The effects of meclofenamate, captopril and phentolamine on organ blood flow in the conscious rabbit

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Summary

1. Systemic and regional vascular changes were measured in conscious rabbits after intravenous sodium meclofenamate, captopril and phentolamine. These drugs were used respectively to inhibit prostaglandin synthesis and angiotensin-converting enzyme, and to block $\alpha$-adrenoceptors.

2. Meclofenamate reduced renal and adrenal blood flow by 11 and 28% respectively, and doubled hepatic artery flow. The effect on renal and adrenal flow persisted in the presence of phentolamine.

3. Captopril decreased estimated peripheral resistance and increased cardiac output without changing arterial pressure. Kidney and adrenal flow increased by 70 and 21% respectively.

4. Phentolamine reduced arterial pressure and doubled flow to skeletal muscle and increased hepatic artery flow to the liver.

5. Splenic blood flow was unaffected by meclofenamate, captopril or phentolamine alone. Meclofenamate given after captopril caused a halving of splenic flow and a rise in arterial pressure; these effects were prevented by phentolamine.

6. The results point to a continuing effect of prostaglandin synthesis in maintaining blood flow to the kidney and adrenal gland independent of $\alpha$-adrenoceptor activation in resting conscious rabbits. An important modulating effect of prostaglandins on sympathetic vascular tone in the spleen is suggested.

Key words: Angiotensin-converting enzyme, captopril, meclofenamate, organ blood flow, phentolamine, prostaglandins, sympathetic nervous system.

Introduction

Blood flow in different vascular beds is influenced by local, nervous and humoral control mechanisms. The local factors affecting vascular tone enable nutrient perfusion to vital organs, such as the kidney and brain, to be regulated independently of changes in systemic haemodynamics. Amongst the factors which may be important in local blood flow regulation are prostaglandins, bradykinin and angiotensin II (ANG II). Most investigations of tissue flow have used isolated organs or anaesthetized animals and, although these provided much valuable information, they can only give an indication of events which might occur in the intact conscious animal.

The present experiments were designed to study tissue blood flow under physiological conditions in the conscious rabbit. This was done with particular reference to interaction between prostaglandins, the sympathetic nervous system and vasoactive substances controlled by converting-enzyme activity (ANG II and bradykinin). To study effects of these systems we used sodium meclofenamate (Meclomen; Parke Davis Co.), an inhibitor of prostaglandin synthesis, phentolamine (Regitine; Ciba Pharmaceuticals), an $\alpha$-adrenoceptor antagonist, and captopril (SQ 14 225; E. R. Squibb and Sons Ltd), an inhibitor of the angiotensin-converting enzyme.

Methods

Animals

Male, semi-lopeared rabbits from a single-strain colony were used. Weights ranged from
Cardiac output and distribution

Cardiac output and its distribution was measured in conscious animals by thermodilution and radioactive-microsphere techniques respectively. Sodium chloride solution (150 mmol/l: saline) at room temperature acted as the thermodilution indicator injected through a left ventricular cannula. This was given by a 2-0 ml multiple-dose pipetting syringe (Becton, Dickinson and Co.) calibrated to deliver 0.7 ml at room temperature. The temperature of the saline was kept constant by a water jacket and recorded by a thermocouple (Elektrolaboratonet TE3). The temperature transient was measured by a thermistor bead (Phillips NTC 2322 634 01152) positioned in the lower aorta between the renal arteries and the aorta bifurcation. The bead was embedded in epoxy resin at the tip of a 60 cm length of vinyl tubing (internal diameter 0.58 mm, external diameter 0.96 mm) and soldered to insulated steel wires. The wires were included within the tubing and connected in turn to a miniature plug (R. S. Components Ltd 466–472). Thus temperature changes could be recorded as voltage changes when the thermistor was connected to one arm of a Wheatstone bridge and the output displayed on a flat-bed potentiometer pen recorder (Rikadenki R-10). The area described by the voltage change was measured by planimetry after allowance had been made for recirculation of indicator. The cardiac output could then be calculated by a modification of the Stuart–Hamilton equation [1] after calibration of the thermistor bead. At least three curves were recorded for each cardiac output measurement and the mean was calculated. Cardiac output distribution was measured by left ventricular infusions of radioactive microspheres of 15 μm nominal diameter (New England Nuclear). These spheres distribute in proportion to the cardiac output, lodging in the capillary beds with less than 1% recirculation in conscious dogs [2] and anaesthetized rabbits [3]. Portions containing approximately 150 000 spheres suspended in 1.0 ml of saline were drawn into 2.0 ml plastic syringes and sonicated for 10 min in an ultrasound dissonifying bath (Brandson B-220) to disperse clumps. The radioactivity in the syringes was counted in a well gamma counter (Packard 5986).

The suspension was then infused through the left ventricular cannula over approximately 10 s and flushed with 1.0 ml of saline. The remaining activity in the syringe was counted and the total injected radioactivity calculated. The radioactivity trapped within each organ was measured in the same counter after the tissue had been divided into suitable sized portions. Previous experiments had shown a counting efficiency of greater than 98% for samples of less than 3.5 cm in height. Organ perfusion could then be calculated by the equation: organ blood flow = \( a \times CO/b \), where \( a \) = total organ radioactivity in c.p.s., \( b \) = total injected radioactivity in c.p.s. and \( CO \) = cardiac output (determined by thermodilution).

Batches of microspheres were labelled with one of three isotopes; \(^{46}\)Sc, \(^{113}\)Sn and \(^{57}\)Co were chosen because of their wide peak-energy separation. By giving one infusion after another in random sequence, it was thus possible to obtain measurement of cardiac output distribution at three different times. On a separate series of 16 rabbits no adverse effects were seen on blood pressure, pulse rate, cardiac output or renal blood flow after three successive infusions of 150 000 microspheres.

Surgery

Surgery was performed under sterile conditions. Anaesthesia was induced with intravenous phenobarbitone (50–100 mg) and maintained with halothane administered through an endotracheal tube. At sites of incision, the skin was infiltrated with 0.5% lignocaine which enabled the general anaesthesia to be kept to a minimum. The left femoral artery was exposed 1 cm below the femoral triangle and cannulated by the vinyl tubing supporting the thermistor bead. The bead was positioned in the lower aorta and the cannula and steel wires were channelled subcutaneously to the neck where the plug was sutured. The left common carotid artery was exposed and cannulated with a 20 cm length of vinyl tubing (internal diameter 0.50 mm, external diameter 0.98 mm) filled with saline. The other end of the tubing was connected to a pressure transducer (Statham P231d), the signal from which was displayed, via a pre-amplifier (Sanborn 7700 series 3501100C) on the flat-bed pen recorder. The tubing was fed down the carotid artery towards the aortic valve, arrival at which was detected by physical resistance or

2.9 to 4.3 kg, corresponding to 4–5 months maturity. The animals were housed in cages in rooms kept at a constant temperature of 23°C. Lighting was controlled on a 12 h cycle. Free access was given to tap water and standard commercial feeding pellets (Milnes Ltd) supplemented by lucerne, lettuce and cabbage leaves both before and after surgery. This supplied approximately 20 mmol of sodium/day.
pressure damping. The left ventricle was then cannulated under pressure monitoring. The tubing was carefully positioned so that its presence did not cause ventricular premature beats and its tip was not touching the endocardium. The cannula was filled with heparin/saline (1000 units/ml), secured in position and taken subcutaneously to the neck to emerge adjacent to the thermistor plug. The wounds were closed in layers and the animals returned to their cages after injections of benzyl penicillin subcutaneously). Surgery was completed within 60–90 min and the animals were awake within 15 min.

Experimental protocol (Fig. 1)

Twenty-four hours after surgery the animals were placed in a wooden box that allowed a limited degree of movement without harsh restraint. The right ear was infiltrated with 0.5% lignocaine and the central ear artery cannulated with polyethylene tubing (external diameter 0.90 mm, internal diameter 0.50 mm). The pressure signal was amplified and recorded as described above. The animals were then left undisturbed for 15 min after which 2 ml of blood was slowly withdrawn from the central ear artery for measurement of blood gases and plasma renin activity. Five minutes later the first thermocilusion curves were recorded, followed immediately by infusion of microspheres labelled with the first isotope. The resulting measurements represented the pretreatment state. The animals were then divided into four groups. Group 1 (n = 5) received a control infusion of saline (1 ml/kg) through a marginal ear vein over 5 min. After a further 5 min, cardiac output and organ blood flow were measured with a second isotope. An intravenous infusion of sodium meclofenamate (6 mg/kg) was then given to inhibit prostaglandin synthesis [4]. Fifteen minutes later the effect of this drug alone on organ flow was measured by thermodilution and administration of the third isotope. Group 2 (n = 5) received an infusion of captopril (1 mg/kg) in place of saline, but was otherwise identical with that of group 1. Group 3 (n = 7) was given an infusion of phentolamine (2 mg/kg) over 10 min, followed 5 min later by measurement of organ blood flow. Meclofenamate was then given as for groups 1 and 2, followed by a reinforcing infusion of phentolamine (1.5 mg/kg). This latter was found necessary to ensure adequate α-adrenoceptor blockade at the time of flow measurements with the third isotope. Doses were the same as in groups 2 and 3. In group 4 (n = 4) both captopril and phentolamine were given before the first isotope, and meclofenamate and phentolamine before the third. Doses were the same as in groups 2 and 3.

Thus for each group the first measurement was made before any treatment, the second measurement recorded organ blood flow 5 min after infusion of either saline, captopril, phentolamine or the combination of the two last-named drugs (first ‘treatment’ period). The final measurement in each group, 25 min after completion of these infusions, recorded the added influence of meclofenamate (second treatment period). The timing of observations relative to drug administration was kept constant throughout. At the end of each experiment a further 1 ml of blood was collected for measurement of plasma renin activity. Animals from each group were studied in rotation with the exception of one inadvertently transposed to group 3. Three animals in whom the left ventricular cannula was not in place at the time of autopsy were excluded from subsequent analysis.

The animals were killed with a barbiturate overdose. At autopsy kidneys, adrenals, testis, hip-flexor skeletal muscle, right hepatic lobule, spleen, heart and brain were carefully removed and weighed, and the total radioactivity contained within them was counted as outlined above.

Captopril and phentolamine inhibitory activity

In two separate groups of six rabbits each the inhibitor activity of captopril (1 mg/kg) and
phenolamine (2, followed by 1.5 mg/kg) was determined by measuring the rise in blood pressure induced by bolus intravenous injection of angiotensin I (300 μg/kg; [Asp', Ile5]ANG I; Schwarz–Mann) or noradrenaline (1 μg/kg; Levophed; Winthrop Laboratories) respectively. Pressures were measured by cannulation of the central ear artery as described above. A control pressure response was measured before and 5 and 25 min after infusion of inhibitor. In this way a measure of inhibitor activity was obtained which corresponded in time to observations made with isotopes 2 and 3 in the main experiments. Captopril reduced the hypertensive effect of intravenous ANG I on average by 97 and 89% at 5 and 25 min after the inhibitor respectively. The pressor response to noradrenaline was reduced by an average of 77 and 79% at 5 and 25 min respectively after phenolamine.

**Assay of plasma renin activity**

The method is a modification of that of Boyd, Adamson, Fitz & Peart [5] involving radioimmunoassay of ANG I with antisera directed towards [Asp’, Ile5]ANG I (Schwarz–Mann). Blood (1.0 ml) was taken into ice-cold tubes containing 10 μl of EDTA (0.27 mol/l). The final plasma activity was determined after incubation at 37°C for 1 h at pH 7.5 in the presence of 8-hydroxyquinoline sulphate and 2,3-dimercaptopropanol.

**Solutions**

Phentolamine and captopril were dissolved in saline and meclofenamate in a sodium bicarbonate/chloride buffer at pH 8.5. All solutions were made up in concentrations permitting the administered dose to be given in a millilitre volume, numerically equal to the kilogram weight of the animal.

**Statistical analysis**

Unless otherwise stated Student’s paired t-test was used to compare pretreatment values with those after drug administration, and group comparisons were made by one-way analysis of variance [6]. The null hypothesis was rejected when P < 0.05. Group values are stated as means ± SEM in the text and Figures.

**Results**

In group 1 intravenous saline (1 ml/kg) produced no significant systemic or regional vascular changes.

There were no significant differences (P < 0.05) in pretreatment blood gases between any of the groups (analysis of variance). For all the animals (n = 21) the mean values were: Pao2 98 ± 2 mmHg (13.1 ± 0.3 kPa); Paco2 34.1 ± 1 mmHg (4.5 ± 0.1 kPa); pH 7.41 ± 0.01.

An indirect measure of biological activity of meclofenamate was obtained by its ability to suppress plasma renin activity [7]. Thus in group 1 mean pretreatment plasma renin activity was reduced from 23.9 ± 1.7 to 12.7 ± 1.7 ng of ANG I h⁻¹ ml⁻¹ (P < 0.01; n = 5) after 6 mg of meclofenamate/kg.

<table>
<thead>
<tr>
<th>Group</th>
<th>Treatment</th>
<th>Mean arterial pressure (mmHg)</th>
<th>Cardiac output (ml min⁻¹ kg⁻¹)</th>
<th>Estimated total peripheral resistance (kPa⁻¹ s × 10⁻⁶)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>First</td>
<td>Second</td>
<td>Pre-treatment</td>
<td>Second</td>
</tr>
<tr>
<td>1</td>
<td>Saline</td>
<td>Meclofenamate</td>
<td>92 ± 5</td>
<td>92 ± 2</td>
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<td></td>
<td>(n = 5)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
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<td>Captopril</td>
<td>Meclofenamate</td>
<td>79 ± 4</td>
<td>75 ± 6</td>
</tr>
<tr>
<td></td>
<td>(n = 5)</td>
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<td></td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>Phentolamine</td>
<td>Meclofenamate/ phentolamine</td>
<td>84 ± 3</td>
<td>72 ± 2</td>
</tr>
<tr>
<td></td>
<td>(n = 7)</td>
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<td></td>
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<tr>
<td>4</td>
<td>Captopril/</td>
<td>Meclofenamate/ phentolamine</td>
<td>90 ± 7</td>
<td>64 ± 6</td>
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<td></td>
<td>(n = 4)</td>
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</table>

TABLE 1. Sequential effects of intravenous drugs on systemic haemodynamics in conscious rabbits

Estimated total peripheral resistance was derived from the values for mean arterial pressure and cardiac output. Results are means ± SEM. *P < 0.05.
Effect of drugs on systemic haemodynamics

(Table I)

In group 1 meclofenamate given after saline caused no significant changes ($P > 0.05$) in mean arterial pressure, cardiac output or estimated total peripheral resistance. Five minutes after an infusion of captopril (group 2) cardiac output increased in all five animals ($P < 0.05$, paired t-test on log values), with little change in mean arterial pressure and a fall in estimated total peripheral resistance of 18% ($P < 0.05$). In the presence of captopril meclofenamate significantly increased mean arterial pressure ($P < 0.05$) to pretreatment values without further affecting cardiac output. In group 3 arterial pressure fell after phentolamine ($P < 0.01; n = 7$) and was not significantly changed by subsequent infusion of meclofenamate. In group 4 mean arterial pressure and estimated peripheral resistance were reduced ($P < 0.05; n = 4$) by the combined infusion of captopril and phentolamine and were not further affected by meclofenamate.

Renal haemodynamics

Meclofenamate, given after saline in group 1, reduced renal blood flow from $11.7 \pm 1.2$ to $10.5 \pm 1.4 \text{ ml min}^{-1} \text{ g}^{-1}$ ($P < 0.05; n = 5$), a fall of 11%.

In group 3 renal blood flow was unaffected by phentolamine ($10.4 \pm 0.4$ and $10.2 \pm 1.0 \text{ ml min}^{-1} \text{ g}^{-1}$ before and after phentolamine respectively), but after meclofenamate flow fell by 12% with a significant rise in estimated renal vascular resistance from $1.85 \pm 0.1 \times 10^{-5}$ kPa $\text{ l}^{-1} \text{s}$ pretreatment to $2.36 \pm 0.1 \times 10^{-5}$ after meclofenamate ($P < 0.05; n = 7$).

Renal haemodynamic changes induced by intravenous captopril are shown in Fig. 2. Renal blood flow increased 70% from $8.7 \pm 0.2$ to $14.9 \pm 2.6 \text{ ml min}^{-1} \text{ g}^{-1}$ ($P < 0.05; n = 5$). The percentage cardiac output received by the kidneys also increased by 50% ($P < 0.05; n = 5$) after captopril and estimated renal vascular resistance fell from $2.02 \pm 0.13 \times 10^{-5}$ to $1.22 \pm 0.2 \times 10^{-5}$ unit ($P < 0.01; n = 5$). When meclofenamate was given after captopril, estimated renal vascular resistance [$1.54 \pm 0.07 \times 10^{-5}$] remained below control values ($P < 0.025; n = 5$). Although flow fell towards normal after meclofenamate in the four animals showing the greatest response to captopril (Fig. 2), these changes were not statistically significant.

The combination of captopril and phentolamine (group 4) resulted in an increase in renal blood flow ($8.3 \pm 0.6$ to $12.3 \pm 1.4 \text{ ml min}^{-1}$

![Fig. 2. Drug effects on renal haemodynamics in individual rabbits in group 2. Estimated renal vascular resistance was calculated by dividing systemic arterial pressure by renal blood flow per unit weight of kidney. Mean values ± SEM are shown. * $P < 0.05$; ** $P < 0.025$; *** $P < 0.01$. CAPT, Captopril; MECL, meclofenamate.](image)

Adrenal haemodynamics (Fig. 3)

In group 1 adrenal blood flow was reduced from $3.5 \pm 0.2$ to $2.6 \pm 0.2 \text{ ml min}^{-1} \text{ g}^{-1}$ ($P < 0.005; n = 5$) after meclofenamate infusion, a mean reduction of 28%.

In group 2 captopril increased mean adrenal blood flow from $2.9 \pm 0.3$ to $3.5 \pm 0.2 \text{ ml min}^{-1} \text{ g}^{-1}$ ($P < 0.01; n = 5$) and reduced adrenal vascular resistance from $6 \pm 0.4 \times 10^{-5}$ to $4.67 \pm 0.2 \times 10^{-5}$ kPa $\text{ l}^{-1} \text{s}$ ($P < 0.05; n = 5$).
However, captopril increased cardiac output and the percentage received by the adrenal glands did not change. Meclofenamate given after captopril resulted in a fall in adrenal flow to pretreatment values.

Phentolamine did not affect adrenal blood flow (group 3), but reduced estimated adrenal vascular resistance from 5.95 (±0.67) × 10⁻⁵ to 4.73 (±0.2) × 10⁻⁵ unit (∗ ∗ < 0.01; n = 7). Subsequent infusion of meclofenamate reduced adrenal blood flow from 3.5 ± 0.2 to 2.6 ± 0.2 ml min⁻¹ g⁻¹ (∗ < 0.02; n = 7), a fall almost identical with that seen in group 1 in the absence of α-adrenoreceptor blockade. At the same time estimated adrenal resistance rose to 5.6 (±0.4) × 10⁻⁵ unit (∗ ∗ < 0.01; n = 5; compared with phentolamine).

In group 4 adrenal blood flow was unchanged by the combination of captopril and phentolamine (2.9 ± 0.2 to 2.9 ± 0.6 ml min⁻¹ g⁻¹) or by the subsequent addition of meclofenamate (2.6 ± 0.4 ml min⁻¹ g⁻¹). Changes in groups 1 to 3 are represented in Fig. 3.

**(a)**

### Splenic haemodynamics (Fig. 4)

Meclofenamate given after saline (group 1) or phentolamine (group 3) had no effect on splenic haemodynamics. Captopril or phentolamine given alone in the first treatment period were also without effect.

In the presence of captopril (group 2) meclofenamate caused a marked fall in splenic blood flow from 9.9 ± 1.3 in the pretreatment state to 4.8 ± 0.8 ml min⁻¹ g⁻¹ (∗ < 0.01; n = 5; Fig. 4a). When both captopril and phentolamine were given in the first treatment period (group 4), meclofenamate was without effect (Fig. 4b).

**Effects on blood flow in hepatic artery and to skeletal muscle, brain, heart and testis (Table 2)**

Meclofenamate alone and phentolamine alone doubled hepatic arterial flow in groups 1 and 3 (P < 0.05; n = 5). Flow remained elevated when meclofenamate was given after phentolamine in group 3 (P < 0.005 compared with pretreatment).

Phentolamine alone increased skeletal muscle flow (P < 0.05), an effect that persisted after meclofenamate (P < 0.05; n = 7).

There were no significant changes observed in flow to the brain, heart or testis.
Vasoactive drugs and organ blood flow

### TABLE 2. Drug effects on blood flow to skeletal muscle and the hepatic artery

Results are means ± SEM. *P < 0.05 (Wilcoxon's test for paired values).

<table>
<thead>
<tr>
<th>Group</th>
<th>Treatment</th>
<th>Blood flow (ml min⁻¹ 100 g⁻¹)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Pretreatment</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Skeletal muscle</td>
</tr>
<tr>
<td></td>
<td></td>
<td>First</td>
</tr>
<tr>
<td>1</td>
<td>Saline</td>
<td>23</td>
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<tr>
<td>(n = 5)</td>
<td>Meclofenamate</td>
<td>± 3</td>
</tr>
<tr>
<td>2</td>
<td>Captopril</td>
<td>14</td>
</tr>
<tr>
<td>(n = 5)</td>
<td>Meclofenamate</td>
<td>± 2</td>
</tr>
<tr>
<td>3</td>
<td>Phentolamine/</td>
<td>20</td>
</tr>
<tr>
<td>(n = 7)</td>
<td>Meclofenamate/</td>
<td>± 4</td>
</tr>
<tr>
<td>4</td>
<td>Captopril/</td>
<td>24</td>
</tr>
<tr>
<td>(n = 4)</td>
<td>Meclofenamate/</td>
<td>± 11</td>
</tr>
<tr>
<td></td>
<td>Phentolamine</td>
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</tbody>
</table>

**Discussion**

Meclofenamate, captopril and phentolamine were used to study the relative influence of tissue prostaglandins, converting-enzyme activity and sympathetically mediated α-adrenoceptor stimulation on organ blood flow in the resting state. This was done with the knowledge that any effect of converting-enzyme inhibition has to be interpreted with respect to its dual action on angiotensin conversion and bradykinin destruction.

The efficacy of captopril and phentolamine as inhibitors was confirmed by blockade of the pressor effects of intravenous ANG I and noradrenaline respectively. The degree of blockade of prostaglandin synthesis by meclofenamate was not measured directly; however, the dose used has been shown to produce more than 90% inhibition of renal prostaglandin synthesis in rabbits [8]. We were able to provide indirect evidence of this action by suppression of plasma renin activity by 50%, renin release being mediated in part by renal prostaglandin synthesis [9].

The effects were diverse and the principal changes demonstrated are perhaps best summarized as follows: (a) meclofenamate caused a fall in adrenal and renal flow and an increase in hepatic artery flow (the fall in adrenal and renal flow were unaffected by phentolamine); (b) captopril caused systemic renal and adrenal vasodilatation and an increase in cardiac output; (c) phentolamine alone reduced arterial pressure and increased muscle and hepatic artery flow. Renal, adrenal and splenic flow were unaffected by phentolamine alone; (d) the fall in adrenal and renal blood flow induced by meclofenamate was unaffected by phentolamine; (e) meclofenamate given after captopril caused a pronounced fall in splenic flow (this interaction was prevented by prior phentolamine administration).

The function of tissue prostaglandins as regulators of vascular tone is uncertain. Work on isolated organs and anaesthetized animals has shown species differences [10]. Moreover in some instances a particular prostaglandin may dilate one vascular bed and constrict another [11]. Recently attention has been focused on the vasodilator prostacyclin, which is synthesized in the vessel walls. Of all the prostaglandins, prostacyclin is perhaps most likely to influence vascular tone under physiological circumstances. The reduction of renal and adrenal flow by meclofenamate suggests that synthesis of vasodilator prostaglandins normally helps to maintain flow to these organs. In the isolated perfused rabbit kidney release of noradrenaline from renal nerve terminals is inhibited by prostaglandin E₂ [12], whereas prostaglandin I₂ appears to reduce the postjunctional effect of this catecholamine [13]. In anaesthetized dogs renal artery infusion of noradrenaline results in vasoconstriction, but despite continuing infusion this is followed by vasodilatation concurrent with the appearance of urinary prostaglandin E activity [14]. Similarly canine renal prostaglandin production is increased by ANG II and the renal vasoconstriction so produced is potentiated by inhibitors of prostaglandin synthesis [15].

Prostaglandins seem to play only a minor role in regulating renal blood flow in the conscious dog [16], unless the renal circulation is stressed by haemorrhage or vasoconstriction [16, 17]. The conscious resting rabbit, however, does respond to structurally dissimilar inhibitors of prosta-
glandin synthesis by renal vasoconstriction [18, 19]. Man may be similarly susceptible, especially in the presence of renal failure [20].

These experiments examined the question as to whether renal vasoconstriction after inhibition of prostaglandin synthesis may be due to unopposed neurogenic tone. In the anaesthetized dog after haemorrhage the effect of the prostaglandin synthesis inhibitor, indomethacin, was abolished by renal nerve section together with infusion of a specific ANG II antagonist [21]. However, in the present study, meclofenamate caused a fall in renal blood flow which, although small, was not reduced by prior adrenal $\alpha$-receptor blockade, suggesting that the vasoconstriction was independent of sympathetic tone.

The large increase in renal blood flow seen after captopril is of considerable interest. An increase in renal blood flow after converting-enzyme inhibition has been demonstrated in anaesthetized animals [22, 23]. In conscious dogs [24] and man [25] the effects seem dependent on sodium depletion. However, our rabbits are maintained on a relatively high sodium diet by human standards (5.7 mmol day$^{-1}$g$^{-1}$).

There has been uncertainty as to the anti-hypertensive mechanism of captopril and speculation as to whether this is due to inhibition of ANG II formation or to a concomitant decrease in bradykinin breakdown [26].

Captopril increased renal flow to kidneys and adrenals, both target organs for ANG II, and hence may simply be causing vasodilatation by inhibiting ANG II formation. The relatively high concentration salt diet the animals were eating could be expected to render them sensitive to ANG II; however, observations that salt-loaded animals are generally resistant to the hypertensive effects of captopril and angiotensin receptor antagonists casts some doubt on this explanation.

Murphy, Waldron & Goldberg [27] found that indomethacin markedly attenuated the enhancement by captopril of the hypertensive response to bradykinin in intact, but not anephric rabbits. They suggested that bradykinin was stimulating release of renal vasodilator as has been demonstrated in perfused dog kidneys [28]. In our experiments although blood pressure rose when meclofenamate was given after captopril renal vasodilatation persisted. Thus the renal vasodilatation induced by captopril was unlikely to be mediated by prostaglandins, unless one postulates a stimulus sufficient to overwhelm the inhibitory effect of the dose of meclofenamate given. This leaves open the possibility that captopril exerts its vasodilatation effect by causing local accumulation of bradykinin. The possibility that captopril causes renal vasodilatation by a mechanism unrelated to angiotensin or bradykinin also needs to be considered.

The adrenal gland showed qualitatively similar effects to the kidney. Indeed adrenal vasocostriction after meclofenamate was more pronounced and has not previously been reported. As with the kidney, this effect was not altered by phentolamine and hence seems independent of sympathetic tone. Renal prostaglandins may modulate adrenal aldosterone production via an effect on the renin-angiotensin system and perhaps by an additional direct action on aldosterone release [29]. The present work points to the possibility that prostaglandin-stimulated aldosterone release may also depend on changes in adrenal blood flow per se.

The pronounced reduction in splenic blood flow to half the pretreatment values when meclofenamate was given after captopril points to an important buffering role for prostaglandins in the spleen. Prevention of this reduction in splenic flow by phentolamine (Fig. 4) suggests the following sequence of events: captopril causes systemic vasodilatation and hence induces increased sympathetic nerve activity through the baroreflex. In the spleen the resulting vasoconstriction would normally be countered by enhanced prostaglandin synthesis. Inhibition of prostaglandin synthesis by meclofenamate thus results in sympathetically mediated vasocostriction. This in turn can be prevented by the $\alpha$-adrenoceptor-blocking action of phentolamine.

The increase in blood flow to skeletal muscle, and to a lesser extent in the hepatic artery, after phentolamine (Table 2), implies resting $\alpha$-adrenergic tonic control in these circulations. Hepatic arterial vasodilatation after meclofenamate is less easy to explain. It may reflect inhibition of local vasoconstrictor prostaglandins or passive shunting of blood flow from constricted vascular beds. In a separate series of experiments we have observed a reciprocal relationship between gastric and hepatic artery blood flows (unpublished observations). It has been reported that gastric blood flow falls after indomethacin [30]. If inhibition of prostaglandin synthesis reduces intestinal blood flow in the conscious rabbit, the increase in hepatic artery blood flow after meclofenamate may be an attempt to maintain total hepatic perfusion in the presence of a diminished portal venous flow.

In summary, in the conscious rabbit the reduction in renal and adrenal blood flow by
sodium meclofenamate points to a continuing effect of prostaglandin synthesis in maintaining flow to these organs, as the effects persist even in the presence of pharmacological α-adrenoceptor blockade. Endogenous prostaglandins appear to have an important role in modulating the effects of sympathetic vascular tone in the spleen and to influence hepatic artery flow. The mechanism by which captopril induced renal and adrenal vasodilatation remains uncertain; with the effect persisting in the presence of meclofenamate in the kidney, but not the adrenal.

Acknowledgments

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