Intraportally delivered GLP-1, in the presence of hyperglycemia induced via peripheral glucose infusion, does not change whole body glucose utilization

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After a meal, glucagon-like peptide-1 (GLP-1) is secreted from the L cell in the gut, such that its level in the hepatic portal vein is approximately twice that in peripheral blood (7). Similarly, at the same time, postprandial glucose levels are higher in hepatic portal vein blood than in the artery, due to absorption of the meal. We have previously reported that, in the presence of intraportal glucose delivery, a physiological increase of GLP-1 in the hepatic portal vein increases nonhepatic glucose uptake via a mechanism independent of changes in pancreatic hormone secretion. The aim of the present study was to determine whether intraportal glucose delivery is required to observe this effect. Experiments consisted of a 40-min basal period, followed by a 240-min experimental period, during which conscious 42-h fasted dogs received glucose peripherally to maintain arterial plasma glucose levels at ~160 mg/dl. In addition, either saline (n = 6) or GLP-1 (1 pmol·kg−1·min−1; GLP-1, n = 6) was administered intraportally during the experimental period. As in the previous study, the presence of GLP-1 did not alter pancreatic hormone levels; however, in the present study, intraportal GLP-1 infusion did not result in an increase in whole body glucose utilization. This is despite the fact that arterial and hepatic portal vein GLP-1 levels were maintained at the same level as the previous study. Therefore, a physiological elevation of GLP-1 in the hepatic portal vein does not increase whole body glucose uptake when hyperglycemia is induced by peripheral glucose infusion. This indicates that a physiological increase in GLP-1 augments glucose utilization only when GLP-1 and glucose gradients conditions mimic the postprandial state.

dog; glucagon-like peptide-1; hepatic portal vein; portal signal

AFTER A MEAL, GLUCAGON-LIKE PEPTIDE-1 (GLP-1) is secreted from the L cell in the gut, such that its level in the hepatic portal vein is approximately twice that in peripheral blood (7). Similarly, at the same time, postprandial glucose levels are higher in hepatic portal vein blood than in the artery, due to absorption of the meal. We have previously reported that, in the presence of intraportal glucose delivery, a physiological increase of GLP-1 in the hepatic portal vein increases nonhepatic glucose uptake (Non-HGU) via a mechanism independent of changes in pancreatic hormone secretion (11). This increase in Non-HGU did not occur when GLP-1 was infused at the same rate through the hepatic artery, even though infusion at this site elevated GLP-1 concentrations in the liver and peripheral blood to the same levels as those seen in the presence of intraportal GLP-1 infusion (11). We concluded, therefore, that the elevation of GLP-1 in the hepatic portal vein was itself responsible for the increase in Non-HGU.

In addition to showing that GLP-1 delivery into the hepatic portal vein results in changes in glucose utilization, our laboratory (2, 9, 14, 15, 22) and others (5, 8, 21) have assembled evidence that intraportal delivery of glucose per se alters the distribution of a glucose load in the body. Compared with peripheral glucose delivery, intraportal delivery of glucose results in greater net hepatic glucose uptake, even when the liver is presented with equal hepatic glucose loads and sinusoidal insulin levels (2). In addition, we have shown that intraportal delivery of glucose decreases Non-HGU. Thus portal glucose delivery results in a preferential deposition of glucose in the liver (9, 14, 15). It is believed that these unique effects of portal glucose delivery are mediated by neural signals initiating within the hepatic portal region (1, 12).

Firing of afferent vagal nerve fibers, originating in the hepatic portal region, decreases upon initiation of an intraportal glucose infusion, thus suggesting that the portal glucose signal is neurally mediated (16, 20). In contrast to glucose, intraportal GLP-1 delivery increases neural firing from the region (17, 25); therefore, both glucose and GLP-1 have the ability to initiate effects via neural changes within the hepatic portal vein. It has been suggested that a negative arterial-hepatoportal gradient for both GLP-1 and glucose levels must be present for GLP-1 to exert its effect on glucose utilization (4, 10, 11). The aim of the present study was to test this hypothesis.

RESEARCH DESIGN AND METHODS

Animals and surgical procedures. Mongrel dogs of either sex, with a mean weight of 22.2 ± 0.4 kg, were used. All animals were maintained on a diet, as previously described (11). The animals were housed in a facility that met American Association for Accreditation of Laboratory Animal Care guidelines, and the protocol was approved by the Vanderbilt University Medical Center Animal Care Committee.

Approximately 16 days before experimentation, a laparotomy was performed, as previously described (11). In brief, under general anesthesia, sampling catheters were placed into the hepatic portal vein, hepatic vein, and a femoral artery; catheters to be used for intraportal infusion were inserted into a jejunal vein and a splenic vein; and ultrasonic flow probes were placed around the portal vein and hepatic artery. Approximately 2 days before each study, the animals were shown to be in good health, as previously described (11).

Experimental design. After a 42-h fast, catheters and transonic leads were exteriorized, as previously described (11). The protocol consisted of a 100-min equilibration period (−140 to −40 min), a
using the formula \[GGNa \times \text{PGa}/PFa + GGNp \times \text{PGp}/PFp\], where GGNa and GGNp are the arterial and portal vein plasma glucagon concentrations, respectively; and PFa, PFp, and PFi are hepatic artery, portal vein, and total hepatic plasma flow, respectively.

Statistical analysis. All data are presented as means ± SE. Repeated measures two-way ANOVA was used for time course analysis, with post hoc data analysis determined by Student-Newman-Keuls method. Statistical significance was accepted at \(P < 0.05\).

RESULTS

Plasma glucose levels. Plasma glucose levels in the artery (159 ± 1 and 162 ± 1 mg/dl) and hepatic portal vein (159 ± 1 and 162 ± 1 mg/dl) in the Sal and GLP-1 groups, respectively, were increased similarly in response to peripheral glucose infusion (Fig. 1A).

Plasma GLP-1 levels. There was no difference between groups in basal GLP-1 levels in either arterial or hepatic portal vein plasma (Fig. 1B), nor did the levels change in response to saline infusion. On the other hand, they rose in both the artery (29.4 ± 1.5 pm) and hepatic portal vein (57.7 ± 4.5 pm) in response to intraportal GLP-1 infusion (Fig. 1B).

GIR. There was no difference in the GIRs, required to maintain the glucose clamp, between the two groups (6.1 ± 1.0 vs. 6.4 ± 1.2 mg·kg⁻¹·min⁻¹, average over final 2 h, Sal vs. GLP-1) (Fig. 2).

Plasma insulin and glucagon levels. The arterial and hepatic portal plasma insulin levels were similar between groups during the basal period (Fig. 3A). Both groups exhibited similar increases in response to the hyperglycemia (to 24 ± 1 and 28 ± 5 μU/ml in arterial plasma, and to 81 ± 12 and 86 ± 10 μU/ml in hepatic portal vein plasma in the Sal and GLP-1 groups, respectively) (Fig. 3A). The sinusoidal plasma glucagon levels were similar between groups during the basal period (Fig. 3B) and decreased in a similar manner (to 22 ± 5 and 22 ± 5 pg/ml, Sal and GLP-1, respectively) during the experimental period.

Hepatic blood flow, NHGB, and non-HGU. Hepatic arterial blood flow during the basal period and the experimental period were similar between groups, as were the hepatic portal vein blood flows. In the basal state, net hepatic glucose output was

![Fig. 1. Plasma glucose and glucagon-like peptide-1 (GLP-1) levels in 42-h fasted conscious dogs. A: arterial and portal plasma glucose levels for dogs that received either intraportal GLP-1 or saline (Sal). Levels were basal initially (−40 to 0 min), but both arterial and portal levels increased significantly (\(P < 0.05\)) during the experimental period (0 to 240 min) in response to the glucose clamp. There were no significant differences between groups in either the basal or experimental period. B: arterial and portal plasma GLP-1 levels for dogs that received either intraportal GLP-1 or saline. In the animals that received the GLP-1 infusion, GLP-1 levels were basal initially (−40 to 0 min), but both arterial and portal levels increased significantly (\(P < 0.05\)) during the experimental period (0 to 240 min). Levels remained unchanged in Sal. Values are means ± SE.](#)

![Fig. 2. Total glucose infusion rate (GIR) during the infusion of intraportal GLP-1 or Sal into the hepatic portal vein (0 to 240 min). There was no statistical difference between groups. Values are means ± SE for each time point.](#)
of GLP-1 in the hepatic portal vein stimulated Non-HGU via a mechanism independent of changes in pancreatic hormone secretion (11). The results from the present study indicate that, in the absence of a hepatic portal vein glucose infusion, portal vein GLP-1 infusion does not bring about such an effect. This observation is supported by our observation that there was no significant difference in GIR (6.1 ± 1.0 vs. 6.4 ± 1.2 mg·kg⁻¹·min⁻¹, average over final 2 h, Sal vs. GLP-1, respectively) in the two groups (Fig. 2). The failure of the GIR to rise in the presence of elevated GLP-1 in the absence of portal glucose infusion is explained by the absence of an increase in Non-HGU (4.9 ± 1.0 and 4.9 ± 1.2 mg·kg⁻¹·min⁻¹, final 2 h, Sal and GLP-1, respectively) (Fig. 4B). In our previous study, the presence of a portal glucose infusion GLP-1 delivery significantly increased (3.8 ± 0.7 to 5.5 ± 0.8 mg·kg⁻¹·min⁻¹) Non-HGU (11).

The question thus arises as to the mechanism by which a physiological increase in hepatic portal vein GLP-1 levels similar between groups (1.5 ± 0.2 and 1.3 ± 0.1 mg·kg⁻¹·min⁻¹, Sal and GLP-1, respectively). In response to the hyperglycemic clamp, the liver switched to net glucose uptake in both groups. Although there was no significant difference between groups, there was a tendency for net hepatic glucose uptake to be higher in the GLP-1 group [NHGB = −1.2 ± 0.1 and −1.6 ± 0.2 mg·kg⁻¹·min⁻¹, Sal and GLP-1, respectively, (P = 0.52) during final 2 h] (Fig. 4A). There was no effect of GLP-1 on non-HGU (4.9 ± 1.0 and 4.9 ± 1.2 mg·kg⁻¹·min⁻¹, final 2 h, Sal and GLP-1, respectively) (Fig. 4B).

**DISCUSSION**

After a meal, the increases in both GLP-1 (7) and glucose levels in the hepatic portal vein are significantly greater than in the peripheral blood. It has been suggested that this elevation of both GLP-1 and glucose must exist for a physiological increase of GLP-1 to exert its effect on glucose utilization (4, 10, 11). We have previously reported that, in the presence of hepatic portal vein glucose infusion, a physiological increase

![Fig. 3. Plasma insulin and glucagon levels. A: arterial and portal plasma insulin levels for dogs that received either intraportal GLP-1 or saline. Levels were basal initially (−40 to 0 min), but both arterial and portal levels increased significantly (P < 0.05) during the experimental period (0 to 240 min) in response to the glucose clamp. B: sinusoidal plasma glucagon levels for animals that received either intraportal GLP-1 or saline. There was a significant decrease in sinusoidal glucagon levels in both groups during the experimental period compared with respective basal period values (P < 0.05). Values are means ± SE.](http://ajpendo.physiology.org/)

![Fig. 4. Glucose production and utilization. A: net hepatic balance (NHGB) in animals that received either intraportal GLP-1 or Sal. Rates in each group were significantly decreased (P < 0.05) during the experimental period compared with their respective basal period values. There was no significant difference between groups. B: nonhepatic glucose uptake (Non-HGU) during the infusion of intraportal Sal or GLP-1 during the experimental period (30 to 240 min). There was no significant difference between groups. Data are the average of values over 30 min segments. Values are means ± SE.](http://ajpendo.physiology.org/)
might bring about increased whole body glucose uptake solely in the presence of intraportal glucose delivery. Elevation of portal vein glucose decreases vagal firing by afferent fibers originating in the wall of the hepatic portal vein (16, 20). Portally delivered GLP-1, on the other hand, increases neural firing in afferents originating in the hepatoporal region (17, 25). It has been clearly shown that the portal glucose delivery is associated with a decrease in Non-HGU (9, 14). Thus, if GLP-1 were to override the impact of portally delivered glucose on vagal afferent firing to nonhepatic tissue, one would predict an increase in Non-HGU. Since our laboratory’s earlier work has suggested that the effect of the portal signal occurs in muscle, it seems likely that this is the site of the GLP-1-induced effect (9).

Our laboratory’s earlier data (7) showed that intraportal infusion of GLP-1 (1 pmol·kg⁻¹·min⁻¹) in the presence of hyperglycemia, induced by peripheral glucose infusion, and a pancreatic hormone clamp resulted in a small increase in net hepatic glucose uptake (−0.8 mg·kg⁻¹·min⁻¹), but no change in Non-HGU. This agrees with data from the present study, in which animals that received glucose only via the peripheral route tended to have slightly greater net hepatic glucose uptake when GLP-1 was given intraportally than when it was not (NHGB = −1.6 ± 0.2 vs. −1.2 ± 0.1 mg·kg⁻¹·min⁻¹, GLP-1 vs. Sal, during final 2 h, not significant, P = 0.16) (Fig. 4A). As noted in our earlier study (11), in the presence of intraportal glucose infusion, GLP-1 delivery also tended to have a small, direct effect on the liver. Collectively, therefore, our data suggest that physiological increases in GLP-1 can have a direct, albeit small, stimulatory effect on the liver.

It has been well established that exogenously infused GLP-1 acts as an incretin in both healthy humans and those with Type 2 diabetes (19). As noted above, however, in the past (11), as well as in the present study, there was no difference in arterial or portal plasma insulin levels in the presence or absence of GLP-1 infusion. This agrees with earlier data that showed that dogs that received a systemic infusion of glucose to simulate postprandial peripheral glucose levels showed no change in insulin levels when GLP-1 was infused peripherally to create a physiological increase in its level (10). The fact that a physiological elevation in GLP-1 did not result in changes in pancreatic hormone levels in blood in the dog was discussed at length in our previous publication (11); however, a recent study indicates that, upon stimulation of endogenous GLP-1 release in the rat, total levels of GLP-1 in the hepatic portal vein are less than those in the lymph draining from the gut (6). In our present study, an increase in the intestinal lymph GLP-1 level presumably did not occur, since the elevation in GLP-1 was brought about by infusion. To the extent that lymph GLP-1 is involved in the incretin effect of the peptide, that effect would be absent when the hormone is infused intraportally. On the other hand, numerous investigators (3, 13, 18, 23) have infused GLP-1, presumably in the absence of a rise in lymph GLP-1 levels, and yet they observed an incretin effect of the peptide. The impact, it any, of the increased lymph GLP-1 remains to be clarified.

In conclusion, we have shown that a physiological elevation of plasma GLP-1 in the hepatic portal does not increase whole body glucose utilization when hyperglycemia is induced by peripheral glucose infusion alone. The data presented here, combined with evidence from our earlier study, thus suggest that the GLP-1 secretion that occurs following feeding, when a glucose gradient exists between the periphery and the hepatic portal vein, plays a role in limiting postprandial hyperglycemia. This occurs by increasing glucose utilization independently to form the incretin effect of the peptide.

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DISCLOSURES
A. D. Cherrington has consultancies with Merck, and Novo Nordisk; is on the Scientific Advisory Board of Amylin and owns stock in Amylin.

REFERENCES


