Fructose augments infection-impaired net hepatic glucose uptake during TPN administration

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Fructose augments infection-impaired net hepatic glucose uptake during TPN administration. Am J Physiol Endocrinol Metab 280: E703–E711, 2001.—During chronic total parenteral nutrition (TPN), net hepatic glucose uptake (NHGU) and net hepatic lactate release (NHLR) are markedly reduced (∼45% and ∼65%, respectively) with infection. Because small quantities of fructose are known to augment hepatic glucose uptake and lactate release in normal fasted animals, the aim of this work was to determine whether acute fructose infusion with TPN could correct the impairments in NHGU and NHLR during infection. Chronically catheterized conscious dogs received TPN for 5 days via the inferior vena cava at a rate designed to match daily basal energy requirements. On the third day of TPN administration, a sterile (SHAM, n = 12) or Escherichia coli-containing (INF, n = 11) fibrin clot was implanted in the peritoneal cavity. Forty-two hours later, somatostatin was infused with intraportal replacement of insulin (12 ± 2 vs. 24 ± 2 μU/ml, SHAM vs. INF, respectively) and glucagon (24 ± 4 vs. 92 ± 5 pg/ml) to match concentrations previously observed in sham and infected animals. After a 120-min basal period, animals received either saline (Sham+S, n = 6; Inf+S, n = 6) or intraportal fructose (0.7 mg·kg⁻¹·min⁻¹; Sham+F, n = 6; Inf+F, n = 5) infusion for 180 min. Isoglycemia of 120 mg/dl was maintained with a variable glucose infusion. Combined tracer and arteriovenous difference techniques were used to assess hepatic glucose metabolism. Acute fructose infusion with TPN augmented NHGU by 2.9 ± 0.4 and 2.5 ± 0.3 mg·kg⁻¹·min⁻¹ in Sham+F and Inf+F, respectively. The majority of liver glucose uptake was stored as glycogen, and NHLR did not increase substantially. Therefore, despite an infection-induced impairment in NHGU and different hormonal environments, small amounts of fructose enhanced NHGU similarly in sham and infected animals. Glycogen storage, not lactate release, was the preferential fate of the fructose-induced increase in hepatic glucose disposal in animals adapted to TPN.

THE PRESENCE OF AN INFECTION substantially alters hepatic carbohydrate metabolism. In stressed states such as trauma, injury, or infection, nutritional support is often provided to patients in the form of total parenteral nutrition (TPN). During feeding or glucose infusion, the liver removes glucose from the bloodstream and thereby limits the severity of hyperglycemia. In healthy human volunteers, the liver removes 30–35% of an acute oral glucose load (23). When nutrients are continuously infused on a chronic (i.e., days) basis, glucose uptake by the liver is markedly augmented. The liver of a TPN-adapted dog consumes 45% of the infused glucose, and ∼70% of liver glucose uptake undergoes glycolytic conversion to lactate, which is released into the peripheral circulation (18). In contrast, the presence of an infection reduces net hepatic glucose uptake (NHGU) to only 25% of the glucose infused in chronic TPN-infused dogs and reduces net hepatic lactate release (NHLR) to ∼45% of NHGU (18). Analogously, splanchnic (gut and liver) glucose uptake in stressed patients receiving chronic TPN accounts for ∼20% of the glucose infused (13).

Despite the importance of the liver as a site of glucose disposal and the impact of infection on liver metabolism, no studies have attempted to specifically improve hepatic glucose disposal during nutritional support. The stress-induced impairment in NHGU with chronic nutritional support may be caused by inadequate suppression of hepatic glucose production (HGP) and/or by a failure to stimulate hepatic glucose uptake (HGU). Previous studies (19, 26) indicate that both processes contribute to the reduced NHGU. In response to a large (8 mg·kg⁻¹·min⁻¹) glucose infusion, gluconeogenesis was not suppressed to the same extent in septic patients as in normal volunteers (25). Strategies that improve the suppression of HGP and/or enhance HGU would facilitate NHGU and thus improve whole body glucose disposal.

Recent evidence suggests that low concentrations of fructose (<0.5 mM) augment NHGU in vivo (27), presumably by activation of glucokinase (32). Because TPN-adapted animals exhibit elevated NHGU, it is unknown whether fructose can further augment NHGU and NHLR in a chronic TPN setting. Moreover, whether fructose can enhance NHGU in the presence...
of an infection has not been determined. Therefore, our work addressed the following questions: 1) does fructose enhance NHGU during TPN administration; 2) does infection impair the ability of fructose to stimulate NHGU; and 3) does fructose infusion improve hepatic glucose disposition?

METHODS

Animal preparation. Twenty-three female mongrel dogs were fed a standard Kal-Kan meat (Vernon, CA) and Purina Lab Canine #5006 (Purina Mills, St. Louis, MO) diet once daily and had free access to water. The composition of the diet, based on dry weight, was 52% carbohydrate, 31% protein, 11% fat, and 6% fiber. Dogs were housed in a facility that met the guidelines of the Association for the Accreditation of Laboratory Animal Care International. The protocols were approved by the Vanderbilt University Medical Center Animal Care Committee. Health of the animals was determined before surgery and before TPN administration as having a good appetite (i.e., consumed ≥75% of the daily ration) and normal stools, hematocrit >0.35, and leukocyte count <18,000/mm^3.

Surgical preparation. A laparotomy was performed under general anesthesia (15 mg/kg iv of thiopental for induction and 1.0% isoflurane as an inhalant during surgery) on healthy dogs weighing 17–24 kg. During the laparotomy, Silastic (Dow Corning, Midland, MI) infusion catheters (0.03 in. ID) were placed in the splenic and jejunal veins, and the gastroduodenal vein was ligated. Catheters (0.04 in. ID) for blood sampling were positioned in the portal and left common hepatic veins. Two infusion catheters (0.04 in. ID) for TPN were placed in the inferior vena cava (IVC), and the free ends were exteriorized and tunneled subcutaneously behind the left clavicle. Flow probes (Transonic Systems, Ithaca, NY) were positioned about the portal vein, hepatic artery, and right external iliac artery. Incisions were made in the right and left inguinal regions, and a sampling catheter (0.04 in. ID) was placed in the left common iliac vein as the tip was advanced distal to the IVC anastomosis; another catheter was placed in the right external iliac artery, and its tip was advanced to the abdominal aorta.

All catheters were filled with 0.9% NaCl (saline) containing heparin (200 U/ml; Elkins-Sinn, Cherry Hill, NJ). The free ends of the catheters and flow probes were exteriorized and placed in subcutaneous pockets. The dogs received penicillin G (600,000 U iv; Anthony Products, Irwindale, CA) to minimize the possibility of infection. Flunixin (0.1 mg/kg body wt; Fort Dodge Lab, Fort Dodge, IA) was injected intramuscularly immediately after wound closure for acute pain relief. Dogs received ampicillin (500 mg; Bristol-Myers Squibb, Princeton, NJ) orally for 3 days after surgery.

Nutritional support. After an allowance of ≥14 days for recovery from surgery, the IVC catheters were exteriorized under local anesthesia (2% lidocaine, Abbott, North Chicago, IL). TPN was infused as the sole source (i.e., all enteral nutrients were discontinued) of calories into one or both of these catheters continuously for 5 days by use of an ambulatory infusion pump (Dakmed, Buffalo, NY, or Walkmed-350, McKinley, Lakewood, CO). Dogs were a jacket (Alice King Chatham, Los Angeles, CA) with two large pockets for the nutrition and the pump. The TPN was designed to be isocaloric on the basis of predicted resting energy expenditure (30). The composition of the TPN included glucose, lipids, amino acids, saline, potassium phosphate (90 mg·kg⁻¹·day⁻¹), and a multivitamin supplement (MVI-12, Astra, Westborough, MA). Glucose (50% dextrose, Abbott, ~10 mg·kg⁻¹·min⁻¹) made up 75% of the nonprotein calories, whereas 20% Intralipid (Baxter, Deerfield, IL) constituted the remaining 25% of the energy requirements. Travasol (10%, Baxter) was infused to supply basal nitrogen requirements (1.5 × body wt⁰.⁶⁷ g protein/day). During the 5 days of TPN, the animals had free access to water.

Induction of infection. A 1% fibrinogen (Sigma, St. Louis, MO) solution was filtered (0.45 μm) under sterile conditions. To initiate clot formation, thrombin (1,000, U, Gentrac, Middleton, WI) was added to the filtrate. The bacterial clot also contained a nonlethal dose (2 × 10⁸ organisms/kg body wt) of Escherichia coli determined by serial dilution followed by plating. Bacteria (American Type Tissue Culture no. 25922) were prepared by inoculation of 1 liter of Trypticase soy broth (Becton-Dickinson, Cockeysville, MD) and incubation overnight at 37°C. Bacteria were pelleted by centrifugation on the next day and were washed with and reconstituted in sterile saline before addition to the filtrate.

After the third day of the 5-day TPN infusion, a second laparotomy was performed under anesthesia, and the TPN infusion was continued. An abdominal midline incision was made at a point below the incision made during the previous surgery. Either a sterile (SHAM) or a bacterial (INF) fibrin clot was implanted in the peritoneal cavity. In addition to the TPN, animals received saline intravenously during the laparotomy and on the next day.

Experimental protocol. On the 5th day of TPN and 42 h after clot implantation, a study was performed. Free ends of all catheters were exteriorized under local anesthesia, and their contents were aspirated and flushed with saline. Leads from the flow probes were also exteriorized and connected to a flowmeter. The dog was placed in a Pavlov harness for the duration of the study. Angiocaths (18 gauge, Abbott) were inserted into both cephalic veins for infusion of radioactive tracers, glucose, and somatostatin (SRIIF; Bachem, Torrance, CA). Blood pressure, heart rate (Micro-Med, Louisiville, KY), and rectal temperature (Yellow Springs Instruments, Yellow Springs, OH) were measured at the basal period.

The study consisted of a 120-min basal period followed by a 180-min experimental period. Primed (44 and 27 μCi) constant infusions of [3-3H]- and [U-14C]glucose (New England Nuclear, Wilmington, DE) were begun at rates of 0.4 and 0.3 μCi/min, respectively, with syringe pumps (Harvard Apparatus, Holliston, MA) and continued for the duration of the study. A fresh TPN solution containing the nonglucose nutrients was prepared separate from the glucose. In this way, the glucose infusion rate was adjusted to maintain isoglycemia (120 mg/dl), whereas the other TPN components were infused at a constant rate. Small blood samples (0.4 ml) were taken every 10 min and centrifuged immediately to measure arterial plasma glucose concentration with a Beckman glucose analyzer II (Beckman Instruments, Fullerton, CA). The exogenous glucose infusion rate was adjusted to maintain isoglycemia. Saline was infused to replace blood volume withdrawn by sampling.

During tracer equilibration, SRIIF (0.8 μg·kg⁻¹·min⁻¹) was infused to suppress endogenous pancreatic hormone secretion. Insulin (regular Iletin II; Eli Lilly, Indianapolis, IN) and glucagon (Eli Lilly) were replaced by intraportal infusion via the splenic and jejunal veins. In the sterile clot group (SHAM, n = 12), insulin and glucagon were infused at ~400 μU·kg⁻¹·min⁻¹ and 0.1 ng·kg⁻¹·min⁻¹, respectively. In the animals that received a bacterial clot (INF, n = 11), insulin (1,000 μU·kg⁻¹·min⁻¹) and glucagon (~2.5 ng·kg⁻¹·min⁻¹) were infused to match the higher levels observed during infection (18). Arterial plasma insulin and...
glucagon levels in INF were 26 ± 2 μU/ml and 70 ± 9 pg/ml, respectively, before initiation of SRIF infusion.

After the basal period, SHAM and INF animals received either saline (+S) or fructose (+F; 0.74 mg·kg⁻¹·min⁻¹ or 4.1 μmol·kg⁻¹·min⁻¹; Sigma) in the portal vein via jejunal and splenic infusion catheters. Blood samples from the iliac artery and portal, hepatic, and iliac veins were taken every 20 min during the last 60 min of the basal period and every 30 min during the 180-min experimental period. Blood flows and hematocrit were recorded at each sampling point. At the end of the experiment, animals were killed with an overdose of pentobarbital sodium (Veterinary Lab, Lenexa, KS). Tissue samples from each of the seven liver lobes and a muscle (adductor magnus et brevis) were freeze-clamped with Wallenburg clamps precooled in liquid nitrogen and were stored at −70°C until analysis. The entire liver was removed rapidly and weighed.

Sample processing. Blood samples were placed in chilled tubes containing EDTA and were processed as described in Ref. 16. For analysis of catecholamines, whole blood (1 ml) was treated with 20 μl of a solution containing 90 mg/ml of EGTA and 50 mg/ml of glutathione. Samples for glucoseo-regenetic metabolite, fructose, and glutamine content were processed by adding 1 ml of whole blood to 3 ml of 4% perchloric acid (PCA). Blood ¹⁴C-CO₂ was assessed in triplicate on arterial, portal, and hepatic vein samples, as described by Chan and Dehaye (4). To assess ¹⁴C incorporation into lactate and amino acids, 3 ml of blood were added to 3 ml of 8% PCA. The sample was centrifuged, neutralized with 2 N KOH, and placed over ion exchange columns to separate into ¹⁴C-lactate, ¹⁴C-glucose, and ¹⁴C-labeled amino acids (22). Blood samples were centrifuged at 3,000 rpm for 10 min, and the remaining plasma was stored at −70°C for later analyses. Plasma treated with aprotinin (500 kallikrein inactivator units/ml plasma; Miles, Kankakee, IL) was analyzed for glucagon content. Plasma glucose specific activity (SA) was determined spectrophotometrically (Wako Chemicals, Richmond, VA).

Analysis. Immunoreactive insulin and glucagon were assayed using a double antibody disequilibrium procedure (21) [coefficients of variation (CV) 9 and 8%, respectively], and cortisol was assayed using a single antibody technique (12) with a Diagnostic Products radioimmunoassay kit (Los Angeles, CA; CV 8%). HPLC techniques were used to assay plasma epinephrine and norepinephrine (CV 11 and 6%, respectively) (14). Analysis of lactate, alanine, and glycerol was performed on an automated centrifugal analyzer (Monarch 2000; Instrumentation Laboratory, Lexington, MA) by using a modification of the method of Lloyd et al. (15). The concentration of nonesterified fatty acids (NEFA) was determined spectrophotometrically (Wako Chemicals, Richmond, VA).

The methods of Bernt and Bergmeyer (1) and of Beutler (2) were adapted to measure blood glutamine and fructose (lower detection limit = 15 μM) content, respectively, with a Technicon Autoanalyzer II (Bran Luebbe, Buffalo Grove, IL). Fructose content was measured in blood deproteinized with 4% PCA. The extract was neutralized with 10% KOH and incubated for 60 min with an equal volume of 0.1 M phosphate buffer (pH 7.4) containing glucose oxidase (10 U/ml) and catalase (600 U/ml). Glycogen content was determined using the enzymatic method of Chan and Exton (5). Hepatic lipids were extracted with the Folch method (11), and the ¹⁴C radioactivity of the extract was determined with liquid scintillation counting.

Calculations. The rates of total glucose appearance (Ra) and utilization (Ru) were determined according to the method of Wall et al. (33), as simplified by de Bodo et al. (9). Whole body endogenous glucose production (EGP) was the difference between Ra and exogenous glucose infusion rate. The substrate (glucose, lactate, alanine, glycerol, fructose, and NEFA) load entering the liver was calculated as the sum of the loads in the hepatic artery and portal vein, (Aa × HABF) + (Pa × PBF), where Aa and Pa represent the substrate concentrations in the iliac artery and portal vein, and HABF and PBF represent blood flow in the hepatic artery and portal vein. Similarly, the substrate load leaving the liver was Hl × THBF, in which Hl and THBF represent the hepatic vein substrate concentration and total hepatic blood flow (THBF = HABF + PBF). Net hepatic substrate balance was calculated as the difference between the entering and exiting substrate loads and was denoted as either uptake or output. Likewise, net hindlimb substrate uptake was calculated using the formula (Aa − Vv) × ABF, where Vv is the substrate concentration in the iliac vein and ABF is the iliac artery blood flow. Net hepatic fractional extraction (FHE) of substrate was calculated as the net hepatic substrate balance divided by the substrate load entering the liver. Plasma flow was used for NEFA balance by multiplying blood flow and (1 − hematocrit).

Unidirectional hepatic glucose uptake (HGU) was calculated as [3H]glucose uptake by the liver divided by inflowing [3H]glucose SA. Hepatic glucose production (HGP) was the difference between unidirectional HGU and NHGU. Hepatic glucose oxidation was measured as the ratio of liver ¹⁴CO₂ production and inflowing [¹⁴C]glucose SA, substituting ¹⁴CO₂ disintegrations per minute in the previous balance equation. Hepatic glucose storage was the portion of NHGU not accounted for by glucose oxidation and NHLR.

Because somatostatin suppresses endogenous insulin and glucagon secretion, we calculated the insulin and glucagon levels in the portal vein using the formula [(Aa − Vv) × ABF], where Vv is the substrate concentration in the iliac vein and ABF is the iliac artery blood flow, and the portal vein blood flow, and the portal vein hormone infusion rate, respectively. In the case of insulin, the equation was modified to account for the extraction of insulin by the nonhepatic splanchnic tissues (28).

The glycogen synthetic rate was calculated by dividing hepatic [¹⁴C]glycogen accumulation (dpm·kg⁻¹·min⁻¹) by the average inflowing [¹⁴C]glucose SA. Net glycogen accumulation attributed to fructose was calculated as the increment in [¹⁴C]glycogen synthetic rate (difference between fructose-infused group and corresponding saline-infused group) multiplied by the fraction of time of fructose infusion (total min/180 min). Analogous to glycogen accumulation, hepatic [¹⁴C]-labeled lipid deposition was calculated as total radioactivity divided by inflowing [¹⁴C]glucose SA.

Statistics. Results are expressed as means ± SE in the basal period (average of 3 sampling points) and during the last 60 min of the experimental period (average of 3 sampling points). Fructose concentrations are expressed as means ± SE for the last 120 min of the experimental period. Basal data are presented as average of sham (SHAM, n = 12) and infected (INF, n = 11) animals, unless otherwise indicated. Student’s unpaired t-test was used for comparisons of sham and infected animals in the basal period. To determine the effect of fructose, statistical comparisons over time were made with two-way ANOVA (SYSTAT, Evanston, IL) between fructose-infused groups and their corresponding saline-infused groups. Statistical significance was designated as a P value < 0.05. Results from infected + saline (Inf+S)
and sham + saline (Sham+S) groups have been presented previously (10).

RESULTS

General characteristics. Body temperature, heart rate, mean arterial pressure, and blood flow in the hepatic artery and portal vein are shown in Table 1. Infected animals were typically hyperthermic, normotensive, and tachycardic, with significantly elevated hepatic arterial blood flow. There were no changes in blood flow over time in any group (data not shown).

Hormones. As expected with the infusion rates, arterial plasma insulin concentrations were higher ($P < 0.05$) in the basal and experimental periods of the INF group, $24 \pm 2$ and $25 \pm 2 \mu$U/ml, than in the SHAM group, $12 \pm 2$ and $13 \pm 2 \mu$U/ml (Fig. 1). Basal arterial plasma glucagon concentrations were also higher in INF ($24 \pm 4$ vs. $92 \pm 5$ pg/ml, SHAM vs. INF, respectively; $P < 0.05$) and did not change in the experimental period ($22 \pm 3$ and $89 \pm 7$ pg/ml). The calculated insulin ($40 \pm 2$ and $100 \pm 14 \mu$U/ml) and glucagon ($32 \pm 3$ and $285 \pm 26$ pg/ml) concentrations in the portal vein for the SHAM and INF groups were also higher. Basal concentrations of cortisol, epinephrine, and norepinephrine were similar in SHAM and INF groups (Table 1) and did not change in the experimental period (data not shown).

Fructose. Arterial fructose concentrations in the basal period were $41 \pm 11$ and $23 \pm 10 \mu$M in Sham+F ($n = 6$) and Inf+F ($n = 3$). During the final 120 min of the intraportal fructose infusion, arterial fructose concentrations increased modestly to $101 \pm 10$ and $116 \pm 24 \mu$M, Sham+F and Inf+F, respectively; Fig. 2. Fructose concentrations reached $321 \pm 1$ and $367 \pm 36 \mu$M in the portal vein and $91 \pm 8 \mu$M and $127 \pm 29 \mu$M in the hepatic vein. Hepatic fructose loads were similar ($7.8 \pm 0.6$ vs. $9.2 \pm 2.0 \mu$mol·kg$^{-1}$·min$^{-1}$, Sham+F vs. Inf+F). Net hepatic fructose uptake ($4.9 \pm 0.3$ vs. $4.6 \pm 1.7 \mu$mol·kg$^{-1}$·min$^{-1}$) and HFE of fructose ($0.63 \pm 0.03$ vs. $0.46 \pm 0.11$) were also similar.

Whole body glucose kinetics. Arterial plasma glucose concentrations were $120 \pm 2$ mg/dl in both SHAM and INF groups and were maintained for the duration of the study (Fig. 3). Exogenous glucose infusion rate (GIR) was $8.6 \pm 0.6$ and $11.5 \pm 0.6$ mg·kg$^{-1}$·min$^{-1}$ in SHAM ($n = 11$) and INF ($n = 11$, $P < 0.05$) and did not increase with saline infusion. In the fructose-infused groups, GIR increased by $3.7 \pm 1.0$ and $2.1 \pm 0.6$ mg·kg$^{-1}$·min$^{-1}$, Sham+F and Inf+F, respectively. Basal endogenous glucose production rates were similar ($1.5 \pm 0.3$ and $1.6 \pm 0.7$ mg·kg$^{-1}$·min$^{-1}$, SHAM and INF) and did not change significantly with fructose ($0.7 \pm 0.5$ vs. $0.9 \pm 0.4$ mg·kg$^{-1}$·min$^{-1}$, Sham+F vs. Inf+F) or saline ($0.1 \pm 0.4$ vs. $0.4 \pm 0.2$ mg·kg$^{-1}$·min$^{-1}$, Sham+S vs. Inf+S) infusion.

Hepatic glucose balance. Basal NGU was higher in SHAM ($3.7 \pm 0.3$ mg·kg$^{-1}$·min$^{-1}$) than in INF ($1.7 \pm 0.3$ mg·kg$^{-1}$·min$^{-1}$, $P < 0.05$; Fig. 3). This was paralleled by higher basal unidirectional HGU, $3.9 \pm 0.3$ vs. $2.2 \pm 0.2$ mg·kg$^{-1}$·min$^{-1}$, SHAM vs. INF ($P < 0.05$). There were no changes in NGU over time in the groups that received saline. Within 30 min of fructose infusion, NGU markedly increased ($P < 0.05$) in both Sham+F and Inf+F groups and remained elevated

![Graph](http://ajpendo.physiology.org/)

**Fig. 1.** Arterial plasma insulin and glucagon concentrations in sham and infected animals receiving saline (Sham+S, $n = 6$; Inf+S, $n = 6$) or fructose (Sham+F, $n = 6$; Inf+F, $n = 5$) during a pancreatic clamp [somatostatin (SRIF), insulin, and glucagon]. Data are expressed as means ± SE.

**Table 1. Baseline arterial hormone concentrations and hemodynamics of sham ($n = 12$) and infected ($n = 11$) groups receiving TPN for 5 days**

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<th>Sham</th>
<th>Infected</th>
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<tr>
<td>Body temperature, °C</td>
<td>38.5 ± 0.2</td>
<td>39.4 ± 0.2$^*$</td>
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<tr>
<td>Heart rate, beats/min</td>
<td>87 ± 5</td>
<td>134 ± 10$^*$</td>
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<tr>
<td>Hepatic arterial blood flow, ml·kg$^{-1}$·min$^{-1}$</td>
<td>9 ± 1</td>
<td>22 ± 1$^*$</td>
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<tr>
<td>Portal vein blood flow, ml·kg$^{-1}$·min$^{-1}$</td>
<td>22 ± 1</td>
<td>21 ± 1</td>
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<tr>
<td>Mean arterial pressure, mmHg</td>
<td>98 ± 4</td>
<td>96 ± 4</td>
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<tr>
<td>Cortisol, µg/dl</td>
<td>2.8 ± 0.3</td>
<td>3.1 ± 0.4</td>
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<tr>
<td>Epinephrine, pg/ml</td>
<td>134 ± 25</td>
<td>100 ± 24</td>
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<tr>
<td>Norepinephrine, pg/ml</td>
<td>208 ± 59</td>
<td>350 ± 75</td>
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Values are means ± SE. TPN, total parenteral nutrition. $^*$P < 0.05 vs. sham group.
for the duration of the study. NHGU increased to 6.8 ± 0.7 mg·kg⁻¹·min⁻¹ in Sham+F and 4.3 ± 0.4 mg·kg⁻¹·min⁻¹ in Inf+F (Δ2.9 ± 0.4 and Δ2.5 ± 0.3 mg·kg⁻¹·min⁻¹, respectively). Net HFE of glucose in the basal period was 0.14 ± 0.01 and 0.05 ± 0.01 in SHAM and INF (P < 0.05). Analogous to NHGU, HFE increased during fructose infusion (Δ0.10 ± 0.01 and Δ0.08 ± 0.02, Sham+F and Inf+F, respectively). Basal HGP was similar in SHAM and INF (0.2 ± 0.2 and 0.5 ± 0.2 mg·kg⁻¹·min⁻¹) and did not change significantly with fructose or saline (data not shown).

**Hepatic glucose disposition.** Arterial lactate concentrations in the basal period were higher in SHAM relative to INF (892 ± 62 vs. 669 ± 64 μM; P < 0.05; Fig. 4). When fructose was infused, arterial lactate concentrations rose (P < 0.05) by 340 ± 107 and 324 ± 31 μM in Sham+F and Inf+F. Basal hepatic lactate release (NHLR) was 23 ± 2 μmol·kg⁻¹·min⁻¹ in SHAM and did not change significantly in the experimental period (Δ1.3 ± 1.0 and Δ−1.3 ± 1.7 μmol·kg⁻¹·min⁻¹, Sham+F and Sham+S). As expected, basal NHLR was lower during infection (10 ± 3 μmol·kg⁻¹·min⁻¹, P < 0.05). NHLR tended to increase with fructose infusion (Δ7 ± 1 μmol·kg⁻¹·min⁻¹, Inf+F), although this was not significant with respect to Inf+S (Δ−1 ± 2 μmol·kg⁻¹·min⁻¹). Hepatic glucose oxidation rates in the basal period were 0.4 ± 0.1 and 0.5 ± 0.2 mg·kg⁻¹·min⁻¹ in SHAM (n = 12) and INF (n = 10) and did not increase with saline or fructose infusion (data not shown).

The percentage of NHGU released as lactate in the basal period was 60 ± 3% in SHAM (n = 12) and 35 ± 7% in INF (n = 10; P < 0.05). The proportion of NHLR derived from NHGU decreased with fructose infusion in Sham+F (Δ−23 ± 4%) but did not change in other groups (Δ5 ± 6, Δ−11 ± 7, and Δ−2 ± 11% in Sham+S, Inf+F, and Inf+S, respectively). The proportion of hepatic [¹⁴C]glucose uptake released as [¹⁴C]lactate in Sham+F (n = 5) was 33 ± 5% and did not change with fructose infusion (35 ± 7%). Infected groups had very low hepatic [¹⁴C]lactate release, which made it difficult to measure a change in glycolysis with tracer techniques. The proportion of hepatic [¹⁴C]glucose uptake released as [¹⁴C]lactate in INF (n = 7) was 8 ± 10%, and we could not detect a significant change in [¹⁴C]lactate release with fructose infusion. The conversion of
[14C]glucose to [14C]alanine was minimal, 4% in Sham+S and ~1% in Inf+F.

The proportion of NHGU oxidized, released as lactate, and stored in SHAM was 12, 60, and 28%, respectively, and fructose infusion shifted the proportion to 7, 32, and 61% (Sham+F). In INF (n = 10), 28% of NHGU was oxidized, 35% was released as lactate, and 37% was stored. As in Sham+F, fructose infusion during infection shifted the fate of glucose to storage, with 15, 36, and 49% in the respective pathways (Inf+F; Fig. 5).

Terminal hepatic glycogen mass in Sham+S (n = 5) and Sham+F (n = 6) was 77 ± 2 and 75 ± 9 mg glycogen/g liver tissue, respectively. With infection, glycogen mass was 21 ± 3 mg/g in Inf+S (n = 4) and 36 ± 9 mg/g in Inf+F (n = 5), but we could not detect a significant increase in glycogen mass with fructose infusion. Tracer 14C-determined glycogen synthetic rates were 0.5 ± 0.1 and 0.5 ± 0.3 mg·kg−1·min−1 in Sham+S and Inf+S. The increment in glycogen synthesis during fructose infusion was 2.0 ± 0.3 mg·kg−1·min−1 in Sham+F (n = 6) and 0.7 ± 0.4 mg·kg−1·min−1 in Inf+F (n = 4). Basal hepatic 14C-labeled lipid deposition rates were ~0.1 mg·kg−1·min−1 in both SHAM (n = 5) and INF (n = 4), and there was no increase with fructose infusion (data not shown).

**Hepatic metabolites.** Arterial alanine concentrations in blood were lower with infection (471 ± 48 vs. 205 ± 0.15 μM, SHAM vs. INF; P < 0.05) and did not change during the experimental period. Basal net hepatic alanine uptake rates (1.4 ± 0.2 and 2.7 ± 0.3 μmol·kg−1·min−1; P < 0.05) and net HFE of alanine (0.20 ± 0.02 vs. 0.29 ± 0.02, P < 0.05; Table 2) were higher during infection and did not increase with fructose infusion.

![Fig. 4. Arterial blood lactate concentration and net hepatic lactate release (NHLR) in Sham+S (n = 6) and Inf+S (n = 6) or Sham+F (n = 6) and Inf+F (n = 5) animals during a pancreatic clamp (SRIF, insulin, and glucagon). Data are expressed as means ± SE.](image-url)

![Fig. 5. NHGU divided into components of oxidation, lactate release, and storage in Sham+F (n = 6) and Inf+F (n = 5) animals before (Basal) and during fructose infusion (+F). Data are expressed in mg of glucose equivalents·kg−1·min−1.](image-url)

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<th>Table 2. Concentrations and uptake rates</th>
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<td><strong>Alanine, μM</strong></td>
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<td><strong>Net hepatic alanine uptake, μmol·kg−1·min−1</strong></td>
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Values are means ± SE. Concentrations and uptake rates for alanine, glycerol, and nonesterified fatty acids (NEFA) in the basal period and in final 60 min of experimental (Exp) period from sham and infected groups receiving saline (Sham+S, Inf+S) or fructose (Sham+F, Inf+F).
Basal arterial plasma NEFA concentrations were higher in SHAM (307 ± 32 vs. 220 ± 11 μM, SHAM vs. INF; P < 0.05; Table 2) and remained stable with saline or fructose infusion. Basal net hepatic NEFA uptake rates were similar (0.7 ± 0.2 and 0.8 ± 0.1 μmol·kg\(^{-1}\)·min\(^{-1}\)) and did not change significantly with saline or fructose infusion. Arterial glycerol concentrations (66 ± 4 and 64 ± 4 μM) were similar in SHAM and INF and also did not increase in the experimental period. Although basal net hepatic glycerol uptake was higher during infection (1.2 ± 0.1 and 1.6 ± 0.1 μmol·kg\(^{-1}\)·min\(^{-1}\), SHAM and INF, P < 0.05), there were no significant changes in the experimental period.

**Hindlimb.** Net hindlimb glucose uptake rates during the basal period in SHAM and INF were 14 ± 2 and 16 ± 2 mg/min, and net hindlimb fractional extraction of glucose was 0.09 ± 0.02 and 0.12 ± 0.02. Basal net hindlimb lactate uptake was 2.0 ± 0.3 and 0.5 ± 0.3 μmol·kg\(^{-1}\)·min\(^{-1}\) in SHAM (n = 11) and INF (n = 11); net hindlimb fractional extraction of lactate was 0.37 ± 0.07 and 0.09 ± 0.05. Fructose infusion did not alter these variables (data not shown). Muscle glycogen mass was not different between groups (data not shown).

**DISCUSSION**

Although chronic TPN administration markedly enhances NHGU and NHLR in dogs, the presence of an infection sharply reduces both processes despite elevated insulin and glucose concentrations (18). Fructose is known to augment liver glucose uptake in non-TPN adapted animals. In the present study, we demonstrated that fructose enhanced NHGU during chronic TPN administration in both sham and infected dogs. Although acute fructose infusion overcame the infection-induced impairment in NHGU, it did not correct the impairment in NHLR.

Unexpectedly, the ability of fructose to enhance glucose disposal in normal TPN-adapted animals was comparable to the effect observed in fasted glucose-infused animals. Acute fructose infusion during chronic TPN increased NHGU by 2.9 mg·kg\(^{-1}\)·min\(^{-1}\) in the presence of slightly elevated glucose (120 mg/dl) and insulin (12 μU/ml) concentrations. In 42-h-fasted dogs in the presence of greater hyperglycemia (225 mg/dl) and hyperinsulinemia (30 μU/ml), a similar fructose infusion raised liver glucose uptake by 7 mg·kg\(^{-1}\)·min\(^{-1}\) (27). The increments in HFE of glucose, however, were similar (Δ0.14 vs. Δ0.10; fasted vs. TPN), suggesting that nutritional support does not substantially blunt the response to fructose. The mechanism by which fructose activates NHGU is presumably via an increase in fructose 1-phosphate concentration, which allows translocation and activation of glucokinase (31, 32). The ability of fructose to stimulate NHGU beyond that seen with TPN implies that, despite the already high NHGU with TPN, the majority of GK protein is not already maximally translocated.

Given the different hormone environments, the response to fructose in infected animals was remarkably similar to that of sham animals. Insulin and glucagon concentrations were clamped at the elevated levels seen during infection, allowing us to examine the effects of fructose in the absence of changes in these hormones. As we have observed previously (18), infection impaired NHGU by ~50%. Given the impaired NHLR and the known in vitro effects of glucagon to partially reverse the effect of fructose (3, 8), we expected infection to impair the response to fructose; in fact, infection did not impair the response to fructose. It is possible that the acute inhibitory effect of glucagon on fructose action does not persist chronically. Consistent with this, the acute and chronic effects of glucagon are markedly different. Acute changes in glucagon primarily alter hepatic glycogen metabolism, the primary fate of glucose in response to fructose. In contrast, glucagon chronically modulates gluconeogenesis (17, 20). In four additional experiments, acute reduction (↓ 40%) of the glucagon replacement rate did not alter the ability of fructose to stimulate NHGU (data not presented). However, the glucagon replacement rate was still substantially higher than in sham animals receiving chronic TPN; therefore, the inhibitory effect of glucagon may still be present. Nevertheless, it is also possible that the underlying impairment in NHGU seen with infection is distinct from the site of action of fructose.

The effect of fructose was mediated rapidly and specifically on the liver, as hindlimb glucose uptake was not altered by acute fructose infusion. This is consistent with a previous report in the dog (27). Circulating arterial levels of fructose were relatively low (~100 μM) because of the high first-pass extraction by the liver. The fructose-induced increase in NHGU (Δ2.9 ± 0.4 and Δ2.5 ± 0.3 mg·kg\(^{-1}\)·min\(^{-1}\) in Sham+F and Inf+F) roughly accounted for the increase in whole body glucose disposal (Δ3.8 ± 0.6 and 2.9 ± 0.6 mg·kg\(^{-1}\)·min\(^{-1}\)).

Once consumed by the liver, glucose undergoes one of three possible fates: oxidation, release as lactate, or storage in the form of glycogen or lipid. Acute fructose infusion did not increase hepatic glucose oxidation. Given the marked activation of glycolysis during TPN administration, we expected fructose infusion to substantially increase lactate release; however, an increase was not observed. In fact, in Sham+F, the proportion of NHGU diverted to lactate fell in response to fructose infusion, whereas the proportion of \([^{14}C]\)glucose diverted to \([^{14}C]\)lactate was unaltered. These data suggest that the lack of a rise in NHLR, despite a rise in NHGU, was probably a result of an inhibition of hepatic glycogenolysis and subsequent conversion to lactate. In contrast, in infected animals, although basal NHLR was lower, NHLR tended to increase in Inf+F during fructose infusion. Thus glycogenolysis may not have decreased to the same extent as in Sham+F. In previous studies in fasted animals (27), fructose infusion did enhance NHLR, possibly because hepatic glycogenolysis was already suppressed by the accom-
panying hyperglycemia and hyperinsulinemia. Why glycogenolysis remains active in the TPN-adapted state is unknown.

Sham and infected animals demonstrated a twofold increase in hepatic glucose storage in response to fructose. The increase reflects greater glycogen deposition because tracer incorporation into lipid was minimal. Glycogen was also the major glucose fate during acute fructose infusion in fasted dogs (27). Fructose is known to enhance both the direct and indirect pathways of glycogen synthesis (24), and our data support increases in both pathways. Although fructose increased tracer incorporation into glycogen in Sham+F (2.0 mg·kg⁻¹·min⁻¹), which accounts for the majority of the increase in NHGU, we did not observe a net increase in glycogen mass. It is possible that 1) near-maximal hepatic glycogen capacity is achieved with chronic TPN or 2) we lacked the statistical power to detect an increase in glycogen mass in Sham+F because of a large variance. In contrast, the 15 mg/g increase in hepatic glycogen mass in Inf+F approximates the theoretical increase in mass (12 mg/g), assuming that all of the additional carbohydrate taken up was stored as glycogen. However, the increase (0.7 ± 0.4 mg·kg⁻¹·min⁻¹) in tracer-determined glycogen synthetic rate attributed to fructose is lower than predicted, suggesting that some of the glycogen was formed via the indirect pathway. Although our data cannot differentiate between a stimulation of glycogen synthase and/or an inhibition of phosphorylase, they are consistent with in vitro evidence suggesting that fructose stimulates glycogen synthase (6).

Acute fructose infusion had no effect on hepatic lipid metabolism. Tracer incorporation into lipid was insignificant, and neither arterial NEFA concentration nor hepatic NEFA uptake increased significantly during fructose infusion. In contrast, some studies have found adverse effects (increased triglyceride and cholesterol levels) of fructose in at-risk patients when larger amounts of fructose (e.g., 10% of calories) were consumed for several weeks (7). In the present study, however, a 3-h infusion of small amounts of fructose did not alter hepatic lipid metabolism.

Thus, in the TPN-adapted state, intraportal fructose infusion enhanced NHGU in sham and infected dogs. Although the ability of fructose to stimulate NHGU was not impaired by infection, fructose did not correct the impairment in hepatic lactate release. Glycogen was a major fate of the additional glucose utilized by the liver. Future studies can be done to determine whether the addition of small amounts of fructose to TPN will chronically improve liver glucose uptake and limit the hyperglycemia seen during stress.

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