Vagal cooling and concomitant portal norepinephrine infusion do not reduce net hepatic glucose uptake in conscious dogs

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Neurophysiological data provide evidence for CNS involvement in the portal signal. The afferent firing rate in the hepatic branch of the vagus nerve is inversely related to the portal vein glucose concentration (27). Similarly, glucose-sensitive neurons in the lateral hypothalamus decrease their firing rates when glucose is injected in the portal vein, a phenomenon that is mediated by noradrenergic mechanisms (41). Infusion of glucose in the portal vein was also associated with a decrease in the firing rate of the hepatic branch of the splanchnic (sympathetic) nerve (32) and an increase in the firing rate of the pancreatic branch of the vagus nerve (25). To our knowledge, the efferent firing rate in the hepatic branch of the vagus nerve has not been assessed during portal glucose administration, but efferent hepatic vagal activity is enhanced by hyperglycemia brought about by intravenous glucose injection (33).

Therefore, we hypothesized that the portal signal brings about its effect as a result of suppression of afferent firing in the hepatic branch of the vagus nerves, with a resulting enhancement of efferent hepatic parasympathetic activity and an inhibition of the efferent firing rate in the hepatic branch of the splanchnic (sympathetic) nerve (32). Parasympathetic stimulation enhances hepatic glucose uptake, glycogen synthase activity, and glycogen storage in the perfused liver (40), and the portal signal brings about all of these effects in vivo (34). Thus we initially postulated that the effects of the parasympathetic nervous system are dominant. To examine the role of the parasympathetic nervous system, we cooled the vagus nerves of conscious dogs during portal glucose delivery, thus interrupting the efferent signal. Cooling would also interrupt afferent transmission, but this should be equivalent to a maximal stimulus, since portal glucose delivery induces a fall in the firing rate of the vagal afferents (27). When an initial series of studies indicated that vagal cooling did not suppress NHGU, we expanded the experimental design to examine the role of sympathetic signaling in bringing about the effects of the portal signal. In other words, we postulated that the suppression of sympathetic input to the liver during portal glucose administration removes a tonic block to NHGU. To mimic augmentation of sympathetic tone to the liver, we infused norepinephrine (NE) in the hepatic portal circulation at a low rate during the cooling period in a second group of dogs.

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MATERIALS AND METHODS

Animal Care

Experiments were conducted on conscious, mongrel dogs of either sex weighing 23.1 ± 0.8 kg. The animals were fed meat and chow one time daily (KaliKan, Vernon, CA and Purina Lab Canine Diet No. 5006, St. Louis, MO; 34% protein, 46% carbohydrate, 14.5% fat, and 5.5% fiber based on dry weight). The animals were housed in a facility that met the standards of the American Association for the Accreditation of Laboratory Animal Care International, and the protocols were approved by the Institutional Animal Care and Use Committee.

Surgical Procedures

Before the experiment (2 wk), all dogs underwent a laparotomy under general anesthesia, and infusion catheters were placed in a splenic and a jejunal vein for intraportal infusions. Catheters for blood sampling were inserted in the left common hepatic vein, the portal vein, and the femoral artery, as previously described (23). Doppler flow probes (group 1, n = 3; Instrumentation Laboratories, Baylor University, Houston, TX) or ultrasonic flow probes (group 2, n = 6; Transonic Systems, Ithaca, NY) were placed around the hepatic artery and the portal vein to determine hepatic blood flow (HBF). The flow probe leads and the catheters were placed in subcutaneous pockets before closure of the abdominal skin. Stainless steel cooling coils, probe leads and the catheters were placed in subcutaneous pockets and the portal vein to determine hepatic blood flow (HBF). The flow probe leads and the catheters were placed in subcutaneous pockets before closure of the abdominal skin. Stainless steel cooling coils, probe leads and the catheters were placed in subcutaneous pockets and the portal vein to determine hepatic blood flow (HBF).

Portal glucose infusate so that the PAH delivery rate would be 0.4 mg·kg⁻¹·min⁻¹ was administered to assess mixing of the infused glucose with blood in the portal and hepatic veins, as described previously (34). In addition, a primed continuous peripheral infusion of 50% dextrose was begun so that the hepatic glucose load could be quickly clamped at twofold basal. The experimental period was divided into a 90-min period during which the vagal coils were infused with solution at body temperature (37°C; 0–90 min, period 1), a 60-min period (90–150 min, period 2) of vagal cooling (−15°C in the bath), and a subsequent period (150–210 min, period 3) when the coils were again perfused with a body temperature solution. For group 2, study conditions were identical to those in group 1, with three exceptions. First, the glucagon infusion during the entire experimental period was 0.5 ng·kg⁻¹·min⁻¹ because the infusion rate in group 1 appeared to result in concentrations slightly above basal for 42-h-fasted dogs. This might have tended to reduce NHGU in group 1, but it should not have affected the overall conclusion (16), since the same rate was used during all three periods. Second, periods 2 and 3 were lengthened to 90 min (i.e., period 2 = 90–180 min and period 3 = 180–270 min) to make them equivalent in length to period 1. Third, simultaneously with vagal cooling during period 2, NE was infused intraportally at 16 ng·kg⁻¹·min⁻¹ to augment hepatic sympathetic tone; the NE infusion was stopped concomitantly with the cessation of vagal cooling at 180 min.

The effectiveness of the cooling-induced blockade of parasympathetic signaling was verified by measuring the heart rate (primarily under vagal control in the basal state and during normal fluctuations of blood pressure; see Refs. 21 and 38), verifying a decrease in rate and depth of respirations, and observing bilateral Horner’s syndrome (8).

Processing and Analysis of Samples

Plasma glucose, insulin, glucagon, NE, and cortisol and blood lactate were measured as described elsewhere (7, 34). PAH concentrations were also measured in perchloric acid-deproteinized blood, as previously described (34).

Calculations

Mixing of the infused glucose with the portal vein blood was assessed by comparing the recovery of PAH (which was mixed with the portal glucose infusate) in the portal and hepatic veins with the PAH infusion rate (23, 34). An experiment was deleted from the database if poor mixing, as defined previously (34), was observed at more than one of the four time points in the portal glucose infusate phase. In this study, 15 dogs were used; six were not included because of catheter failure or poor mixing. In the animals that were retained, the ratio of PAH recovery in the portal vein to the PAH infusion rate was 1.1 ± 0.2; whereas the ratio of PAH recovery in the hepatic vein to the PAH infusion rate was 0.9 ± 0.2 (a ratio of 1.0 would represent perfect mixing). When a dog was retained in the database, all of the points were used whether they were mixed or not, because mixing errors occur randomly.

HBF was calculated by two methods, Doppler or ultrasonic flow probes and dye extraction (34). The results obtained with flow probes and ICG extraction were not significantly different, but the data shown are those obtained with the flow probes, because their use did not require an assumption regarding the distribution of arterial and portal contributions to HBF.

The rate of substrate delivery to the liver, or hepatic substrate load; net hepatic substrate balance; net hepatic fractional glucose extraction; and nonhepatic glucose uptake were calculated as described previously (37). Glucose data were calculated with both direct and indirect methods (37). There were no significant differences with the results of the two methods of calculation, but the results reported here utilize the indirect calculation, unless specified otherwise, to minimize any error resulting from incomplete mixing of the glucose infusate in the portal circulation. Plasma glucose values were converted to whole

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mg·kg⁻¹·min⁻¹).
blood glucose values for all calculations using correction factors derived from previous studies in our laboratory (17, 18, 34). Hepatic sinusoidal insulin and glucagon concentrations (37) and hepatic NE spillover (9) were calculated as previously described. Arterial cortisol concentrations and portal vein concentrations of insulin, glucagon, and peripheral glucose were infused throughout the 3 experimental periods (periods 1–3). During period 2, the vagus nerve was cooled. Period 3 followed the discontinuance of vagal cooling.

Hormone concentrations. Insulin concentrations increased statistically threefold between the basal and experimental period, but there were no significant changes over time in glucagon and NE. Moreover, the concentrations for insulin, glucagon, and NE did not differ among periods 1–3 (Table 1).

Glucose and lactate metabolism. Arterial blood glucose did not differ among periods 1–3, averaging approximately twofold basal (Table 2). The arterial-portal vein glucose gradient was negative during the experimental period, and did not differ significantly among periods 1–3. The hepatic glucose load increased approximately threefold during periods 1–3 and did not differ among these periods. The dogs switched from net hepatic glucose output to NHGU with the onset of the experimental period, with no significant difference in rates among periods 1–3 (Table 2). Net hepatic fractional extraction of glucose did not differ significantly among periods 1–3 (0.07 ± 0.01, 0.10 ± 0.02, and 0.10 ± 0.03, respectively). The total glucose infusion rates and nonhepatic glucose uptakes were statistically similar among periods 1–3.

Arterial blood lactate concentrations increased over basal during periods 1–3 but did not differ among the three periods. The livers of the animals shifted from net lactate uptake to net output during the experimental period. Net hepatic lactate output was greatest during period 1, but it did not differ significantly between periods 2 and 3.

Group 2 (Vagal Cooling Plus NE Infusion)

Heart rate and HBF. Blockade of vagal firing was evident based on the significant (nearly 3-fold) increase in heart rate during period 2 (Table 3). As in group 1, all dogs evidenced prolapose of the third eyelid (Horner’s sign) and a deepening and slowing of respirations during period 2, which was re-
versed during period 3. Hepatic artery blood flow increased significantly and portal vein blood flow decreased significantly after the basal period, concomitant with the initiation of the somatostatin infusion. Neither hepatic artery nor portal vein blood flow changed significantly after that time (Table 3).

Hormone concentrations. Arterial and hepatic sinusoidal plasma insulin concentrations were increased three- to fourfold above basal during periods 1–3, respectively [Table 4; not significant (NS) among experimental periods]. Arterial and sinusoidal plasma glucagon concentrations remained at basal throughout the experiments (Table 4).

There were no significant differences in arterial plasma NE levels among the test periods (Table 4). NE infusion in the portal vein increased the portal vein plasma NE level so that the sinusoidal concentration rose fivefold during period 2, but the concentration fell below basal during period 3. Hepatic NE spillover increased ~60% during NE infusion and then returned to basal after cessation of the infusion. As shown in our previous work (20), vagal cooling created a threefold increase in the arterial plasma cortisol level; the cortisol levels decreased with the cessation of vagal cooling.

Glucose metabolism. Arterial blood glucose increased approximately twofold, from 75 ± 2 mg/dl in the basal period to 170 ± 3, 167 ± 2, and 162 ± 2 mg/dl during the last 30 min of periods 1, 2, and 3, respectively (NS among experimental periods; Fig. 1). Because of the infusion of glucose in the portal vein during the experimental periods, the arterial-portal difference shifted from a positive to a negative value in periods 1, 2, and 3 (NS among periods 1–3; Table 5).

The hepatic glucose load increased approximately twofold above basal in periods 1, 2, and 3, respectively, and did not differ among the periods (Fig. 1).

In the basal state, net hepatic glucose output was 1.9 ± 0.3 mg·kg⁻¹·min⁻¹ (Fig. 1). The combination of increases in the hepatic glucose load and the insulin level as well as the presence of the portal signal caused the liver to exhibit a net glucose uptake of 5.0 ± 0.9 mg·kg⁻¹·min⁻¹ (using the indirect calculation) in period 1. Interruption of vagal firing by hepatic portal delivery of NE did not decrease NHGU (5.6 ± 0.7 mg·kg⁻¹·min⁻¹). Likewise, there was no significant change in NHGU when vagal cooling and NE infusion were stopped (6.1 ± 0.9 mg·kg⁻¹·min⁻¹). The values for NHGU using the direct calculation were 4.4 ± 1.2, 4.2 ± 0.7, and 5.6 ± 0.8 mg·kg⁻¹·min⁻¹ during periods 1–3, respectively (NS compared with the values from the indirect calculation). The fractional extraction of glucose by the liver was also unaffected by vagal cooling (0.08 ± 0.01, 0.11 ± 0.02, and 0.11 ± 0.03 in periods 1, 2, and 3, respectively, Fig. 1).

Glucose EndoRa declined from 2.5 ± 0.2 mg·kg⁻¹·min⁻¹ in the basal period to 0.5 ± 0.3 mg·kg⁻¹·min⁻¹ under the influence of hyperglycemia, hyperinsulinemia, and the portal signal. There were no statistical differences in EndoRa during

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**Table 4. Plasma hormone data in group 2**

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Basal Period</th>
<th>Period 1</th>
<th>Period 2</th>
<th>Period 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>Insulin, μU/ml</td>
<td>5 ± 1</td>
<td>17 ± 5</td>
<td>22 ± 5</td>
<td>21 ± 4</td>
</tr>
<tr>
<td>Arterial</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Glucagon, pg/ml</td>
<td>18 ± 5</td>
<td>47 ± 11</td>
<td>48 ± 7</td>
<td>58 ± 11</td>
</tr>
<tr>
<td>Arterial</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hepatic sinusoidal</td>
<td>30 ± 3</td>
<td>31 ± 2</td>
<td>32 ± 2</td>
<td>28 ± 2</td>
</tr>
<tr>
<td>Glucagon, pg/ml</td>
<td>38 ± 3</td>
<td>41 ± 2</td>
<td>39 ± 2</td>
<td>35 ± 2</td>
</tr>
<tr>
<td>Arterial</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Noradrenaline, pg/ml</td>
<td>196 ± 17</td>
<td>194 ± 19</td>
<td>157 ± 25</td>
<td>151 ± 18</td>
</tr>
<tr>
<td>Heparic sinusoidal</td>
<td>210 ± 27</td>
<td>184 ± 20</td>
<td>993 ± 174*</td>
<td>148 ± 15*</td>
</tr>
<tr>
<td>Noradrenaline spillover, hepatic</td>
<td>0.6 ± 0.2</td>
<td>0.8 ± 0.3</td>
<td>1.3 ± 0.4</td>
<td>1.0 ± 0.3</td>
</tr>
<tr>
<td>Cortisol, arterial, μg/dl</td>
<td>5.9 ± 1.2</td>
<td>1.3 ± 0.2</td>
<td>3.9 ± 0.8</td>
<td>1.0 ± 0.2</td>
</tr>
</tbody>
</table>

Data are means ± SE; n = 6 dogs. Somatostatin, intraportal insulin, glucagon, and glucose, and peripheral glucose were infused throughout the 3 experimental periods (periods 1–3). During period 2, the vagus nerve was cooled and noradrenaline was infused intraportally. Period 3 followed the discontinuance of vagal cooling. Cortisol was not measured in the portal vein. P < 0.05 vs. all other periods (*), vs. basal period and period 1 (**), and vs. periods 1 and 3 (†).
periods 1–3. Glucose R₄ showed a tendency to increase progressively during periods 1–3 (13.5 ± 2.8, 16.3 ± 3.7, and 17.8 ± 5.6 mg·kg⁻¹·min⁻¹, respectively), but there was no statistically significant change over time (P = 0.2). Moreover, nonhepatic glucose uptake did not change significantly during periods 1–3 (Table 5).

Lactate metabolism. The arterial blood lactate level rose approximately threefold above basal during periods 1–3 and did not differ among the periods (Table 5). The liver switched from net hepatic lactate uptake to output at the onset of the experimental periods, and the rate did not differ among the periods.

DISCUSSION

Group 1 (Vagal Cooling Without NE Infusion)

In the current studies, NHGU was initially stimulated by hyperinsulinemia, hyperglycemia, and the presence of the portal signal. When vagus nerve activity was then blocked by cooling (group 1), NHGU was not inhibited. Although group 1 was composed of only three dogs, there appears to be little possibility of a type II error. NHGU increased modestly, rather than decreased, between periods 1 and 2 in every animal and did not increase further during period 3.

If the portal signal is transmitted to the CNS via a suppression of afferent firing in the hepatic vagus, as neurophysiological data strongly suggest (29–31), then blocking the afferent signaling by vagal cooling should have created a maximal stimulus. Thus our data suggest that efferent parasympathetic signaling is not required for enhancement of NHGU in response to the portal signal. However, there are possible alternatives to this conclusion. First, the efferent signal could be transmitted via vagal parasympathetic fibers not blocked by cooling the nerve. This possibility seems unlikely, given that the heart rate increased significantly during cooling. Horner’s sign was present, and respirations changed as expected. We have previously shown (20) that vagal cooling bath temperatures of −13 to −15°C produce temperatures of −2°C at the nerve, which were sufficient to halt vagal signaling in the dog (8) and cat (13). Injection of atropine (known to maximally stimulate heart rate) is not able to produce an additional increase in heart rate over cooling at this temperature (20). Second, the stimulation of NHGU could result from a local parasympathetic reflex within the hepatoporal region, without evoking a centrally mediated response. A local cholinergic reflex was suggested by work in the isolated, perfused rat liver (43). However, the existence of an intrahepatic reflex does not rule out the possibility that a centrally mediated response forms a component of the portal signal. That complete surgical denervation of the liver blunted NHGU during portal glucose infusion in the dog (2) is consistent with central mediation of the portal signal. Additionally, the ability of the portal signal to enhance insulin secretion (12) is apparently mediated by reflex stimulation of the firing rate in the pancreatic branch of the vagus nerve (25, 46, 47), and this stimulation is prevented by sectioning the hepatic branch of the vagus (25, 47), consistent with central control. Thus it is possible that both local and centrally mediated mechanisms could work together to bring about the full response to the portal signal. Finally, there is the possibility that cooling the vagus produced offsetting signals by halting both parasympathetic and sympathetic signaling. As mentioned previously, portal glucose delivery results in a fall in the efferent firing rate in the hepatic branch of the splanchnic (sympathetic) nerve, and the fall is prevented in animals that undergo sectioning of the hepatic branch of the vagus nerve (32). Thus it appears that a fall in afferent parasympathetic signaling in response to entry of glucose in the portal vein results in an enhancement of efferent parasympathetic firing and a concomitant suppression of hepatic sympathetic tone, with the two efferent signals cooperating to enhance NHGU. Vagal cooling completely halts the firing in the nerve (13); although afferent firing diminishes in an inverse relationship with the portal vein glucose concentration, it apparently never ceases altogether (29). Thus we were concerned that we might have induced a greater suppression of sympathetic input to the liver than normally occurs, offsetting any impact of the reduction in parasympathetic efferent signaling on NHGU. Therefore, we conducted further studies in which study conditions were essentially the same as those in the first group, except that a low-dose intraportal infusion of NE, designed to target the liver without raising systemic NE concentrations, was used to simulate maintenance of hepatic sympathetic tone.

Group 2 (Vagal Cooling Plus NE Infusion)

The combination of vagal cooling and low-dose intraportal NE infusion in the presence of the portal signal also had no suppressive effect on NHGU. Although this suggests that our revised hypothesis was incorrect, it does not completely disprove it. Infusion of NE in the portal vein might not have adequately simulated maintenance of sympathetic signaling to the liver. First, infusion of NE in the blood may not have positioned it to contact receptors located in the hepatic parenchyma of the dog liver (3). However, portal vein NE administration in the isolated, perfused rat liver closely mimics the effects of hepatic nerve stimulation on glucose and lactate output (14, 44). Moreover, both in the sheep (4) and the dog

Table 5. A-P blood glucose gradient, glucose infusion rate, nonhepatic glucose uptake, and lactate data in group 2

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Basal Period</th>
<th>Period 1</th>
<th>Period 2</th>
<th>Period 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>A-P glucose gradient, mg/dl</td>
<td>1.5±0.3</td>
<td>−12.9±1.3</td>
<td>−10.9±0.5</td>
<td>−13.4±2.1</td>
</tr>
<tr>
<td>Glucose infusion rate, mg·kg⁻¹·min⁻¹</td>
<td>0</td>
<td>13.3±1.3</td>
<td>16.4±2.2</td>
<td>17.7±2.6</td>
</tr>
<tr>
<td>Nonhepatic glucose uptake, mg·kg⁻¹·min⁻¹</td>
<td>1.9±0.3</td>
<td>9.4±1.1</td>
<td>11.1±2.0</td>
<td>12.1±2.4</td>
</tr>
<tr>
<td>Arterial blood lactate, μmol/l</td>
<td>505±58</td>
<td>1,364±129</td>
<td>1,457±122</td>
<td>1,509±107</td>
</tr>
<tr>
<td>Net hepatic lactate balance, μmol/kg·min⁻¹</td>
<td>−5.3±1.1</td>
<td>13.5±1.4</td>
<td>8.1±2.4</td>
<td>10.4±2.1</td>
</tr>
</tbody>
</table>

Data are means ± SE; n = 6 dogs. Somatostatin, intraportal insulin and glucagon, intraportal glucose, and peripheral glucose were infused throughout the 3 experimental periods (periods 1–3). During period 2, the vagus nerve was cooled, and norepinephrine was infused intraportally. Period 3 followed the discontinuance of vagal cooling. Negative value for hepatic balance indicates net hepatic uptake. There were no significant differences among periods 1–3.
(7), intraportal NE infusion has been used in studying the effects of a selective rise in sinusoidal NE on hepatic glucose production. Second, this rate may not have been sufficient to mimic normal sympathetic tone. In a previous study, hepatic NE spillover fell modestly but significantly, from 1.6 ± 0.4 to 1.1 ± 0.3 ng·kg⁻¹·min⁻¹, during portal glucose delivery (19), and we chose an NE infusion rate calculated to maintain but not exceed a basal rate of spillover. In this we were successful. The rate of hepatic NE spillover evident in this study, 1.3 ± 0.4 ng·kg⁻¹·min⁻¹, is virtually identical to that observed in normal dogs studied in the basal state and during moderate-intensity exercise (10) and is substantially less than that observed in the dog during heavy exercise (9). Higher levels of intraportal NE infusion (50 ng·kg⁻¹·min⁻¹), elevating the hepatic sinusoidal NE concentration to 4,100 pg/ml) markedly stimulated hepatic glycogenolysis (7), and our infusion rate was chosen to avoid this direct effect on the liver, as well as potential indirect effects resulting from NE action on peripheral tissues. Finally, it is possible that a neurotransmitter other than NE, e.g., serotonin (26, 28), is normally involved in a sympathetic component of the efferent limb of the portal signal. Nevertheless, if the portal signal normally causes a coordinated change in parasympathetic and sympathetic signaling, it is unlikely that blocking part of the efferent signaling (the rise in vagal efferent firing) while leaving the sympathetic limb intact would have resulted in a full response to the portal signal. Certainly, the intraportal infusion of α- and β-adrenergic receptor antagonists did not enhance NHGU during hyperinsulinemia and hyperglycemia (achieved via peripheral glucose infusion) in the conscious dog (42).

Role of Parasympathetic Innervation in Liver Glucose Metabolism

A large body of data indicates that the parasympathetic nervous system is involved in the regulation of NHGU and hepatic glucose metabolism (reviewed in Ref. 5). In vitro evidence is especially convincing. Stimulation of the distal cut end of the vagus nerve increases glycogen synthase activity and glycogen synthesis independent of changes in insulin (39). In a perfused liver preparation under hyperglycemic and hyperinsulinemic conditions, the addition of ACh to the perfusate increased NHGU, whereas the addition of atropine decreased it (43). However, both nerve stimulation and ACh administration may have produced a supra-physiological stimulus. In addition, they represented an isolated parasympathetic stimulus to the liver, and it may be that, when the sympathetic nervous system is functional, it dominates in the regulation of hepatic glucose metabolism.

In vivo data regarding parasympathetic regulation of hepatic glucose metabolism are less abundant than in vitro data. Using conscious dogs, Chap et al. (6) found that NHGU in response to ingestion of a glucose load was reduced 44% by portal infusion of atropine. Atropine also altered the rate of portal blood flow and the absorption of the glucose load, however, raising the question of whether its effects on NHGU might have been indirect rather than a direct effect of muscarinic blockade. Shiota et al. (42) showed that, during a hyperglycemic, hyperinsulinemic clamp in the conscious dog, ACh infusion in the hepatic portal vein caused an increase in NHGU. It is important to note that, in this experiment, the infusion of ACh increased hepatic arterial blood flow and, as a result, the hepatic glucose load. Nevertheless, the fractional extraction of glucose by the liver increased, indicating that hepatic glucose uptake was stimulated even when the increase in the hepatic glucose load was taken into account. It remains possible, however, that the increase in fractional extraction resulted from the same stimulus as that causing the increased HBF. Moreover, both the studies of Chap et al. (6) and Shiota et al. (42) suffer from the fact that it is difficult to be sure that neurotransmitters or receptor blocking agents reach their physiological targets when they are delivered via the bloodstream, as previously noted in regard to the NE infusion in the current study. In regard to surgical interruption of vagal signaling, it is noteworthy that rats with isolated hepatic cholinergic denervation exhibited significantly less hepatic glycogen in the fed state than did sham-operated controls (45). However, the rats were studied 4 wk postoperatively. It is not clear that these findings in a chronically denervated model are relevant in a more acute setting, such as changes in neural firing in response to feeding. Thus, although it seems clear that the parasympathetic nervous system can enhance hepatic glucose uptake and storage in vitro, neither the published literature nor our current findings provide compelling evidence that the hepatic parasympathetic efferent innervation plays a major role in acute regulation of NHGU in vivo.

In conclusion, the present data do not confirm the hypothesis that the portal signal is mediated by a decrease in afferent vagal firing stimulating an increase in parasympathetic efferent stimuli from the CNS to the liver. Intraportal NE infusion also failed to blunt NHGU, but this may have been because it did not adequately simulate maintenance of sympathetic tone. The current data thus suggest that the enhancement of NHGU by the portal signal is mediated by a local intrahepatic reflex or another neurotransmitter. We cannot rule out, however, that suppression of sympathetic stimuli to the liver plays a part in bringing about the portal signal.

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GRANTS

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