Presinusoidal vessels predominantly contract in response to norepinephrine, histamine, and KCl in rabbit liver

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Shibamoto, Toshishige, Hong-Gang Wang, Takashige Miyahara, Satoshi Tanaka, Hisao Haniu, and Shozo Koyama. Presinusoidal vessels predominantly contract in response to norepinephrine, histamine, and KCl in rabbit liver. J. Appl. Physiol. 87(4): 1404–1412, 1999.—In rabbit livers, it is not well known which segments of the hepatic vasculature are predominantly contracted by various vasoconstrictors. We determined effects of histamine, norepinephrine, and KCl on hepatic vascular resistance distribution in isolated rabbit livers perfused via the portal vein with 5% albumin-Krebs solution at a constant flow rate. Hepatic capillary pressure was measured by double vascular occlusion pressure (Pdo) and was used to determine portal (Rpv) and hepatic venous (Rhv) resistances. A bolus injection of either histamine or norepinephrine dose-dependently increased portal venous pressure but not Pdo, resulting in a dose-dependent increase in Rpv and no changes in Rhv. KCl (50 mM), when injected in antegrade-perfused livers, contracted the presinusoidal vessels selectively with liver weight loss. Although KCl significantly increased Rhv in retrogradely perfused livers, the increase in Rpv by 400% of baseline predominated over the increase in Rhv by 85% of baseline. In the retrogradely perfused livers, KCl produced an initial liver weight loss followed by a profound weight gain. We conclude that histamine and norepinephrine selectively contract the presinusoidal vessels. The results on KCl effects suggest that this selective presinusoidal constriction might be possibly due to predominant distribution of functionally active vascular smooth muscle in the presinusoidal vessels rather than the hepatic vein in rabbit livers.

We have recently shown, by using vascular occlusion methods for measurement of hepatic capillary pressure, that hepatic longitudinal vascular responsiveness differs depending on vasoconstrictive substances in isolated perfused canine livers (20, 22, 23, 25). Either histamine (20, 22) or the thromboxane A2 analog (22) predominantly contracts the canine hepatic vein with resultant hepatic congestion, whereas norepinephrine constricts both the presinusoidal vessels and hepatic artery more vigorously than the hepatic vein, with reduction of liver weight (20). In contrast, acetylcholine (20) and platelet activating factor (23) constrict both portal and hepatic veins in a similar magnitude. However, there could be species differences in the primary site of hepatic vasoconstriction. In isolated perfused rabbit livers, we have recently reported that endothelin-1 selectively contracts the presinusoidal vessels (24). However, only a limited number of studies have been reported to determine effects of vasoconstrictors on rabbit hepatic vascular resistance distribution (19).

Therefore, in the present study, we determined, by using the double-vascular-occlusion method, the effects of histamine and norepinephrine on longitudinal vascular resistance distribution in isolated rabbit livers perfused with 5% albumin-Krebs buffer. The reason for adopting histamine and norepinephrine as the vasoconstrictors in the present study was that norepinephrine predominantly constricts the presinusoidal vessels over the hepatic vein (3, 19, 20), whereas histamine predominantly contracts the hepatic vein over the presinusoidal vessels in canine livers (12, 16, 20, 22). In this study, we assumed that the distribution of vascular smooth muscle between presinusoidal vessels and hepatic veins may play a key role in hepatic vasoreactivity. Thus another purpose of the present study was to determine which segment of the hepatic vasculature was primarily contracted by KCl, which in turn could act directly on vascular smooth muscle without mediation of receptor activation. We used the reverse-perfusion technique to clarify more accurately the hepatic vascular response to KCl.

MATERIALS AND METHODS

Isolated liver preparation. Twenty rabbits, weighing 2.2 ± 0.1 (SE) kg, were anesthetized with pentobarbital sodium (30 mg/kg iv) and mechanically ventilated with room air. The experiments were performed in adherence to the guidelines of the Physiological Society of Japan for the use of experimental animals, and the protocols were approved by the Shinshu University Animal Care Committee. The method for isolated rabbit liver preparation was previously described (24). Catheters were placed in the left jugular vein and in the right carotid artery. After left thoracotomy and laparotomy, loose ligatures were placed around the hepatic artery, portal vein, inferior vena cava, and common bile duct. At 5 min after heparinization (500 U/kg iv), the rabbit was rapidly bled through the carotid arterial catheter. After ligation of the aforementioned vessels and bile duct, the liver was rapidly excised and weighed. Then, the portal vein and vena cava were cannulated with plastic cannulas (3 mm ID), whereas the hepatic artery was ligated to simplify analysis of the intrahepatic vascular circuit. The common bile duct was also cannulated with polyethylene tubing. Perfusion was begun within 5 min after excision of the liver.

The cannulated liver was suspended from an electric balance (LF-6, Murakami Koki) and perfused via the portal vein at a constant perfusion flow rate of 0.157 ± 0.008 l/min.

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the perfusate in the reservoir was continuously bubbled with perfusate was maintained at 37°C by using a water bath, and the perfusate in the reservoir was continuously bubbled with 95% O₂-5% CO₂ (inflow perfusate P O₂, 300 Torr). A bubble trap was placed in the inflow line. Portal (Ppv) and hepatic venous (Phv) pressures were measured through the corresponding sidearm cannula by using pressure transducers (Gould) referenced to the level of the portal vein at the hepatic hilus. The perfusion flow rate (Q) was measured with an electromagnetic flowmeter (MFV 1200, Nihonkohden), and the flow probe was positioned in the outflow line. To occlude the portal and hepatic venous lines simultaneously for measurement of the double occlusion pressure (Pdo), solenoid valves were placed around the perfusion tubes upstream from the Ppv sidearm cannula and downstream from the Phv sidearm cannula. Bile was continuously collected in a small tube suspended from the force transducer (45196A, NEC-Sanei). The weight of the tube was continuously measured, and the bile flow rate was expressed as grams per minute per 100 g liver weight. The liver weight, bile flow, and hemodynamic variables were continuously monitored and displayed on a thermal physiograph (8K23, NEC-Sanei). Initially, an isogravimetric state (neither weight gain nor loss) was obtained by adjusting the flow rate and the height of the reservoir independently to maintain Ppv and Phv at a level within the normal perfusion range, as described below.

Experimental protocol. Hepatic hemodynamic parameters were observed for at least 20 min after the start of perfusion, during which an isogravimetric state was reached at a Ppv of 6–9 mmHg, a Phv of 0–2 mmHg, and a Q of 0.195 ± 0.014 (SE) l·min⁻¹·100 g liver wt⁻¹ (n = 15). After the baseline measurements were taken, the perfused livers were challenged with either histamine, norepinephrine, or KCl. In the norepinephrine group (n = 5), 0.33, 3.3, 33, or 330 µg, or 3.3 mg of norepinephrine were injected as a bolus into the portal line and attained the final perfusate concentration of 0.01, 0.1, 1, 10, or 100 µM, respectively. In the histamine group (n = 5), 0.37, 3.7, 37, 110, or 370 µg, or 3.7 mg of histamine were injected as a bolus into the portal line and attained the final perfusate concentration of 0.01, 0.1, 1, 10, 30, or 100 µM, respectively. In the KCl group (n = 5), KCl was injected as a bolus into the portal line and attained the final perfusate concentration of 50 mM. In the retrograde perfusion KCl group (n = 5), the liver was perfused retrogradely from the hepatic vein, and KCl was injected into the inflow (hepatic venous) line to attain the final perfusate concentration of 10, 25, and 50 mM. The volume of each agent injected was adjusted to <1 ml.

Hepatic sinusoidal pressure was measured by the double-occlusion method (24, 25). Both the inflow and outflow lines were simultaneously and instantaneously occluded by using the equipped solenoid valves, after which Ppv and Phv rapidly equilibrated to a similar or identical pressure, which was Pdo. In each experimental group, Pdo was measured at baseline and 1, 2, 3, 6, 10, 20, and 30 min after an injection of each agent. When low doses of agents were injected, Pdo was determined only at maximal vasoconstriction.

The total portal-hepatic venous (Rtot), portal or presinusoidal (Rpv), and hepatic venous (Rhv) resistance were calculated as follows

\[
R_{\text{tot}} = \frac{(\text{Ppv} - \text{Phv})}{Q} \\
R_{\text{pv}} = \frac{(\text{Ppv} - \text{Pdo})}{Q} \\
R_{\text{hv}} = \frac{(\text{Pdo} - \text{Phv})}{Q}
\]

Statistics. All results are expressed as means ± SE. Comparisons were made by using Student's t-tests. A P value < 0.05 was considered significant.

RESULTS

Effects of histamine and norepinephrine on hepatic hemodynamics, liver weight, and bile flow. The initial wet liver weight measured immediately after excision was 86 ± 6 g (n = 15). Pdo at baseline state of 15 anterogradely perfused livers was 3.7 ± 0.2 mmHg, with Ppv of 7.3 ± 0.3 mmHg and Phv of 12.0 ± 2.2 mmHg at Q of 0.195 ± 0.014 l·min⁻¹·100 g liver wt⁻¹. The calculated Rtot was 33.0 ± 4.1 mmHg·l⁻¹·min⁻¹·100 g liver wt⁻¹. The segmental vascular resistances of Rpv and Rhv were 20.2 ± 3.8 and 12.9 ± 1.2 mmHg·l⁻¹·min⁻¹·100 g liver wt⁻¹, respectively, and the Rhv-to-Rtot ratio (Rhv/Rtot) was 0.41 ± 0.04.

Figure 1 (left) shows a representative example of the response to an injection of 330 µg norepinephrine (10 µM) into a perfused rabbit liver. Soon after an injection of norepinephrine, Ppv increased from 5.3 to 14.7 mmHg, indicating that vasoconstriction occurred. Phv at 1 mmHg was not changed because the perfusion rate of 0.12 l/min was maintained. Capillary (sinusoidal) pressure (i.e., Pdo) at 1 min after norepinephrine injection was 3.3 mmHg and was similar to the baseline value of 3.3 mmHg. Thus, after norepinephrine, the pressure gradient between Ppv and Pdo was increased from the baseline value of 2.3 to 11.4 mmHg, whereas the pressure gradient between Ppv and Phv was not much changed. This finding indicates that norepinephrine selectively increased the pressure gradient from the portal vein to the capillary, that is, Rpv. Liver weight was rapidly decreased after norepinephrine injection and began to return toward the baseline when elevated Ppv began to decrease. The bile flow rate also decreased, along with liver weight loss and hepatic vasoconstriction. Hepatic vasoconstriction peaked within 3 min after an injection of norepinephrine, followed by a gradual return to baseline by 30 min after injection. The selective presinusoidal vessel constriction, liver weight loss, and a decrease in the bile flow were all similarly observed in the liver injected with 100 µM histamine, as shown in Fig. 1 (right), although the magnitude of the responses was small compared with that to 10 µM norepinephrine (Fig. 1, left).

Table 1 shows the summary data of Ppv, Pdo, and Phv after norepinephrine and histamine. Ppv increased dose dependently, with peak values of 23.9 ± 2.1 mmHg at 100 µM norepinephrine and 12.0 ± 1.4 mmHg at 100 µM histamine. There were no significant changes in Pdo after norepinephrine or histamine [3.6 ± 0.3 vs. 3.9 ± 0.5 mmHg, baseline vs. after 100 µM norepinephrine (n = 5); 3.3 ± 0.3 vs. 3.3 ± 0.4 mmHg, baseline vs. after 100 µM histamine (n = 5)].

Figure 2 shows the peak changes in Rpv, Rhv, Rhv/Rtot ratio, and liver weight after injection of norepinephrine. In livers injected with norepinephrine, Rpv increased in a dose-dependent manner at 0.1–100 µM, reaching the maximum level of 408 ± 33% of baseline
at 100 µM. In contrast, Rhv did not change significantly even at the highest concentration of 100 µM norepinephrine, and therefore the Rhv/Rtot ratio decreased to 39 ± 2% of the baseline values.

Figure 3 shows the peak changes in Rpv, Rhv, Rhv/Rtot ratio, and liver weight after injection of histamine. In response to histamine, Rpv significantly increased at 1 µM, reaching the maximum level of 197 ± 9% of the baseline at 100 µM. In contrast, Rhv did not change significantly at concentrations ranging from 0.01 to 100 µM. Therefore, the Rhv/Rtot ratio decreased to 64 ± 3% of the baseline values at 100 µM histamine.

Liver weight was decreased dose dependently after either norepinephrine or histamine (Figs. 2 and 3). The maximum liver weight loss was 12.9 ± 2.4 g/100 g liver wt at 10 µM norepinephrine, and 3.6 ± 0.5 g/100 g liver

Table 1. Changes in portal venous pressure, double-occlusion pressure, and hepatic venous pressure after injection of norepinephrine, histamine, or KCl in isolated perfused rabbit livers

<table>
<thead>
<tr>
<th>Drug</th>
<th>Ppv, mmHg</th>
<th>Pdo, mmHg</th>
<th>Phv, mmHg</th>
</tr>
</thead>
<tbody>
<tr>
<td>Norepinephrine</td>
<td>Baseline</td>
<td>After</td>
<td>Baseline</td>
</tr>
<tr>
<td>100 µM</td>
<td>8.5 ± 0.3</td>
<td>23.9 ± 2.1*</td>
<td>3.6 ± 0.3</td>
</tr>
<tr>
<td>10 µM</td>
<td>8.0 ± 0.6</td>
<td>16.7 ± 1.5*</td>
<td>3.7 ± 0.4</td>
</tr>
<tr>
<td>1 µM</td>
<td>7.7 ± 0.4</td>
<td>12.2 ± 1.2*</td>
<td>4.0 ± 0.3</td>
</tr>
<tr>
<td>0.1 µM</td>
<td>7.9 ± 0.5</td>
<td>9.7 ± 0.8*</td>
<td>3.9 ± 0.3</td>
</tr>
<tr>
<td>0.01 µM</td>
<td>7.7 ± 0.5</td>
<td>8.3 ± 0.5</td>
<td>3.9 ± 0.3</td>
</tr>
<tr>
<td>Histamine</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>100 µM</td>
<td>7.8 ± 0.6</td>
<td>12.0 ± 1.4*</td>
<td>3.3 ± 0.3</td>
</tr>
<tr>
<td>10 µM</td>
<td>7.5 ± 0.5</td>
<td>10.1 ± 1.4*</td>
<td>3.4 ± 0.2</td>
</tr>
<tr>
<td>1 µM</td>
<td>7.3 ± 0.5</td>
<td>9.0 ± 0.9*</td>
<td>3.6 ± 0.3</td>
</tr>
<tr>
<td>0.1 µM</td>
<td>7.3 ± 0.4</td>
<td>7.7 ± 0.4*</td>
<td>3.5 ± 0.3</td>
</tr>
<tr>
<td>0.01 µM</td>
<td>7.1 ± 0.4</td>
<td>7.2 ± 0.4</td>
<td>3.6 ± 0.2</td>
</tr>
<tr>
<td>KCl (anterograde)</td>
<td>50 mM</td>
<td>8.5 ± 0.4</td>
<td>20.3 ± 2.2*</td>
</tr>
<tr>
<td>KCl (retrograde)</td>
<td>50 mM</td>
<td>1.0 ± 0.1</td>
<td>-0.1 ± 0.2*</td>
</tr>
<tr>
<td></td>
<td>25 mM</td>
<td>1.0 ± 0.1</td>
<td>1.0 ± 0.1</td>
</tr>
<tr>
<td></td>
<td>10 mM</td>
<td>1.1 ± 0.1</td>
<td>1.2 ± 0.1</td>
</tr>
</tbody>
</table>

Values are means ± SE; n = 5 animals for all drugs and doses. Ppv, Pdo, and Phv: portal venous, double-occlusion, and hepatic venous pressure, respectively. *P < 0.05 vs. baseline.
Fig. 2. Peak changes in portal (Rpv) and hepatic (Rhv) venous resistances, hepatic venous-to-total hepatic venous resistance ratio (Rhv/Rtot), and liver weight at concentrations of 0.01–100 µM norepinephrine as expressed by percentage of baseline. *P < 0.05 from baseline.

Fig. 3. Peak changes in Rpv, Rhv, Rhv/Rtot, and liver weight at concentrations of 0.01–100 µM histamine as expressed by percentage of baseline. *P < 0.05 from baseline.
wt at 100 µM histamine. The bile flow rate was 0.05 ± 0.01 g·min−1·100 g liver wt−1 (n = 10) at baseline and was transiently decreased concomitantly with liver weight loss after injection of norepinephrine or histamine. Table 2 shows the summary data of changes in bile flow. Norepinephrine reduced the bile flow dose dependently to 63 ± 11% of baseline at 0.1 µM, 56 ± 10% at 1 µM, 21 ± 11% at 10 µM, and 11 ± 5% at 100 µM. Histamine also decreased the bile flow to 56 ± 16% of baseline at 100 µM.

Effects of KCl on hepatic hemodynamics, liver weight, and bile flow. We studied the effect of KCl on hepatic vascular resistance distribution to determine the distribution of effective smooth muscle by directly stimulating vascular smooth muscle with KCl. Figure 4 shows a representative recording of the hepatic vascular response to 50 mM KCl in an anterogradely perfused liver. KCl caused presinusoidal constriction, as characterized by an increase in Ppv without changes in Pdo. This KCl-induced vasoconstriction was accompanied by decreases in liver weight and bile flow rate. This response was similar to that to histamine and norepinephrine. Figure 5 shows representative recordings of a retrogradely perfused liver injected with 10, 25, and 50 mM KCl. At low concentrations of 10 and 25 mM, KCl did not change either Ppv or Pdo but produced transient liver weight loss. An injection of the same volume of saline did not cause any changes in liver weight or pressures (data not shown). At the highest concentration of 50 mM KCl, Pdo was increased along with an increase in the inflow pressure of Phv during the retrograde perfusion. Liver weight showed a biphasic response, as characterized by an initial liver weight loss followed by a progressive weight gain.

As shown in Table 1, after 50 mM KCl in anterogradely perfused livers, Ppv was increased from baseline of 8.5 ± 0.4 mmHg to the peak values of 20.3 ± 2.2 mmHg, but Pdo did not significantly change (3.9 ± 0.2 vs. 4.0 ± 0.3 mmHg, baseline vs. after KCl). In contrast, in retrogradely perfused livers, 50 mM KCl caused significant increases in Phv (the inflow pressure) from the baseline of 8.3 ± 0.4 to 22.4 ± 2.0 mmHg and in Pdo from the baseline of 4.8 ± 0.1 to 16.7 ± 2.0 mmHg. In this retrograde perfusion KCl group, neither 10 nor 25 mM KCl caused no significant increases in Phv, Ppv, or Pdo.

Figure 6 shows the summary data for Rpv, Rhv, Rhv/Rtot ratio, and liver weight in both the KCl and retrograde perfusion KCl groups. In anterogradely perfused livers injected with KCl, Rpv was increased but Rhv did not change significantly, with a resultant decrease in the Rhv/Rtot ratio. In retrogradely perfused livers, 10 and 25 mM KCl did not significantly increase either Rpv or Rhv. However, at the highest concentration of 50 mM KCl, Rhv was significantly increased by 85% of baseline, although Rpv was increased by a much greater magnitude to 500% of baseline. Figure 6 (bottom right) shows the response of liver weight to KCl. In retrogradely perfused livers, a dose-dependent liver weight loss was observed within an initial 1 min, but the subsequent weight gain occurred only at the highest dose of 50 mM KCl. In anterogradely perfused liver injected with 50 mM KCl, liver weight progressively decreased. As shown in Table 2, in response to 50 mM KCl, the bile flow rate decreased to 22 ± 14% of baseline in anterogradely perfused livers and to 73 ± 17% in retrogradely perfused livers.

### Table 2. Bile flow rate after injection of norepinephrine, histamine, or KCl in isolated perfused rabbit livers

<table>
<thead>
<tr>
<th>Drug</th>
<th>Norepinephrine</th>
<th>Histamine</th>
<th>KCl (Anterograde)</th>
<th>KCl (Retrograde)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dose After</td>
<td>Dose After</td>
<td>Dose After</td>
<td>Dose After</td>
<td>Dose After</td>
</tr>
<tr>
<td>100 µM</td>
<td>11 ± 5*</td>
<td>100 µM</td>
<td>56 ± 16*</td>
<td>50 mM 22 ± 14*</td>
</tr>
<tr>
<td>10 µM</td>
<td>21 ± 11*</td>
<td>30 µM</td>
<td>56 ± 16*</td>
<td>50 mM 38 ± 12*</td>
</tr>
<tr>
<td>1 µM</td>
<td>56 ± 10*</td>
<td>10 µM</td>
<td>74 ± 9*</td>
<td>25 mM 40 ± 6*</td>
</tr>
<tr>
<td>0.1 µM</td>
<td>63 ± 11*</td>
<td>1 µM</td>
<td>95 ± 5</td>
<td>10 mM 60 ± 7*</td>
</tr>
<tr>
<td>0.01 µM</td>
<td>98 ± 4</td>
<td>0.1 µM</td>
<td>95 ± 5</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.01 µM</td>
<td>95 ± 5</td>
<td></td>
</tr>
</tbody>
</table>

Values are means ± SE (%baseline); n = 5 animals for all drugs and doses. *P < 0.05 vs. baseline (100%).

![Fig. 4. Representative recording of response to injection of KCl (50 mM) in anterogradely perfused rabbit liver.](image)
DISCUSSION

Hepatic vasoconstriction sites in dog livers differ depending on vasoactive substances (12, 16, 20, 22, 23). In fact, histamine predominantly contracts the canine hepatic vein (12, 16, 20, 22), whereas norepinephrine primarily contracts the presinusoidal vessels rather than the hepatic vein (3, 19, 20). There could be species differences in hepatic vascular responsiveness. In the present study, we determined effects of histamine and norepinephrine on hepatic vascular resistance.

![Fig. 5. Representative recording of response to injection of KCl (10, 25, and 50 mM; left, middle, and right, respectively) in retrogradely perfused rabbit liver.](image)

![Fig. 6. Peak changes in $R_{pv}$, $R_{hv}$, $R_{hv}/R_{tot}$, and liver weight after injection of KCl (10, 25, and 50 mM) with retrograde perfusion (open bars) and 50 mM KCl injection with anterograde perfusion (solid bars). In liver weight gain panel (bottom right), left column shows initial 1-min change and right column the maximal change 6–10 min after injection. *$P < 0.05$ from baseline.](image)
tion in rabbit livers. Vasoreactivity to vasoactive substances will depend on the receptor density, the ability to transmit the stimulus message, and the quantity of effective smooth muscle at the effective site. Thus the role of functional vascular smooth muscle in vasoconstrictive responses was estimated by measuring the longitudinal distribution of the resistance increase induced by KCl. In these experiments, Rtot was assigned to Rpv and Rhv by measuring Pdo as hepatic capillary pressure. A bolus injection of either histamine or norepinephrine caused a dose-dependent increase in Rpv, but no changes in Rhv. KCl (50 mM), when injected portally, increased Rpv almost exclusively, whereas the same dose of KCl injected into the hepatic vein in retrogradely perfused livers caused small but definite hepatic venous constriction concomitant with marked portal contraction. This finding suggests that functional smooth muscle may exist in much greater quantity in the presinusoidal vessels than in the hepatic veins in rabbit livers. Thus the present results suggest that histamine and norepinephrine selectively contract the presinusoidal vessels, which may be ascribed, at least in part, to predominant distribution of functionally active vascular smooth muscle in the presinusoidal vessels rather than the hepatic veins.

Our laboratory has recently shown, by measuring the capillary pressure with the use of the triple-vascular-occlusion method (20) and the double-occlusion method (25) in isolated canine livers, that postsinusoidal vascular resistance comprises approximately one-half of total liver vascular resistance. In the subsequent study, we have shown, using the same vascular occlusion method, that 59% of Rtot exists in the presinusoidal vessels and 41% in the hepatic vein at resting state of isolated rabbit liver. This finding was confirmed by the present study and is consistent with the results obtained by using a micropipette servo-null pressure measurement technique in vivo rabbit liver (3, 14, 15, 19). Thus these results suggest that the portal venule and presinusoidal vasculature are the dominant resistant sites in rabbit livers. On the basis of the present results in both the KCl and the retrograde perfusion KCl groups, it is suggested that vascular smooth muscle may be more abundantly distributed in the portal than the hepatic veins, which may explain in part why the presinusoidal vessels provide a predominant resistance in rabbit livers.

In the present study, the dose of histamine required to produce significant hepatic vasoconstriction was 1 µM. This dose of histamine (1 µM) is identical to that used to contract the isolated rabbit portal vein (4, 10). The responsiveness of rabbit hepatic vessels to histamine seems to be weak compared with that of norepinephrine, because the lower concentration of 0.1 µM norepinephrine can induce significant hepatic vasoconstriction, as shown in Fig. 2. In addition, 100 µM histamine increased Rtot only two times baseline, whereas with the same dose of norepinephrine the increase was four times baseline.

We have clearly shown that histamine-induced hepatic vasoconstriction occurs almost exclusively in the presinusoidal vessels in rabbit livers. This result contrasts with the well-established concept that histamine predominantly contracts the hepatic vein rather than the presinusoidal vessels in dogs (12, 16). We believe that part of the explanation lies in species differences in the distribution of functional smooth muscle in the hepatic vessels. However, differences in receptor distribution, density, and affinity between these two species certainly have a role in the response to histamine. Indeed, the histamine-induced hepatic venoconstriction is not observed in the cat (6) or rat (9). On the other hand, Rothe and Maass-Moreno (19) have recently reported, measuring the hepatic venular pressure with micropipettes in vivo anesthetized rabbits, that histamine at the portal venular blood concentration of 11 µmol/l significantly increases hepatic venular pressure and outflow resistance. They also estimated changes in liver volume by recording changes in lobe thickness, using a variable differential transformer, and showed that histamine passively induces hepatic engorgement (19). This finding is not in agreement with the present result. The reason for this discrepancy is not known, but the different preparation might account for it. Indeed, in isolated portally and hepatic arterially perfused rabbit liver, histamine at a concentration higher than 1 µM increases Rpv and hepatic arterial resistance dose dependently but did not increase liver weight at all (17). However, Greenway and Lautt (7) asserted that the isolated perfused rat livers are unsuitable for studying hepatic hemodynamics with respect to distribution of flows and O2 supply. The limitations of using isolated perfused rabbit livers might be similar to those of the isolated perfused rat livers.

It is well known that norepinephrine causes a reduction in liver blood volume in cats (6), dogs (2, 20), and rabbits (19). In addition, Rothe and colleagues, using the micropipette servo-null pressure measurement technique, have recently demonstrated that the increase in Rpv is much more than that of Rhv in dogs (3), rats (3), and rabbits during norepinephrine infusion (19). We confirmed that a similar response to norepinephrine occurs in isolated rabbit livers. Furthermore, this norepinephrine-induced increase in hepatic vascular resistance is caused almost exclusively by portal venular constriction.

In the present study, a decrease in liver weight accompanied selective presinusoidal vessel constriction when histamine, norepinephrine, or KCl was injected in the anterograde perfused livers. The mechanism for this decrease in liver weight cannot be presently clarified. A reduction in unstressed volume of the hepatic venules or even sinusoids will explain the reduction in volume induced by norepinephrine (7, 18). If the area where this vasoconstriction occurs has a very small total resistance to flow, then even a large change in this minute resistance will be negligible with respect to total resistance. This active constriction and also Ito cell contraction might contribute to the present blood volume reduction. Another possibility may be related to possible heterogeneous portal venule constric-
tion. If there was heterogeneity in portal venule constriction among the hepatic lobules, that is, some vessels were closed and others open, the blood volume of sinusoids, which was distal to the closed portal venules, could be passively reduced due to a decrease in distending pressure of the sinusoids. Actually, in rat liver treated with norepinephrine, the heterogeneous vasoconstriction was observed, as revealed by clear heterogeneous dye staining of the liver, when trypan blue dye was infused after norepinephrine infusion (1, 13).

On the other hand, in retrogradely perfused livers, KCl also caused a dose-dependent initial weight loss. This could possibly be explained by vasoconstriction of the inflow vessel of the hepatic vein by using retrograde perfusion, which did not affect so much the calculated hepatic vascular resistance. At the highest dose of 50 mM KCl, the initial weight loss was followed by a progressive weight gain. When sufficient vasoconstriction downstream from the compliant microvasculature might overcome the effects of possible constriction of the inflow vessel of the hepatic vein, the weight begins to increase because of vascular filling and filtration.

We found that the bile flow decreased after injection of either norepinephrine, histamine, or KCl, with elevation of PPv. The mechanism for this cholestasis is not known from the present study. Lenzen et al. (13) proposed that cholestasis caused by norepinephrine is secondary to its hemodynamic effects, on the basis of the finding that papaverine, an unspecific vasodilator, prevented norepinephrine-induced cholestasis and hepatic vasoconstriction in isolated rat livers. Similar findings have been observed, in that administration of vasodilatory agents such as a nitric oxide donor and a prostaglandin I₂ analog restored the decreased biliary flow rate induced by a potent vasoconstrictor of endothelin-1 to normal values, along with the recovery of PPv (11, 21).

In conclusion, we determined, measuring the hepatic capillary pressure with the double-occlusion method, the effects of histamine, norepinephrine, and KCl on hepatic vascular resistance distribution in isolated rabbit livers perfused with 5% albumin-Krebs solution. Histamine or norepinephrine caused a dose-dependent increase in presinusoidal vascular resistance but no changes in Rhv. There was also a decrease in liver weight. This selective constriction of the presinusoidal vessels seems to be due to the predominant distribution of functionally active vascular smooth muscle in the presinusoidal vessels rather than the hepatic vein. This assumption was based on the observation that KCl contracted the presinusoidal vessels selectively in the anterogradely perfused liver, although KCl significantly, but minimally, increased the Rhv in retrogradely perfused livers. It also should be mentioned that the present results were based on sinusoidal pressure, which was analyzed as part of the pressure profile during the double-occlusion maneuver, that is, an indirect measure. In the future, further studies might be required to compare double-occlusion pressure with microvascular pressures measured directly by micropipettes (3, 14, 19) in the same isolated liver preparation, as was performed in isolated lungs (8).

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