerson *et al.* 1951; Green *et al.* 1954), in man in the dosage used here vasoconstriction alone is blocked. This allows us to separate vasoconstrictor and vasodilator components of the response to infused amines. Thus the response of the vessels of the forearm to infused adrenaline can be shown to have both dilator and constrictor components, since the vasoconstriction caused by intra-arterial infusion of adrenaline may be completely reversed. This is in agreement with the results of de la Lande & Whelan (1959).

The response in the forearm to intra-arterial noradrenaline is, however, purely vasoconstrictor, since phenoxybenzamine reduced this effect without reversing it. Nor was there a reversal of action with intravenous noradrenaline, for the slight increase in average blood flow during noradrenaline infusion after phenoxybenzamine (Fig. 2(*b*)) was not significantly different from the mean change in flow during infusion of noradrenaline before phenoxybenzamine.

*Iso*propylnoradrenaline has been shown to cause a marked dilatation in skeletal muscle in man, whether given intravenously or intra-arterially (Barcroft & Konzett, 1949; Cobbold, Ginsburg & Paton, 1960). With intra-arterial *iso*propylnoradrenaline the vasodilatation in the forearm was much the same before and after phenozybenzamine, indicating that there was no blockade of any direct vasoconstrictor component. There was, however, an increased vasodilator response with intravenous *iso*propylnoradrenaline after phenoxybenzamine, indicating the inhibition of some constrictor effect on forearm vessels. Whether this is similar to the reflex vasoconstriction in muscles seen on intravenous *iso*propylnoradrenaline infusion in cats, and initiated by the fall in systemic blood pressure (Bowman, 1959), or whether some other mechanism is involved, is not known. While a reflex vasoconstriction initiated by a fall in blood pressure would explain the decrease in forearm flow during intravenous infusion, it is unlikely to account for the fall in flow from the initial peak level during intra-arterial infusion.

It has been suggested that the sustained phase of the vasodilatation during intravenous adrenaline infusion is due to the action on the muscle vessels of some humoral substance which might be released by adrenaline (Barcroft & Swan, 1953). One substance which has been suggested is lactic acid, since an increase in blood lactate occurs following adrenaline administration (Cori & Buckwald, 1930). Evidence suggesting that lactic acid is responsible for adrenaline vasodilatation in the cat has been presented by Lundholm (1956). In man the venous lactate rises during intravenous infusion of adrenaline (Bell & Stead, 1952; Barcroft & Cobbold, 1956), and also during intra-arterial infusion (Allwood & Cobbold, 1961). There is no conclusive evidence, however, that it is lactic acid which is, in fact, responsible for the sustained vasodilatation in man. The present results indicate further that it may not be necessary to postulate formation or release of another dilator substance to explain this sustained increase in flow, for a dilator response in muscle to both intra-arterial and intravenous infusion of adrenaline is revealed when constrictor effects have been abolished by phenoxybenzamine.

An 'after-dilatation' was a prominent feature soon after the cessation of the control infusion of intra-arterial adrenaline. In the presence of phenoxybenzamine the 'after-dilatation' following intra-arterial adrenaline did not occur. This was also noted by de la Lande & Whelan (1959). It may, as suggested by these authors, be accounted for by a more rapid wearing off of the constrictor component of adrenaline action before the dilator effect has ceased. Although in the dosage used here intra-arterial noradrenaline and intra-arterial adrenaline both caused a comparable decrease in forearm blood flow, there was no after-dilatation following noradrenaline

infusion. This suggests that the phenomenon is not simply a 'rebound' on cessation of vasoconstriction, nor is it a reactive-hyperaemic response to a period of relative ischaemia. Moreover, there was no after-dilatation in the hand following intra-brachial infusion of the closely similar dose of 1·5 μg adrenaline/min (Swan, 1951). Although we do not yet know the cause of the after-dilatation this evidence suggests that it is specifically related to the action of adrenaline on skeletal muscle.

SUMMARY

1. The effect of intra-arterial phenoxybenzamine on the change in forearm blood flow during the infusion of sympathomimetic amines has been determined in healthy students by venous occlusion plethysmography.

2. Phenoxybenzamine reversed the normal constrictor response in the forearm to intra-arterial infusion of adrenaline (2 μg/min). The dilator response to the intravenous infusion of adrenaline (10 μg/min) was also significantly increased.

3. Phenoxybenzamine significantly reduced the vasoconstriction produced by intra-arterial infusion of noradrenaline (2 μg/min), but did not uncover a vasodilatation; the constrictor response to intravenous noradrenaline (10 μg/min) was abolished.

4. Vasodilatation during the intra-arterial infusion of *iso*propylnoradrenaline (0·1 μg/min) was not significantly altered; with intravenous *iso*propylnoradrenaline (4 μg/min) the vasodilator response was increased.

5. The results suggest that it is not necessary to postulate release of another vasodilator to explain the sustained vasodilatation in the forearm during intravenous infusion of adrenaline.

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REFERENCES

ALLWOOD, M. J. & COBBOLD, A. F. (1961). Lactic acid release by intra-arterial adrenaline infusions before and after Dibenyline, and its relationship to blood flow changes in the human forearm. *J. Physiol.* (In the Press.)

ALLWOOD, M. J. & GINSBURG, J. (1959). The effect of Dibenyline on the vascular response to sympathomimetic amines in the forearm. *J. Physiol.* **147**, 57*P*.

BARCROFT, H., BONNAR, K. McK., EDHOLM, O. G. & EFFRON, A. S. (1943). On sympathetic vasoconstrictor tone in human skeletal muscle. *J. Physiol.* **102**, 21–31.

BARCROFT, H. & COBBOLD, A. F. (1956). The action of adrenaline on muscle blood flow and blood lactate in man. *J. Physiol.* **132**, 372–378.

BARCROFT, H. & KONSETT, H. (1949). On the actions of noradrenaline, adrenaline and *iso*propylnoradrenaline on the arterial blood pressure, heart rate and muscle blood flow in man. *J. Physiol.* **110**, 194–204.

BARCROFT, H. & SWAN, H. J. C. (1953). *Sympathetic Control of Human Blood Vessels.* London: Edward Arnold and Co.

BELL, D. M. & STEAD, A. E. (1952). Effects of epinephrine on the vessels of the calf. Observations on the period of initial vasodilatation. *J. appl. Physiol.* **5**, 228–232.

BOWMAN, W. C. (1959). The effect of *iso*propylnoradrenaline on the blood flow through the individual skeletal muscle in the anaesthetized cat. *J. Pharm., Lond.*, **11**, 143–149.

COBBOLD, A. F., GINSBURG, J. & PATON, A. (1960). Circulatory, respiratory and metabolic responses to *iso*propylnoradrenaline in man. *J. Physiol.* **151**, 539–550.

CORI, C. F. & BUCKWALD, K. W. (1930). Effects of continuous intravenous injection of epinephrine on the carbohydrate metabolism, basal metabolism and vascular system of normal men. *Amer. J. Physiol.* **95**, 71–78.

DALE, H. H. (1906). On some physiological actions of ergot. *J. Physiol.* **34**, 163–206.

DALE, H. H. & RICHARDS, A. M. (1927). The depressor (vaso-dilator) action of adrenaline. *J. Physiol.* **63**, 201–210.

DE LA LANDE, I. S. & WHELAN, F. R. (1959). The effect of antagonists on the response of the forearm vessels to adrenaline. *J. Physiol.* **148**, 548–553.

DUFF, R. S. (1952). Effect of sympathectomy on the response to adrenaline of the blood vessels of the skin in man. *J. Physiol.* **117**, 415–430.

DUFF, R. S. (1956). Action of Dibenyline on the peripheral circulation. *Brit. med. J.* ii, 857–860.

DUFF, R. S. & GINSBURG, J. (1957). Some peripheral vascular effects of intra-arterial Dibenyline in man. *Clin. Sci.* **16**, 187–196.

ELLIOTT, T. R. (1905). The action of adrenaline. *J. Physiol.* **32**, 401–407.

GADDUM, J. H., PEART, W. S. & VOGT, M. (1949). The estimation of adrenaline and allied substances in blood. *J. Physiol.* **108**, 467–481.

GINSBURG, J. & DUFF, R. S. (1956). Effect of chlorpromazine on adrenaline vasoconstriction in man. *Brit. J. Pharmacol.* **11**, 180–185.

GINSBURG, J. & DUFF, R. S. (1958). Inhibition of levarterenol by intra-arterial phenoxybenzamine in man. *Circulation Res.* **6**, 751–754.

GREEN, H. D., DENISON, A. B. Jr., WILLIAMS, W. O. Jr., GARVEY, A. H. & TABOR, C. G. (1954). Comparison of the potency of dibenzyline, ilidar, phentolamine (regitine) and tolazoline (priscoline) in blocking the vasoconstrictor responses in canine muscle to lumbar sympathetic stimulation and to intra-arterial injections of L-epinephrine and of L-norepinephrine. *J. Pharmacol.* **112**, 462.

HUIDOBRO, F. (1954). Pharmacological properties of chloro-3(dimethylamine-3propyl)10-phenothiazine or 4·560 RP. *Arch. int. Pharmacodyn.* **98**, 308.

LUNDHOLM, L. (1956). The mechanism of the vasodilator effect of adrenaline. I. The effect on skeletal muscle. *Acta physiol. scand.* **19**, Suppl. 133.

NICKERSON, M., HENRY, J. W. & NOMAGUCHI, G. M. (1951). Blockade of responses to epinephrine and norepinephrine by dibenamine congeners. *Fed. Proc.* **10**, 327.

SWAN, H. J. C. (1951). Observations on a central dilator action of adrenaline in man. *J. Physiol.* **112**, 426–437.