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THE EFFECT OF ADRENALINE AND NORADRENALINE ON THE LIVER BLOOD FLOW

By J. GRAYSON and D. H. JOHNSON
From the Departments of Physiology and Pathology, University of Bristol

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The control of the circulation in the splanchnic area and liver is a matter of considerable importance to the vascular economy of the body. Its investigation has been greatly hindered in the past by technical difficulties. There is a considerable amount of information available concerning the behaviour of the isolated perfused liver (Bauer, Dale, Poulsson & Richards, 1932). Valuable experiments have also been performed using blood-flow recorders in the hepatic artery and portal vein (Burton-Opitz, 1912). Recently, too, the development of the bromsulphalein technique by Bradley and his co-workers (Bradley, Inglefinger, Bradley & Curry, 1945) has enabled observations to be made on liver blood flow in man. There is, however, a disappointing lack of cohesion in the sum total of available knowledge and much of the published work is conflicting.

The technique of internal calorimetry described recently (Grayson, 1952*a*) offers a fresh approach to these problems. A development of Gibbs's heated thermocouple (Gibbs, 1933), it has many advantages in so far as it enables quantitative observations to be made without any interference with the blood supply of the liver. It is eminently suited to recovery experiments, and the presence of the recorders causes no apparent inconvenience to the animal. Early observations on liver blood flow (Birnie & Grayson, 1952) have already justified the use of the method. Nevertheless the first object of the present paper must be to assess more fully its scope and limitations.

In this investigation of the liver circulation we have been concerned with two factors only, namely the effect of circulating adrenaline and the effect of changing blood pressure. Previous work has demonstrated clearly that systemically administered adrenaline produces a rise in liver blood flow (Burton-Opitz, 1912; Bearn, Billing & Sherlock, 1951), whereas the action of adrenaline locally on liver vessels is vasoconstrictor (Burton-Opitz, 1912; Bauer *et al.* 1932). It has been suggested that the initial increase in flow with

systemic administration is a mechanical effect of the raised blood pressure. We shall produce evidence to demonstrate that this cannot be the full explanation and that nervous mechanisms are also involved.

METHODS

Internal calorimetry

The technique of internal calorimetry has been fully described elsewhere (Grayson, 1952*a*). It consists in the measurement of the electric current required to maintain a plus 1° C thermal equilibrium in a heater embedded in the tissue. The temperature of the heater is measured thermoelectrically. The heat output of the wire is given by the equation $H = CI^2R$ (where C is a constant, I = the current in amperes and R = the resistance of the wire in ohms). It has been shown that heat is lost from the wire partly to the circulating blood and partly by direct conduction to the tissues. Provided a sufficient length of lead is embedded in the liver, errors do not arise from loss of heat conducted back along the wire.

Equilibrium being established, the formula $I^2R = k4\pi r\theta$ may be applied (where k is the thermal conductivity of the tissue, r is the radius of a sphere of equivalent thermal properties to the heater, and θ is the temperature increment). For the purpose of these experiments the I^2 value is defined as the square of the current required to maintain equilibrium when $\theta = 1$. In a situation uncomplicated by blood flow, as for example with the recorder embedded in gelatine, the I^2 value is a linear function of thermal conductivity. The fraction I^2/k may, therefore, be used as a standardization factor (F) for any particular recorder. It follows, too, that thermal conductivity, k, is given by the fraction I^2/F.

The determination of the standardization factor, F, has been fully described elsewhere (Grayson, 1952*a*). The recorders used in the present work were made to standard dimensions using the same 'jig' throughout. The factor was determined and found to be 110, lower than the factor determined for the recorders used by Birnie & Grayson (1952), which were made on a different 'jig'. Using these recorders, therefore, thermal conductivity, k, was obtained by recording the I^2 value and dividing by 110.

The thermal conductivity of dead liver has been determined using these methods (Grayson, 1952*a*) and found to be constant within narrow limits. Thus, it has been found that the I^2 value determined after death in the rat or rabbit liver is 0·130–0·135. The figure 0·130 is nearest to the true mean and has been used, for convenience, in determinations on the living animal.

The presence of circulating blood produces changes in the apparent thermal conductivity of animal tissue. Thus, in the present work, the mean I^2 value recorded in rat liver in the conscious animal under resting conditions was 0·290. If this were expressed in terms of thermal conductivity it would mean that the apparent thermal conductivity was $0{\cdot}290/110 = 26{\cdot}4 \times 10^{-4}$. But the true thermal conductivity has been shown to be $0{\cdot}130/110 = 11{\cdot}9 \times 10^{-4}$. There is, therefore, an apparent increase in thermal conductivity due to the presence of circulating blood, of $14{\cdot}5 \times 10^{-4}$. This figure has been termed the conductivity increment (δk) which has been shown to be a linear function of blood flow. It may, in practice, be more simply derived using the subtracted I^2 value, that is, the actual I^2 value recorded less the I^2 value given by dead liver (i.e. 0·130) and dividing this figure by the factor 110. Empirical observations on perfused tissue suggest, however, that a correlation factor exists whereby conductivity increment may be related directly to volume flow of blood. This factor is 12·5. Thus, flow in ml./ml./sec $= \delta k \times 12{\cdot}5$. In practice it gives figures for liver blood flow which are of the same order as those published by other workers. However, it should be remembered that the factor is empirical and may have to be modified in the light of future experience. Meanwhile, it is established that conductivity increment itself is a quantitative function of blood flow in the vicinity of the recorder.

It must be emphasized that measurements using this instrument are purely local. Internal calorimetry gives no measure of total liver blood flow. Evidence will be presented in this paper to demonstrate how greatly the proximity of large vessels may modify the figures obtained. For

this reason, where results have been converted to blood flow, they have been expressed in terms of blood flow per ml. of tissue per min. In the previous paper by Birnie & Grayson the conversion was expressed as blood flow per 100 ml. of tissue per min. This is now regarded as misleading, suggesting as it does a wider sphere of influence than the recorder in fact possesses.

The implantation of recorders

Similar methods were used for the rabbit and the rat. Using ether or pentobarbitone sodium anaesthesia, a ventral, mid-line incision was made exposing the upper part of the abdominal cavity. A broad-gauge hypodermic needle was introduced under one or other skin flap and passed subcutaneously around the body until it emerged through the dorsal skin as near the mid-line as possible. The recorder was threaded down the needle which was then withdrawn leaving the recording end and 1–2 in. of wire free in the abdominal wound. Thus leads for connexion to the recording apparatus emerged from the dorsum of the animal well away from the incision. A small loop was made in the wire 1–1½ cm proximal to the recorder.

A suitable lobe of liver was selected and brought out through the incision. An atraumatic needle, with fine catgut suture attached, was inserted through the capsule and into liver substance, then out again on the side of entry. The recorder was introduced into the liver substance, taking care to ensure that the recording end should be as near the centre of the lobe as possible. The needle was passed through the loop previously made in the wire and the catgut tied, thus anchoring the recorder firmly to the liver.

The practice of anchoring the recorder directly to the liver has been found preferable to the earlier methods used (Birnie & Grayson, 1952) where the only anchor was to the abdominal wall. It has not been found to interfere in any way with the results, the liver damaged by the suture being 1–1½ cm from the recording filament. Haemorrhage was usually slight, and subsequent scarring was limited to the region of the suture.

After suture of the abdominal incision, recovery was permitted. In most cases this was uneventful, the presence of the recorder and the leads on the dorsum appearing to cause little trouble or inconvenience to the animal.

In many cases the method was applied in the acute experiment. The technique of implantation was the same in every respect and the abdomen was closed before proceeding further.

The I^2/θ test

For the method of internal calorimetry to be valid the recorder must be surrounded by a quantity of tissue sufficient, as it were, to absorb all the heat generated in the filament. It has been shown that in a large mass of any given substance, the relation between I^2 and θ is linear over a big range of θ. In the rat liver, however, where the total volume available is relatively small, as θ is increased the zone of influence of the instrument may reach beyond the boundaries of the liver and heat transference take place across the interface into the surrounding medium. In the rat this has a low thermal conductivity and an inflexion appears on the curve relating I^2 and θ, the gradient of which falls off rapidly.

This forms the basis of a useful test which should be applied at the beginning of each experiment. I^2 values are taken with $\theta = 0{\cdot}5$, 1·0, 1·5, 2·0 and 3·0° C. Provided that about 1·5 cm of wire are enclosed within the tissue, linear relation over this range may be taken as evidence that the mathematical requirements are being fulfilled and that the recorder is adequately embedded.

Mechanical control of blood pressure

In some experiments it was required to alter the blood pressure of the animal without the use of drugs; in other cases it was desirable to prevent alteration in blood pressure even during the infusion of, for example, adrenaline. A mechanical blood-pressure compensating device was used. As large a cannula as possible (siliconed polythene tubing was found suitable in later experiments) was introduced into a suitable artery, usually the femoral. The tube was connected to a flask containing saline, or in some cases heparinized blood from another rabbit, communicating with

a reservoir in which pressure could be adjusted as desired by means of a foot pump and manometer. For blood-pressure compensation the pressure in the flask was adjusted to the initial level in the animal as recorded by a mercury manometer in the carotid artery. During the infusion of adrenaline when the animal's blood pressure might be expected to rise, blood passed from the animal into the reservoir. On ceasing the infusion, blood from the reservoir returned through the femoral artery into the rabbit. By this means general systemic blood-pressure changes could be eliminated.

When it was required to produce mechanical increments in blood pressure without using adrenaline or other drugs, the pressure in the compensating bottle was adjusted to a predetermined level higher than that of the animal; on opening the tap leading to the cannula, blood flowed from the reservoir into the animal. The systemic blood pressure rose, and it was thus possible to produce changes in pressure similar to those produced by adrenaline. It was equally practicable to lower the animal's blood pressure. With the reservoir at a pressure below that of the rabbit, opening the tap connecting cannula and reservoir permitted the animal to bleed against the lower pressure until equilibrium was established at the lower level.

RESULTS

Blood-flow variations in rat and rabbit liver under conditions of rest

In a previous paper, Birnie & Grayson (1952) reported observations on the liver blood flow of rats using the technique of internal calorimetry. The levels of flow reported from individual recorders were remarkably constant and conductivity increments ranged from 10·00 to $12{\cdot}36 \times 10^{-4}$. Day-to-day variations in individual rats were usually small; measurements made at the same time on different days showed only relatively small fluctuations.

Table 1 records resting blood flows observed during the present investigation. The position of each implant was checked post-mortem and in every case the recorder was found to be well embedded. In most instances the validity of the implant was further checked by means of the I^2/θ test above. It may be applied in the living animal but is subject to possible errors from changes in blood flow. In practice, with an animal at rest these may usually be neglected. But if doubt arises the test may more reliably be applied after death.

In those cases in which the test was applied the I^2/θ relationships were linear. The readings obtained in these animals must, therefore, be regarded as indicating true conductivity increments. They are consequently linearly related to blood flow.

The wide variation observed between different animals was not regarded as necessarily indicating wide variations in total liver flow. It must again be emphasized that the readings applied to the small quantities of tissue in the neighbourhood of the recorder only. The blood-flow measurements were local in nature. Thus, if the recorder were to lodge near a large vessel the reading would be considerably higher than readings from regions where there were no large vessels. In Table 1 the readings obtained from rats nos. 2 and 22 were considerably above average. The animals were subsequently killed and the livers examined. The recorders were centrally embedded in large liver lobes. No abnormalities were observed. On section, however, in each case, large

vessels were seen coursing near the recording filaments. In rat no. 2, a particularly large vessel was found in close proximity to the recorder.

In all cases where observations were repeated under resting conditions over a number of days the fluctuations in flow were slight compared with the variations shown in Table 1. Measurements from individual recorders were, indeed, remarkably constant. It seems probable, that the position of the

TABLE 1. Liver blood flow in different rats. First day of implantation

Rat no.	I^2 ($\theta=1$)	δI^2	δk ($\times 10^{-4}$)	Blood flow (ml./ml. of tissue/min)
1	0·300	0·170	15·5	1·16
2	0·511	0·380	34·5	2·59
4	0·320	0·190	17·3	1·29
5	0·280	0·150	13·6	1·02
8	0·190	0·050	4·5	0·34
9	0·314	0·184	16·7	1·25
10	0·312	0·182	16·6	1·24
11	0·225	0·095	8·6	0·64
12	0·278	0·148	13·5	1·01
13	0·240	0·110	10·0	0·75
14	0·300	0·170	15·5	1·16
15	0·250	0·120	10·9	0·82
16	0·220	0·090	8·2	0·61
17	0·352	0·222	20·2	1·51
19	0·280	0·150	13·6	1·02
20	0·350	0·220	20·0	1·50
21	0·300	0·170	15·5	1·16
22	0·370	0·240	21·8	1·63
23	0·310	0·180	16·4	1·23
24	0·295	0·165	15·0	1·12
25	0·226	0·096	8·7	0·65
26	0·300	0·170	15·5	1·16
27	0·280	0·150	13·6	1·02
28	0·250	0·120	10·9	0·82
29	0·260	0·130	11·8	0·88
31	0·340	0·210	19·1	1·43
Av.			14·9	1·13

recorder in relation to larger blood vessels was an important factor in determining the level of the reading which was clearly not representative of total liver flow.

Day-to-day variations in liver blood flow. The findings in this respect were similar to those previously reported by Birnie & Grayson. Little fluctuation was observed in flow measured under similar conditions in resting rats. Several livers were sectioned after death. In most cases histological examination up to 10 days after implantation demonstrated fibrous changes not more than 0·23 mm around the recorder. In one rat, however, a progressive decline in conductivity increment from 15·9 initially to $10{\cdot}9 \times 10^{-4}$ was recorded, and histological examination 10 days after implantation showed extensive fibrosis around the recorder. It was considered that the low conductivity increment at the end of this period was due to the low vascularity of this fibrous tissue.

In another animal a progressive rise occurred in conductivity increment from 13·2 initially to $39{\cdot}6 \times 10^{-4}$ on the 11th day. It remained steady at about

this level until the 17th day when the animal died. Post-mortem examination showed the presence of multiple abscesses lining the peritoneum and a severe infestation of the liver by an unclassified taenia.

The importance of these findings in relation to the measurement of liver blood flow will be discussed later.

Blood-flow fluctuations in the rabbit. Similar observations were made on conscious rabbits. The mean conductivity increment was $16{\cdot}3 \times 10^{-4}$. There was no large variation between different animals nor between readings taken on the same animal at different times under similar conditions. Using the correction factor, the mean blood flow in the neighbourhood of the recorders was 1·22 ml./ml./min.

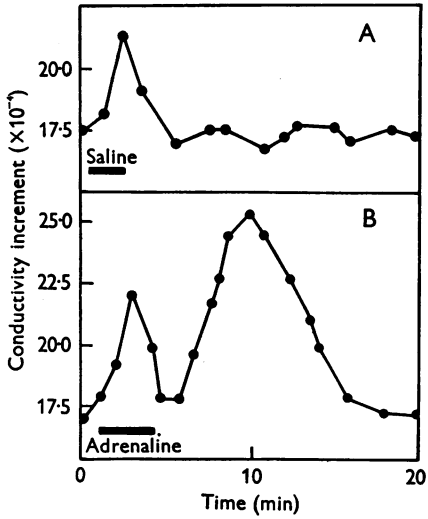

Fig. 1. The effect of subcutaneous adrenaline on liver blood flow in the conscious rat. A, control injection, sterile saline; B, adrenaline tartrate, 15 μg.

The action of adrenaline and noradrenaline on liver blood flow in the rat

The effect of adrenaline in the conscious rat. Most of the experiments were carried out with the rat in a restraining tube made from perforated zinc. Fig. 1 shows a typical result. During the resting period no significant fluctuations were observed. A control injection of sterile saline (Fig. 1A), subcutaneously into the abdominal wall, was followed by an immediate rise in conductivity increment which subsided rapidly, resting levels being reached within 2 min. A similar injection of 15 μg adrenaline tartrate in sterile saline (Fig. 1B), was also followed by an immediate rise in flow with a rapid return to resting levels. This was followed by a slow, more sustained increase in blood flow which reached a maximum in 10 min and then slowly subsided to resting levels.

The delayed increase in blood flow took place in all experiments where adrenaline was injected into the conscious rat (Table 2A). It never occurred following saline alone. It must, therefore, be regarded as a true adrenaline effect, an increase in blood flow.

The immediate increase, however, was variable. In some animals it was not observed (Fig. 2); in others, as in Fig. 1, it was marked. Usually it was only slight. It is possible that when present it was a physiological effect of the fear or slight pain accompanying the injection.

TABLE 2. The effect of subcutaneous adrenaline on liver flow in (A) conscious and (B) anaesthetized rats (doses = 15–20 μg)

A. Conscious rat			B. Anaesthetized rat		
Resting δk ($\times 10^{-4}$)	Max. effect δk ($\times 10^{-4}$)	% flow increase	Resting δk ($\times 10^{-4}$)	Max. effect δk ($\times 10^{-4}$)	% flow increase
17·2	20·9	20·8	12·2	13·6	11·4
28·2	30·9	9·5	13·6	14·5	6·6
25·5	32·7	28·3	24·5	22·7	− 7·4
21·8	25·4	16·5	26·4	17·3	−34·4
19·1	22·7	18·8	22·3	22·7	1·7
19·1	24·5	28·3	16·3	15·5	− 4·9
14·5	19·1	24·1	12·4	11·4	− 8·0
14·1	20·0	41·8	12·7	12·7	00·0
14·5	19·1	24·1	12·7	10·9	−14·1
14·1	20·0	29·5	18·2	21·8	16·5
18·2	21·4	17·6	13·6	15·5	12·2
14·1	18·2	29·0	15·5	15·5	0·0
10·9	15·5	42·2	10·9	10·9	0·01
—	—	—	10·0	10·9	9·0
—	—	—	12·7	12·7	0·0
—	—	—	13·6	13·6	0·0
—	—	—	18·6	19·1	2·6
—	—	—	15·0	14·5	3·3
—	—	—	20·9	20·0	− 4·3
—	—	—	18·6	19·1	2·6
—	—	—	13·6	9·1	−33·1
—	—	—	15·5	19·1	23·2
Mean % increase 25·4			Mean % increase		−1·1
			S.E.		±3·5

The effect of adrenaline on the anaesthetized rat. Table 2B shows the results of twenty-two experiments in which adrenaline tartrate was administered subcutaneously to rats anaesthetized with open ether. In six cases a significant rise in flow was observed, in seven a significant fall; in the rest no significant change occurred. The mean fall was thus not significantly different from zero.

Intravenous administration to the anaesthetized rat, however, produced a different picture. Fig. 3 shows the results of an experiment in which adrenaline was administered first subcutaneously, secondly intravenously. It is seen that on intravenous administration a distinct rise in blood flow occurred. Blood-pressure records from this case showed no rise in blood pressure during or following the subcutaneous injection but a big rise with

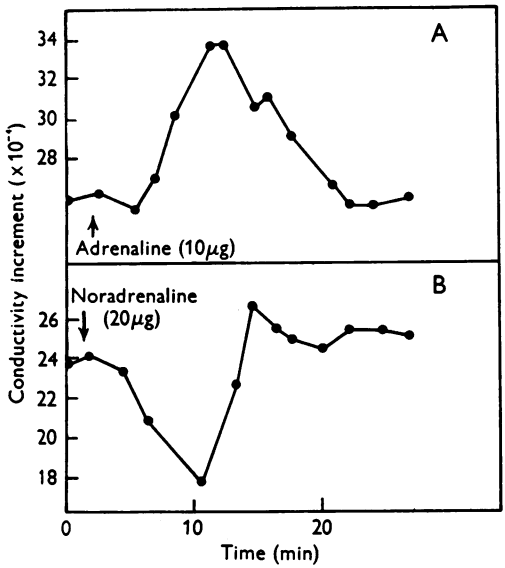

Fig. 2. The effect of subcutaneous adrenaline and noradrenaline on liver blood flow in the conscious rat. A, adrenaline tartrate, 10 μg; B, noradrenaline, 20 μg.

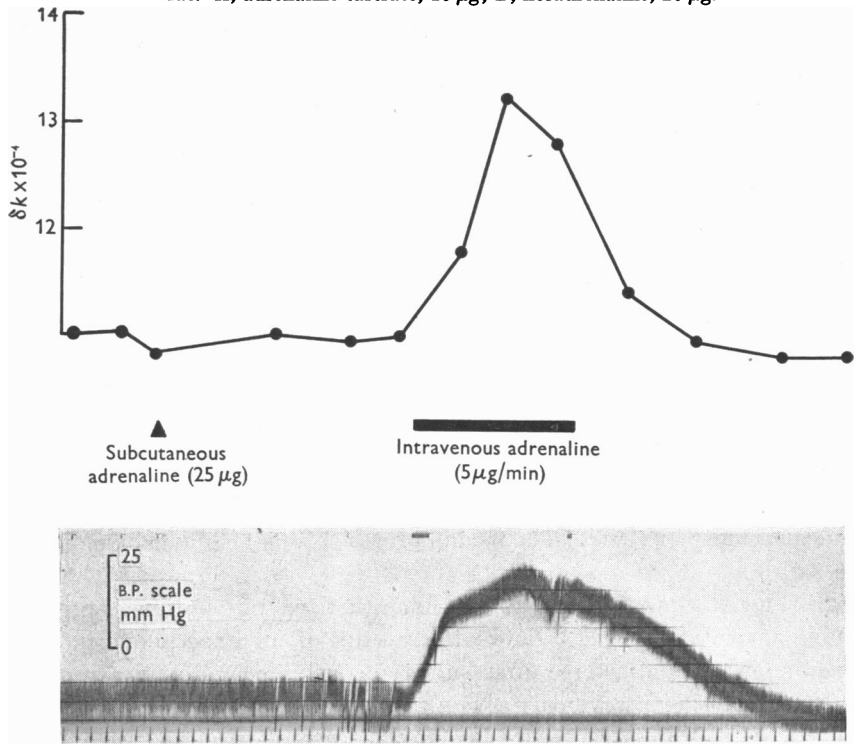

Fig. 3. The effect of subcutaneous and intravenous adrenaline on the anaesthetized rat. Liver blood flow and blood pressure recorded from the femoral artery. Time marker, 30 sec intervals; B.P. base-line, 100 mm Hg.

the intravenous infusion. The blood-flow rise accompanied the rise in blood pressure, but was not sustained, dropping towards resting levels whilst the infusion and the blood-pressure rise were both maintained.

In most of the experiments on the conscious or anaesthetized rat blood-pressure records were not available. In a different series of experiments, however, it was found that the administration of adrenaline in doses of up to 20 μg, subcutaneously, intraperitoneally, or intramuscularly, only occasionally produced a rise in arterial blood pressure.

TABLE 3. The effect of subcutaneous noradrenaline on liver blood flow in the rat (dose 20 μg)

Resting δk ($\times 10^{-4}$)	Max. effect δk ($\times 10^{-4}$)	% flow increase	Anaesthesia
23·7	18·2	−23·2	Ether
19·1	19·0	− 0·5	Ether
19·0	24·5	+28·9	None
19·1	24·3	+27·3	None
15·0	11·4	−24·0	None
14·1	16·4	+16·2	None

The effect of subcutaneous noradrenaline on the rat. Noradrenaline had a less predictable effect when administered subcutaneously to the rat. The results of six experiments are given in Table 3. In three cases there was a significant increase in flow, in two a fall (Fig. 2) and in the other experiment no effect. It was not possible in this short series of experiments to relate the nature of the result to the state of anaesthesia.

The effect of adrenaline and noradrenaline on liver blood flow in the rabbit

Intravenous adrenaline on the conscious rabbit. The actions of adrenaline and noradrenaline were investigated on the conscious rabbit. In both cases the procedure was the same. The animal was restrained in a specially constructed box. The rabbit's head and neck protruded through a hole in one end; its ears were thus beyond the range of the animal's scratch reflexes. A needle was inserted into an ear vein, connected to an electrically driven constant speed infusion apparatus (Grayson & Swan, 1950), and saline was administered for a period of about 10 min at the rate of 0·5 ml./min. Blood flows were recorded throughout. The infusion was then changed to a solution of saline containing adrenaline or noradrenaline and observations continued. Finally, the infusion was changed to simple saline.

Adrenaline administered in doses of 5 μg/min for periods of up to 5 min produced an initial rise in hepatic blood flow (Fig. 4). This was not sustained and, during the period of infusion, the flow returned towards resting levels. Similar results were obtained in all cases, there being an initial rise in blood flow of up to 50% over resting levels followed by a return towards base-line

during the infusion. Occasionally, on stopping the infusion, there was a decrease in hepatic blood flow below the base-line.

Intravenous noradrenaline on the conscious rabbit. Noradrenaline in doses of 5–10 μg/min produced variable diminutions in liver blood flow which were, however, sustained throughout the period of infusion. Fig. 4 shows a typical result.

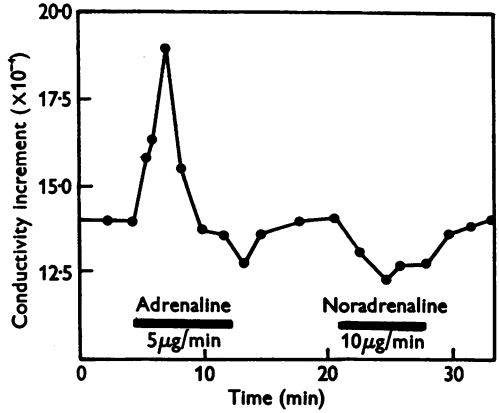

Fig. 4. The effect of intravenous adrenaline (5 μg in 0·5 ml. saline per min) and noradrenaline (10 μg in 0·5 ml. saline per min) on liver blood flow in the conscious rabbit.

Liver blood-flow investigations in the anaesthetized rabbit

For reasons of technical convenience the rabbit was chosen as the most suitable animal in which these phenomena might be further investigated. Rabbits were anaesthetized in three ways, using ether, pentobarbitone sodium (nembutal) intravenously, or urethane. The response obtained did not appear to be affected by the type of anaesthetic, and intravenous pentobarbitone sodium was preferred for the majority of experiments.

Some experiments were performed on animals in which the blood-flow recorders had been implanted some days previously and in which a normal conscious response to adrenaline had been demonstrated. In others the recorders were implanted at the time of the experiment.

After implantation (the technique of which has been described previously in this paper) the operative procedure was the same in all cases. A carotid artery, usually the left, was cannulated for blood-pressure recording by means of a mercury manometer writing on a kymograph. A femoral artery was also cannulated for connexion to the blood-pressure compensator already described. A femoral vein, usually of the opposite side, was also cannulated to facilitate intravenous infusions. In experiments where the depressor nerves were to be cut or stimulated these were isolated carefully at the beginning of the experiment.

The effect of intravenous adrenaline and noradrenaline in the anaesthetized rabbit. Fig. 5 shows the effect on blood pressure and hepatic blood flow of intravenous adrenaline. Liver blood flow and general blood pressure rose simultaneously at the beginning of the infusion. The blood-pressure elevation was maintained throughout, but the hepatic blood flow fell after 2½ min, returning to the base-line despite the elevated blood pressure. In some experiments there was a further variable drop below the base-line on stopping the adrenaline infusion.

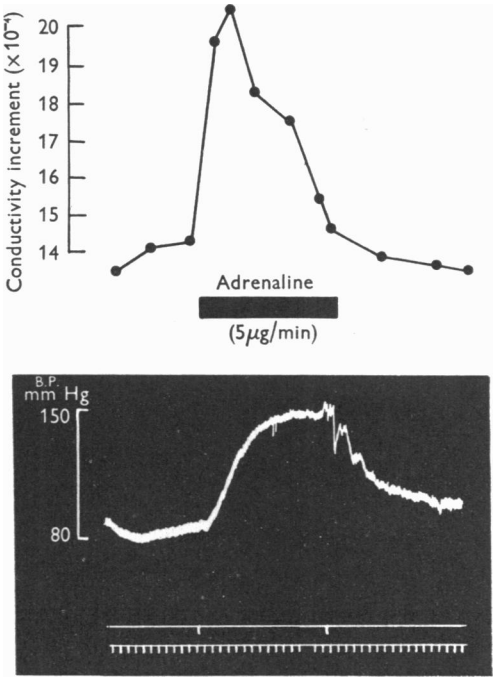

Fig. 5. The effect of intravenous adrenaline (5 μg/min) on blood pressure and liver blood flow in the anaesthetized rabbit. Time interval, 30 sec.

Noradrenaline infusions in equivalent pressor doses produced a fall in liver blood flow which accompanied the rise in blood pressure and was maintained throughout the period of infusion (Fig. 6). In some experiments there was a rise in flow above the basal level on stopping the noradrenaline infusion.

The fall in flow which took place with noradrenaline, in the presence of a rise in systemic blood pressure, can only be interpreted as evidence of active vasoconstriction in the liver or splanchnic bed.

Adrenaline and noradrenaline following blood-pressure compensation. In seven experiments the femoral artery was cannulated and connected to the

compensating device already described. Fig. 7 shows the effects of a slow intravenous infusion of adrenaline in such an experiment. In the first stage of the experiment the compensator was used to prevent the blood pressure from rising. Adrenaline was administered intravenously and blood rose in the compensating reservoir, demonstrating that the adrenaline dose was effective, but the liver blood flow fell. The reduced level of blood flow was maintained throughout the infusion, a return to resting levels occurred on stopping the infusion. In the second stage of the experiment the compensator

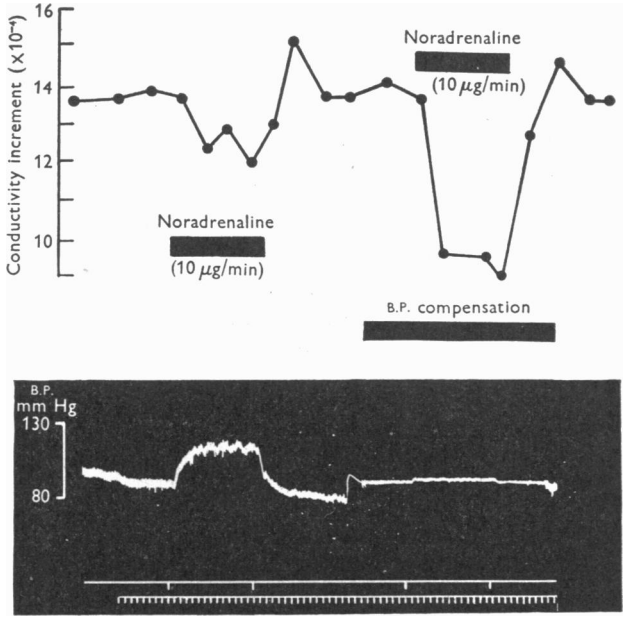

Fig. 6. The effect of noradrenaline (10 μg/min) on liver blood flow (rabbit) with and without compensation of the blood pressure. Time interval, 30 sec.

was disconnected from the animal. On giving adrenaline intravenously the blood pressure rose in response to the infusion and the blood flow rose initially—a typical adrenaline effect.

Fig. 6 shows the results of a similar experiment using noradrenaline. When the blood pressure was permitted to rise noradrenaline produced a small decrease in liver blood flow. When the blood pressure was stabilized the same dose of noradrenaline produced a very much greater fall in liver blood flow.

These experiments demonstrate the importance of changes in systemic blood pressure in relation to the action of adrenaline in the liver or splanchnic bed. They suggest that whereas the local action of adrenaline and noradrenaline in the liver or splanchnic bed is vasoconstrictor the effect of the increased

mean arterial blood pressure is to produce increase in liver blood flow. With adrenaline, the blood pressure changes, temporarily at least, prevailed and the net result of a slow infusion was to produce increase in liver flow. With noradrenaline the local effect frequently predominated and a diminution in blood flow resulted.

The effect of mechanical changes in blood pressure. The above experiments demonstrate the importance of general blood pressure in the control of liver blood flow. In order to test the importance of these factors, experiments were performed in which the blood pressure was varied, using the 'compensator'.

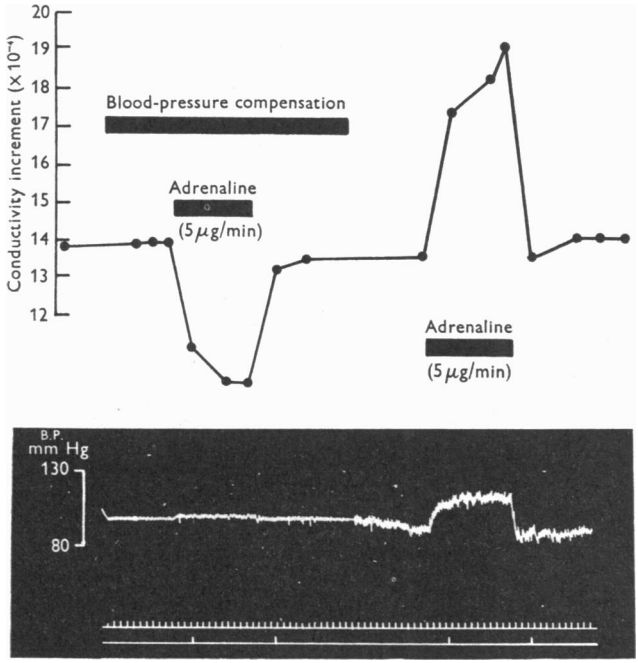

Fig. 7. The effect of intravenous adrenaline (5 μg/min) with and without compensation of the blood pressure. Time interval, 30 sec.

By adjusting the pressure within the compensator reservoir to a suitable level the animal was permitted to bleed into the reservoir, until an equilibrium was established at this level. Alternatively, by raising the pressure within the reservoir, saline was transfused into the animal and the blood pressure raised.

It was realized that these procedures were far from physiological. However, the general condition of the animals seemed little affected by relatively brief changes in blood pressure of limited magnitude, the subsequent return to resting levels of blood pressure being quite remarkable.

Fig. 8 shows a typical result where the pressor effect of adrenaline was

simulated. It will be seen that the blood flow rose initially with the rise in blood pressure, then fell whilst the pressure was maintained at a plateau. On the return of the blood pressure to normal, the blood flow fell again, rapidly at first, to below base-line, then returned to resting levels.

In all experiments of this kind similar results were obtained. The immediate effect of rising blood pressure was to produce a rise in liver flow, but the maximum rise was never maintained.

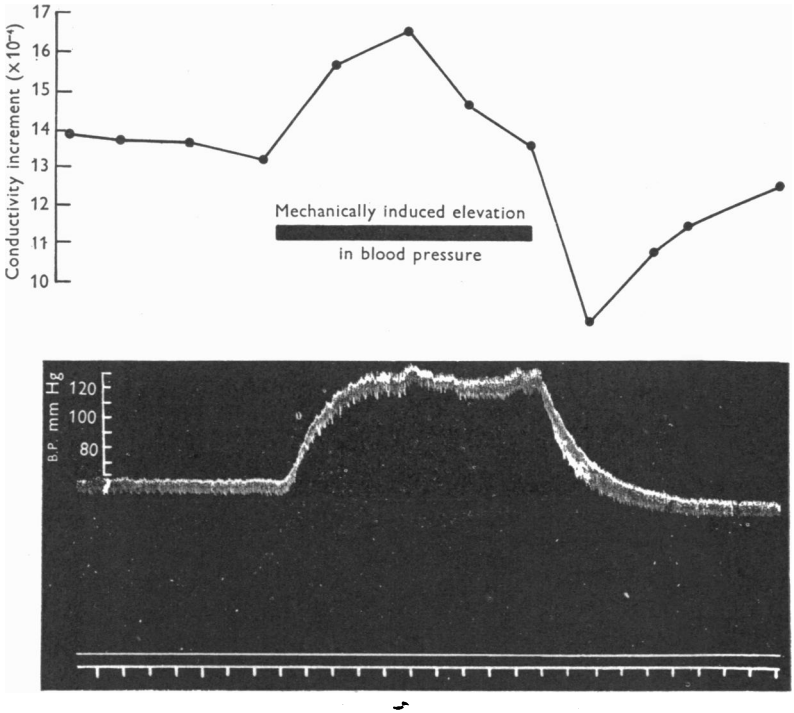

Fig. 8. The effect of a mechanically induced rise in blood pressure on liver blood flow. Time interval, 30 sec.

The effect of nerve section and nerve blocking on the adrenaline effect. The above experiments demonstrated clearly that the increase in hepatic blood flow produced by intravenous infusions of adrenaline only occurred when there was a rise in blood pressure. In fact the local action of both adrenaline and noradrenaline on blood vessels in the liver was vasoconstrictor. The above experiments did not show definitely whether the blood-flow increase following a rise in blood pressure was a passive effect or the result of a reflex action. The fact that the flow increase accompanying a sustained pressor effect was transient only (Fig. 5) suggested, however, that the effect was not simply passive. In order to study the possible important nerve pathways,

experiments were performed using ganglionic blocking agents and detailed nerve section.

The effect of tetraethylammonium bromide. The effect of this drug was investigated using the rabbit and the rat. A single dose of 25 mg followed immediately by a continuous intravenous infusion of the drug into the rabbit produced a fall in blood pressure but had very little effect on blood flow.

Fig. 9 shows the effect on liver blood flow in the rat of an adrenaline infusion before and after the administration of a single dose of tetraethylammonium bromide. It will be seen that after TEA the effect of adrenaline is markedly vasoconstrictor, there being a pronounced drop in liver blood flow with a dose that previously produced a rise.

Similar results were obtained in all such experiments.

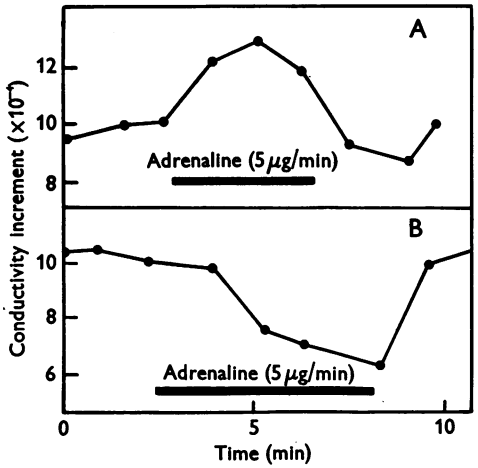

Fig. 9. The effect of intravenous adrenaline (5 μg/min) flow in the rat. Ether anaesthesia. A, before; B, after the administration of tetraethylammonium bromide (5 mg).

The influence of the carotid sinuses and depressor nerves. It was considered that the results with TEA supported the hypothesis that the increased blood flow brought about by pressor doses of adrenaline was reflex in nature. Accordingly, experiments were performed in which the depressor nerves were cut in the neck and the carotid sinuses isolated from the general circulation.

Fig. 10 shows a typical experiment. The left carotid artery was cannulated below the carotid sinus for the recording of blood pressure, the right depressor nerve was cut and the right carotid artery was clamped. Intravenous adrenaline still produced a rise in blood flow which was slow at first. On removing the clamp from the right carotid artery there was a continued rise in liver blood flow which then subsided in the usual way towards the base-line.

The left depressor nerve was now cut, and the adrenaline infusion repeated

with the right carotid artery again clamped. There was now a marked decline in liver blood flow. On removing the clamp from the carotid there was a slight return towards the base-line. Subsequently the blood flow progressively declined. Similar results were obtained in all experiments where the effect of depressor nerve section and carotid clamping on the adrenaline response was investigated. After bilateral nerve section and clamping of carotids intravenous adrenaline invariably produced a drop in hepatic blood flow.

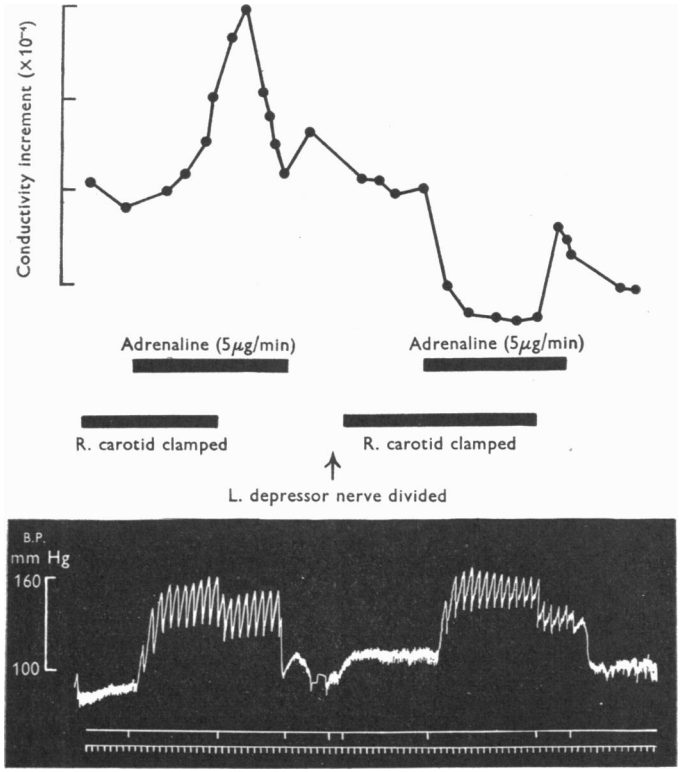

Fig. 10. The influence of the carotid sinuses and depressor nerves on reflex responses to adrenaline (5 μg/min intravenously) in the rabbit liver. Blood-pressure recording from the left carotid artery. Right depressor nerve cut before the record was taken. Time interval 30 sec.

It was found that with either carotid sinus or with one depressor nerve intact blood-pressure elevation usually produced some rise in liver blood flow.

It was concluded, therefore, that the main increase in liver blood flow caused by a rise in blood pressure was reflex in nature, the afferent impulses being conveyed in the depressor nerves or carotid sinus nerves. It was impossible in these experiments to assess the relative importance of these two pathways.

Stimulation of the depressor nerves. In order to demonstrate the relation of the depressor nerves to hepatic blood-flow experiments were performed in which the depressor nerves were stimulated electrically using an electronic square-wave stimulator. It was found that stimulation produced variable effects. It was considered that this may have been due to the conflicting action of the lowered blood pressure on the other depressor nerve and on the carotid sinus receptors.

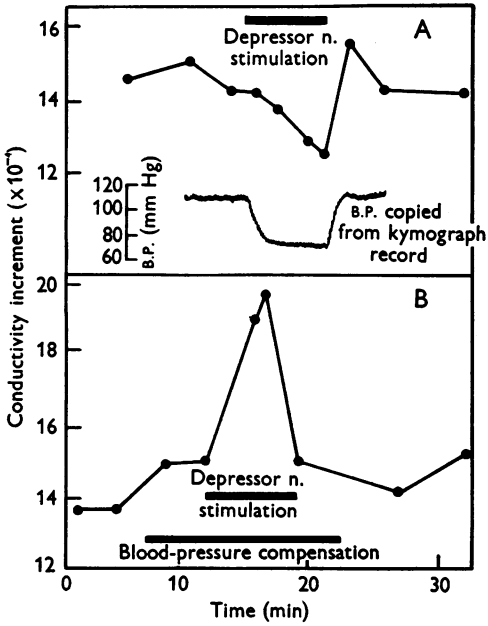

Fig. 11. The effect on liver blood flow in the rabbit of stimulating the right depressor nerve. A, before; B, after blood-pressure compensation. The blood-pressure tracing showed no change in B.

The experiments were repeated using the blood-pressure compensator to prevent alterations in systemic blood pressure. A typical result is shown in Fig. 11. Stimulation of the depressor nerve was accompanied by transfusion of blood from the reservoir into the animal demonstrating that the procedure was effective. The blood flow in the liver, however, rose. In four experiments the effect was consistent, and it must be concluded that stimulation of the depressor nerve produces vasodilatation in the liver.

DISCUSSION

The measurement of the liver blood flow

The technique of internal calorimetry does not measure blood flow in an organ directly. The figures obtained enable the apparent thermal conductivity to be determined and, hence, the conductivity increment, which is the apparent

thermal conductivity in the living liver less the thermal conductivity of the dead liver. Conductivity increment is itself a function of total fluid movement within the zone of influence of the recorder. In such a structure as the liver it may be assumed that the greater part of the total fluid movement is made up of blood flow. This does not mean, however, that the results obtained necessarily indicate blood flow through the whole liver; it can only be claimed that they indicate blood movements within a certain small volume of the tissue.

The scope and limitations of the method in investigations of the liver blood flow are clearer now than initially. One point which can scarcely be overemphasized is that the measurements are strictly local. There is no question of measuring total liver flow. The wide scatter in recorded conductivity increments ($4{\cdot}5$–$34{\cdot}5 \times 10^{-4}$, Table 1) and the demonstrated importance of the situation of the recorder with respect to large vessels emphasize this point. Even so, it is interesting that the mean of flows recorded for the rat under resting conditions in the present work is 1·13 ml./ml. of tissue/min. This may be compared with the figures for mean hepatic flow of 100 ml./100 g of tissue/min using the bromsulphalein technique in the human liver (Bradley *et al.* 1945), and the figures for the whole liver of 40–90 ml./100 g of tissue/min obtained by Grindlay, Herrick & Mann (1941) using thermostromuhrs in dogs. The present figure is of the same order. Indeed, this close correspondence has been considered in itself to be some vindication of the empirical correlation factor 12·5, which was arrived at on the basis of perfusion experiments (Grayson, 1952*a*).

In the present series a few high figures (Table 1) raise the mean above that obtained in the previous series (Birnie & Grayson, 1952). In other respects, however, this investigation bears out the findings reported earlier. Thus the day-to-day variation in flow from any given recorder was slight. There was no evidence to support the hypothesis of intermittent circulatory activity postulated by Grindlay *et al.* (1941), but it is possible that these fluctuations occur in volumes of tissue too small to be detected by our methods. Although internal calorimetry records only from a limited zone, the diameter of that zone may be of the order of 0·3 cm. On the other hand, there was a wide variation in flow from different recorders. It was not possible to correlate this variation with definite regions of the liver. Thus there was no real evidence that flows in the centre of the lobe were any greater than flows at the periphery; and the variations seen in Table 1 were probably the result of the presence or absence of larger vessels in the zone of influence of the recorder.

For an investigation of the present type such variations arising from the local nature of the method are of little importance. In some investigations such localized recordings might indeed be of definite advantage. Great care, however, would be required in the interpretation of results obtained by this method in long-term experiments. The possibility of progressive fibrosis would

have to be carefully examined when diminishing flows over a period of several days or weeks were recorded. Again the possibility of an inflammatory reaction would have to be considered if progressively increasing flows were observed. These difficulties are probably capable of solution. Meanwhile, they remain an obstacle to the investigation of such problems as liver circulatory reactions in hypertension and cirrhosis.

In the present investigation the method has been found very satisfactory. It caused little apparent disturbance, and it was possible to conduct experiments on conscious animals undisturbed by extensive surgical procedures.

In the conditions of an acute experiment it was equally suitable. There was no interference with the liver circulation, the operative trauma was minimal and the animals were in good condition at the beginning of the observations.

The effect of adrenaline and noradrenaline on the liver circulation

A comprehensive review of all the work published on this subject is beyond our present scope. Some of it is, however, outstanding. The experiments of Burton-Opitz (1912) are, perhaps, of especial significance in relation to the present work. Using mechanical stromuhrs in the hepatic artery of dogs, he was able to show that the direct action of adrenaline on the liver was to constrict the hepatic radicles. However, given systemically, small intravenous doses of adrenaline produced simultaneous rises in general blood pressure and of blood flow in the hepatic artery. The flow increase did not occur unless the pressure rose; he suggested, therefore, that this apparently anomalous effect was passive, the mechanical result of increased blood pressure overriding the local constrictor action of adrenaline. McLaughlin (1928) similarly found an initial flow increase in the liver of rabbits and concurred in attributing it to the raised blood pressure. Recently, the work of Bearn *et al.* (1951), using the bromsulphalein technique, has demonstrated that in the human liver an increased flow accompanies the intravenous infusion of adrenaline.

The suggestion that adrenaline might have additional effects in relaxing the outflow mechanism was first made by Mautner (1923), and further work by Dale (1929) and Bauer *et al.* (1932) demonstrated the importance of this phenomenon in the blood-flow responses to adrenaline of the isolated dog's liver. Small doses relaxed the sphincters controlling outflow. This was confirmed by other workers (Grab, Janssen & Rein, 1929*a*, *b*). With larger doses, however, the constrictor action on inflow became of overriding importance. In other species the effect on outflow appeared to be less important, and it may be significant that the dog is apparently unique in the amount of muscle to be found in its hepatic veins. Human hepatic veins are more akin to those of the cat (Popper, 1931) in the localization and extent of hepatic vascular musculature.

Our observations confirm much of the work of Burton-Opitz and others. In

experiments where blood-pressure change was eliminated by means of a mechanical compensator an adrenaline infusion produced a marked and sustained diminution in blood flow. Unpublished experiments of Ginsburg & Grayson, using the same methods, have shown that direct injection into the portal vein of rats produces initially a decrease in flow. This is in agreement with the findings of other workers. There can be little doubt, therefore, that the direct action of adrenaline on liver vessels is vasoconstrictor. In the present experiments without blood-pressure compensation in the intact animal, both conscious and anaesthetized, intravenous adrenaline produced an initial rise in liver flow. It was short-lived, however, for the flow returned to resting levels within a few minutes even when the adrenaline infusion was continued. However, conductivity increment, which was the basic measurement, does not distinguish between inflow and outflow. It might, therefore, have been argued that the initial rise in flow was due to the direct dilator action of adrenaline on hepatic sphincters, to the relaxation of the hepatic veins and a consequent increased outflow. It was not, however, obtained when the blood-pressure compensator was used. It could not, therefore, have been due to the direct action of adrenaline on hepatic sphincters or other vascular elements in the liver.

There can be little doubt from these experiments that an increase in blood pressure was necessary for the production of the initial increase in liver blood flow. In this respect our findings agree with those of Burton-Opitz (1912). But the flow increase was short-lived; though the blood pressure remained elevated the flow returned towards the base-line. The effect on flow cannot, therefore, have been purely passive. Moreover, the initial increase in flow was prevented by section of the depressor nerves and clamping of the carotid arteries. The integrity of nerve pathways is consequently as important as an elevation in blood pressure for the development of the response. The final level of flow may have been determined in part by mechanical factors, but the initial rise must be regarded as a nervous reflex response. Indeed, the nerve section experiments suggest strongly that the carotid sinus nerves and the depressor nerves, in the rabbit at least, constitute the afferent pathway. A similar reflex response to blood pressure has been demonstrated in the human bowel (Grayson, 1952*b*). In this region the effects of blood-pressure change were more long lasting and it was suggested that they were of importance in the nervous regulation of peripheral resistance.

No such hypothesis can be advanced in explanation of the present findings. The dilator response in the liver was too transitory to be strictly comparable to the bowel responses. Whether the quick return to resting levels was a manifestation of adaptation in the carotid sinuses and aortic arch receptors, whether it was significant of some local controlling mechanism in the liver or whether other factors were involved remains to be determined. The question

is complicated by the double blood supply of the liver, and until further information with respect to the relative behaviour of the portal and hepatic radicles is available, further speculation would be ill-advised.

The experiments do, however, explain in part the failure of subcutaneous injections of adrenaline to produce liver blood flow increases in the anaesthetized rat; for in this species, in other than the lightest stages of anaesthesia, this procedure does not raise the blood pressure (unpublished observations, Grayson & Ginsburg).

The experiments described in this paper thus demonstrate the presence of two conflicting mechanisms which come into play to determine liver blood flow when adrenaline or noradrenaline is administered to an animal. First, there is a local constrictor action on the liver vessels, and secondly a reflex action due to the raised blood pressure acting through the carotid sinus and aortic arch receptors. With intravenous adrenaline the reflex effect predominates at first and the net effect is to increase liver flow. With noradrenaline the balance is more even, although, usually, the local effect on the liver is dominant and overwhelms the systemic dilator effect from the outset.

SUMMARY

1. The use of internal calorimetry in the measurement of liver blood flow in the rat and rabbit has been investigated. Subject to precautions detailed in the text, conductivity increment was found to be a reliable index of blood flow around the recorder.

2. In the conscious animal, subcutaneous or intravenous adrenaline produced a rise in liver blood flow.

3. In the anaesthetized animal subcutaneous adrenaline did not affect liver blood flow or systemic blood pressure.

4. The increase in liver blood flow was shown to be mainly reflex in nature, initiated by the rise in blood pressure and mediated through the carotid sinus and aortic arch receptors.

5. The local action of adrenaline and noradrenaline on the liver vessels was constrictor.

6. Intravenous or subcutaneous noradrenaline frequently produced a diminution in liver blood flow, the local action overriding the reflex dilator action of the raised blood pressure.

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