Interaction of a selective serotonin reuptake inhibitor with insulin in the control of hepatic glucose uptake in conscious dogs

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Moore, Mary Courtney, Catherine A. DiCostanzo, Dominique Dardevet, Margaret Lautz, Ben Farmer, and Alan D. Cherrington. Interaction of a selective serotonin reuptake inhibitor with insulin in the control of hepatic glucose uptake in conscious dogs. Am J Physiol Endocrinol Metab 288: E556–E563, 2005.—Whether hyperinsulinemia is required for stimulation of net hepatic glucose uptake (NHGU) by a selective serotonin reuptake inhibitor (SSRI) was examined in four groups of conscious 42-h-fasted dogs, using arteriovenous difference and tracer ([3-3H]glucose) techniques. Experiments consisted of equilibration (120–30 min), basal (30–0 min), and experimental periods (Exp; 0–240 min). During Exp, somatostatin, intraportal insulin [at basal (Ins groups) or 4-fold basal rates (INS groups)], basal intraportal glucagon, and peripheral glucose (to double hepatic glucose load) were infused. In the Fluv-Ins (n = 7) and Fluv-INS groups (n = 6), saline was infused intraportally from 0 to 90 min (P1), and fluvoxamine was infused intraportally at 2 μg·kg⁻¹·min⁻¹ from 90 to 240 min (P2). Sal-Ins (n = 9) and Sal-INS (n = 8) received intraportal saline in P1 and P2. NHGU during P2 was 8.4 ± 1.4 and 6.9 ± 2.3 μmol·kg⁻¹·min⁻¹ in Sal-Ins and Fluv-Ins, respectively (not significant), and 13.3 ± 2.2 and 20.9 ± 3.1 μmol·kg⁻¹·min⁻¹ (P < 0.05) in Sal-INS and Fluv-INS. Unidirectional (tracer-determined) hepatic glucose uptake was twofold greater (P < 0.05) in Fluv-INS than Sal-INS. Net hepatic carbon storage in the presence of hyperinsulinemia but not euinsulinemia, suggesting that hepatocyte-targeted SSRIs may reduce postprandial hyperglycemia.

glicemia; liver; fluvoxamine

INTRAPORTAL INFUSION OF SEROTONIN (5-hydroxytryptamine; 5-HT) (22) and the selective serotonin reuptake inhibitor (SSRI) fluvoxamine (20) enhanced net hepatic glucose uptake (NHGU) in conscious dogs during a hyperinsulinemic hyperglycemic clamp. Although the exact mechanism for this action is unclear, it suggests that SSRIs or 5-HT agonists might provide a tool for reducing postprandial glycaemia in individuals with diabetes. This raises the question of whether SSRI treatment would be effective in the presence of a relative deficiency of insulin. Therefore, we examined NHGU and total body glucose disposal (Ra) in conscious dogs during intraportal infusion of fluvoxamine under conditions of hyperglycemia and either euinsulinemia or hyperinsulinemia. Fluvoxamine was effective in bringing about a significant enhancement of NHGU and hepatic carbon storage in the presence of hyperinsulinemia but not euinsulinemia.

RESEARCH DESIGN AND METHODS

Animals and surgical procedures. Studies were carried out on conscious 42-h-fasted mongrel dogs of either sex with a mean weight of 22.8 ± 0.4 kg. Diet and housing were as previously described (29), and the protocol was approved by the Vanderbilt University Institutional Animal Care and Use Committee. The 42-h fast is well tolerated by the dog, which exhibits little change in blood glucose (Δ 0.2–0.3 mM) and only a 10% increase in plasma glucagon between 18 and 42 h of fasting (24). However, it reduces the hepatic glycogen to a stable concentration, which decreases the variability of NHGU in response to hyperglycemia and hyperinsulinemia. The dog, as well as the human, exhibits marked variability in hepatic glycogen content during a fast shorter than 40 h (8, 32).

Approximately 16 days before study, each dog underwent a laparotomy for placement of ultrasonic flow probes (Transonic Systems, Ithaca, NY) around the portal vein and the hepatic artery, as well as for insertion of silicone rubber catheters for sampling in a hepatic vein, the portal vein, and a femoral artery and for infusion into a splenic and a jejunal vein, as described in detail elsewhere (25, 29). Criteria for inclusion in the study were as previously described (25, 29).

On the morning of the study, catheters and flow probe leads were exteriorized from their subcutaneous pockets (25, 29). The splenic and jejunal catheters were used for intraportal infusion of insulin (Eli Lilly, Indianapolis, IN), glucagon (Glucagen; Bedford Laboratories, Bedford, OH), and fluvoxamine maleate (Tocris, Ellisville, MO). Angiocaths (Deseret Medical, Sandy, UT) were inserted into three peripheral veins.

Experimental design. Each experiment consisted of a 90-min equilibration period (120–30 min), a 30-min basal period (30–0 min), and a 240-min experimental period (0 to 240 min) divided into two subperiods (P1, 0–90 min; P2, 90–240 min; Fig. 1). At 120 min, a primed continuous infusion of [3-3H]glucose and a continuous infusion of indocyanine green (ICG) dye were begun in all dogs (22). At 0 min, a constant peripheral infusion of somatostatin (0.8 μg·kg⁻¹·min⁻¹; Bachem, Torrance, CA) was begun to suppress endogenous insulin and glucagon secretion. Insulin was infused intraportally at either basal (0.3 μU·kg⁻¹·min⁻¹; Ins groups) or fourfold basal rates (1.2 μU·kg⁻¹·min⁻¹; INS groups) and glucagon (0.55 ng·kg⁻¹·min⁻¹) was replaced intraportally in basal amounts. In addition, a primed continuous variable-rate infusion of 50% dextrose was begun through a peripheral vein to maintain the hepatic glucose load at twofold basal. During P1, all dogs received an intraportal saline infusion. At the end of P1, the Ins and INS groups were each divided into two groups. In Sal-Ins (n = 9) and Sal-INS (n = 8), the intraportal saline infusion continued for the remainder of the study. In

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Fluv-Ins (n = 7) and Fluv-INS (n = 6), fluvoxamine was infused into the portal vein at 2 μg·kg⁻¹·min⁻¹ (a rate that we had previously demonstrated to enhance NHGU under hyperinsulinemic hyperglycemic conditions; Ref. 20) during P2. The intraportal infusion allowed us to target the liver specifically with fluvoxamine. Fluvoxamine was chosen because of its selectivity for the serotonin receptor over other monoamine receptors (35) and its extensive (~45%) first-pass extraction by the liver (37), which reduced the potential for circulating fluvoxamine to impact nonhepatic glucose uptake (non-HGU). In addition, the dosage was kept low (a total of 0.3 mg/kg, compared with a usual daily therapeutic dose of 2–4 mg/kg) to reduce the potential for systemic effects of fluvoxamine.

Femoral artery, portal vein, and hepatic vein blood samples were taken every 15–30 min throughout the study as previously described (25, 29). Arterial blood samples were also taken every 5 min throughout the experimental period to monitor the glucose level (25, 29). After completion of each experiment, the animal was anesthetized, biopsies were rapidly taken from three liver lobes and freeze-clamped with tongs chilled in liquid nitrogen, and the dog was euthanized with an overdose of pentobarbital sodium.

Processing and analysis of samples. Hematocrit; blood glucose, lactate, and glycocol; and plasma glucose, nonesterified fatty acids (NEFA), insulin, glucagon, cortisol, catecholamines, and [3H]glucose were measured as described previously (25, 29, 33). 5-HT concentrations were determined on whole blood by an HPLC-amperometric assay (30) with a coefficient of variance (CV) of 4%, as previously described (22). Hepatic glycogen concentrations were determined by the method of Keppler and Decker (14).

Calculations and data analysis. Hepatic blood flow was measured with ultrasonic flow probes and by use of ICG extraction. The two methods yielded similar results, but the data reported here were calculated with the ultrasonic-determined flows, because their measurement did not require an assumption regarding the relative contribution of arterial and portal flow to total hepatic blood flow.

The rate of glucose delivery to the liver, or hepatic glucose load; net hepatic substrate balance (NHB); net hepatic fractional substrate extraction; net hepatic carbon retention; hepatic sinusoidal insulin and glucagon concentrations; non-HGU; and glucose turnover were calculated as described previously (27). NHB was calculated for both cold (nonradioactive) and [3H]glucose, with the values for [3H]glucose being divided by the inflowing (weighted for the proportion of hepatic flow provided by the hepatic artery and portal vein) tritiated glucose specific activity to yield unidirectional hepatic glucose uptake (33). During the first hour of glucose infusion, the non-HGU was corrected for the glucose required to fill the pool, using a pool fraction of 0.65 (5) and assuming that the volume of distribution for glucose equaled the volume of the extracellular fluid, or ~22% of the dog’s weight (34). For all glucose balance calculations, glucose concentrations were converted from plasma to blood values by using correction factors (ratio of the blood to the plasma concentration) as previously established in our laboratory (11, 12, 29).

Net hepatic glycogen synthesis was determined by subtracting the glycogen concentrations of 11 42-h-fasted dogs that did not undergo study (19) from the final hepatic glycogen concentrations in the dogs studied.

Statistical analysis. All data are presented as means ± SE. Time course data were analyzed with repeated-measures analysis of variance, with post hoc comparisons by Tukey’s test (SigmaStat; Jandel Scientific, Chicago, IL). Statistical significance was accepted at P < 0.05. Values for P1 are means of the last 30 min (60–90 min) when steady-state conditions prevailed. Values for P2 are means of all samples between 120 and 240 min.

RESULTS

5-HT and hormone concentrations. Arterial and portal blood 5-HT concentrations remained no different from basal values in all groups throughout the experimental periods (Table 1). The arterial and hepatic sinusoidal plasma insulin concentrations (Table 1) remained at basal levels in the Ins groups and were approximately fourfold greater in the INS groups during P1–P2. There were no differences in the insulin concentrations in the Sal-Ins and Fluv-Ins groups or between the Sal-INS and Fluv-INS groups. Arterial and hepatic sinusoidal plasma glucagon concentrations were basal and indistinguishable in all groups throughout the experiments (Table 1). The cortisol and catecholamine concentrations remained basal in all groups throughout the experiments (data not shown).

Hepatic blood flow, blood glucose concentrations, and hepatic glucose load. Portal vein blood flow decreased significantly in all groups during P1 as a response to somatostatin infusion and did not change significantly thereafter (Table 2). There was a concomitant increase in hepatic artery flow such that total flow fell by ~10%. There were no significant differences in hepatic artery or portal vein blood flow between Sal-Ins and Fluv-Ins or between Sal-INS and Fluv-INS.

Arterial blood glucose levels in all groups increased from basal values of 4.5 ± 0.1 to ~9.3 ± 0.2 mmol/l during P1 and P2 (Fig. 2). The hepatic glucose loads increased from a basal rate of ~140 to 270 μmol·kg⁻¹·min⁻¹ during P1 and P2 and were not significantly different among the four groups at any time (Fig. 2).

Net hepatic glucose balance and net hepatic fractional glucose extraction. The groups exhibited similar rates of net hepatic glucose output during the basal period. Coincident with the start of the experimental period, they switched from net production to net uptake, with no significant difference in the rates between the Sal-Ins and Fluv-Ins groups, respectively, during P1 (6.3 ± 1.7 and 2.4 ± 2.5 μmol·kg⁻¹·min⁻¹) or P2

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<th>Period</th>
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<th>P2</th>
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<tr>
<td>min:</td>
<td>120</td>
<td>-30</td>
<td>0</td>
</tr>
<tr>
<td>[3H]glucose, indocyanine green dye</td>
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<td></td>
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<td>Somatostatin (peripheral), glucagon (basal, intraportal [po])</td>
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<td>Insulin po 0.3 mU·kg⁻¹·min⁻¹ (ins) OR 1.2 mU·kg⁻¹·min⁻¹ (INS)</td>
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<td></td>
<td></td>
</tr>
<tr>
<td>SAL-ins, n = 9</td>
<td>Saline (SAL) po</td>
<td>OR</td>
<td></td>
</tr>
<tr>
<td>FLUV-ins, n = 7</td>
<td>Fluvoxamine (FLUV) po</td>
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<td></td>
</tr>
<tr>
<td>FLUV-INS, n = 6</td>
<td>0 (saline)</td>
<td>2 μg·kg⁻¹·min⁻¹</td>
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</table>

Fig. 1. Protocol design. For details, see RESEARCH DESIGN AND METHODS.
### Table 1. Serotonin and hormone concentrations

<table>
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<tr>
<th>Parameter and Group</th>
<th>Basal Period</th>
<th>P1</th>
<th>90 min</th>
<th>120 min</th>
<th>180 min</th>
<th>240 min</th>
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<td>Arterial blood 5-HT, µg/l</td>
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<tr>
<td>Sal-Ins</td>
<td>1.033±0.321</td>
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<td>Fluv-Ins</td>
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<tr>
<td>Sal-INS</td>
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<td>831±84</td>
<td>642±165</td>
<td>539±132</td>
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<tr>
<td>Fluv-INS</td>
<td>682±292</td>
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<td>691±295</td>
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<td>Portal blood 5-HT, µg/l</td>
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<td>Sal-Ins</td>
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<td>1.722±0.327</td>
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<td>Sal-INS</td>
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<td>Sal-Ins</td>
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<td>27±3</td>
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<tr>
<td>Fluv-Ins</td>
<td>40±5</td>
<td>26±2</td>
<td>25±3</td>
<td>27±4</td>
<td>26±2</td>
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<td>Sal-INS</td>
<td>51±7</td>
<td>141±14</td>
<td>131±11</td>
<td>149±10</td>
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<td>Fluv-INS</td>
<td>52±10</td>
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<td>134±13</td>
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<td>Hepatic sinusoidal insulin, pmol/l</td>
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<tr>
<td>Sal-Ins</td>
<td>94±12</td>
<td>109±16</td>
<td>92±16</td>
<td>93±7</td>
<td>84±9</td>
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<tr>
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<td>90±15</td>
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<tr>
<td>Sal-INS</td>
<td>146±24</td>
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<td>556±73</td>
<td>478±49</td>
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<td>Fluv-INS</td>
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<td>606±122</td>
<td>517±64</td>
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<td>Arterial plasma glucagon, ng/l</td>
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<td>39±4</td>
<td>38±2</td>
<td>40±4</td>
<td>35±3</td>
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<tr>
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<td>37±4</td>
<td>31±4</td>
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<tr>
<td>Sal-INS</td>
<td>46±6</td>
<td>43±7</td>
<td>40±6</td>
<td>38±4</td>
<td>35±6</td>
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<tr>
<td>Fluv-INS</td>
<td>37±4</td>
<td>35±5</td>
<td>35±4</td>
<td>42±6</td>
<td>31±5</td>
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<tr>
<td>Hepatic sinusoidal glucagon, ng/l</td>
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<tr>
<td>Sal-Ins</td>
<td>45±6</td>
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<tr>
<td>Fluv-Ins</td>
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<td>38±5</td>
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<td>54±7</td>
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<tr>
<td>Fluv-INS</td>
<td>42±4</td>
<td>50±10</td>
<td>50±13</td>
<td>50±9</td>
<td>51±10</td>
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Values are means ± SE. INS, dogs received 4-fold basal intraportal insulin during period 1 (P1) to period 2 (P2); Ins, dogs received basal intraportal insulin; Sal, dogs received intraportal saline infusion during P1–P2; Fluv, dogs received intraportal saline during P1 and fluvoxamine at 2.0 µg·kg⁻¹·min⁻¹ during P2; 5-HT, 5-hydroxytryptamine. There were no significant differences between the Sal-INS (n = 9) and Fluv-INS (n = 7) groups or between the SAL-INS (n = 8) and Fluv-INS (n = 6) groups.

### Table 2. Hepatic blood flow and glucose Rₘ

<table>
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<tr>
<th>Parameter and Group</th>
<th>Basal Period</th>
<th>P1</th>
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<th>120 min</th>
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<td>Heparic artery flow, ml·kg⁻¹·min⁻¹</td>
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<tr>
<td>Sal-Ins</td>
<td>5.9±0.7</td>
<td>6.5±1.1</td>
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<tr>
<td>Fluv-Ins</td>
<td>6.1±0.9</td>
<td>6.6±1.7</td>
<td>7.2±1.7</td>
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<tr>
<td>Sal-INS</td>
<td>6.3±1.0</td>
<td>7.3±1.0</td>
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<tr>
<td>Fluv-INS</td>
<td>4.8±0.7</td>
<td>6.1±1.0</td>
<td>5.3±0.9</td>
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<tr>
<td>Portal vein flow, ml·kg⁻¹·min⁻¹</td>
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<tr>
<td>Sal-Ins</td>
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<tr>
<td>Sal-INS</td>
<td>25.2±2.4</td>
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<td>Glucose Rₘ, µmol·kg⁻¹·min⁻¹</td>
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<td>Fluv-INS</td>
<td>12.5±0.6</td>
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<td>48.4±6.0</td>
<td>58.8±10.5</td>
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Values are means ± SE. For group descriptions, see legend to Table 1. Rₘ, rate of disappearance. There were no significant differences between the Sal-Ins (n = 9) and Fluv-Ins (n = 7) groups or between the Sal-INS (n = 8) and Fluv-INS (n = 6) groups.
(8.4 ± 1.4 and 6.9 ± 2.3 μmol·kg⁻¹·min⁻¹; Fig. 3). Unidirectional hepatic glucose uptake (tracer determined) did not differ at any time between groups (data not shown). Net hepatic fractional extraction of glucose was similarly unaffected by fluvoxamine (0.029 ± 0.006 and 0.026 ± 0.010 during P2 in the Sal-INS and Fluv-INS groups, respectively).

NHGU in the Sal-INS and Fluv-INS groups, respectively, averaged 10.7 ± 2.2 and 12.7 ± 2.3 μmol·kg⁻¹·min⁻¹ during P1 [not significant (NS); Fig. 3]. Subsequently the rate of NHGU remained relatively stable in Sal-INS (13.3 ± 2.3 μmol·kg⁻¹·min⁻¹ during P2) but increased significantly in Fluv-INS (20.9 ± 3.1 μmol·kg⁻¹·min⁻¹, P < 0.05 between groups). Unidirectional hepatic glucose uptake was also enhanced by fluvoxamine infusion during P2 (14.2 ± 3.7 vs. 24.6 ± 2.8 μmol·kg⁻¹·min⁻¹ in Sal-INS and Fluv-INS, respectively; P < 0.05). Additionally, net hepatic fractional extraction of glucose during P2 was greater in Fluv-INS than in Sal-INS, averaging 0.085 ± 0.009 vs. 0.056 ± 0.009 (P < 0.05).

Glucose infusion rates, non-HGU, glucose rate of appearance, and glucose Rg. The glucose infusion rates (GIRs) in Sal-INS and Fluv-INS did not differ significantly during either P1 (19.6 ± 3.6 vs. 15.9 ± 4.0 μmol·kg⁻¹·min⁻¹) or P2 (21.4 ± 2.9 vs. 18.4 ± 3.3 μmol·kg⁻¹·min⁻¹). Non-HGUs were not different in Sal-INS and Fluv-INS, respectively, during P1 (11.1 ± 2.9 and 13.5 ± 2.5 μmol·kg⁻¹·min⁻¹) or P2 (12.2 ± 2.2 vs. 11.8 ± 1.1 μmol·kg⁻¹·min⁻¹). Endogenous glucose rate of appearance (Rg) was suppressed by hyperglycemia in both groups to ~35% of basal levels and did not differ between groups at any time. Glucose Rg increased in response to hyperglycemia during P1 and then stabilized (Table 2; NS between groups).

In Sal-INS and Fluv-INS, GIRs averaged 38.9 ± 4.3 and 30.9 ± 2.1 μmol·kg⁻¹·min⁻¹, respectively, during P1 (NS), and they continued to increase during P2, reaching 60.6 ± 7.7 and 49.5 ± 6.8 μmol·kg⁻¹·min⁻¹ by 240 min (P = 0.12 between groups; Fig. 4). Non-HGU was significantly greater in Sal-INS than in Fluv-INS during P1 (27.5 ± 3.8 and 16.7 ±
0.6 μmol·kg⁻¹·min⁻¹, respectively; *P < 0.05). It increased over time in both groups but appeared to be blunted in Fluv-INS (rates averaged 38.4 ± 5.4 and 20.0 ± 1.3 μmol·kg⁻¹·min⁻¹ during P2, *P < 0.01 between groups). Endogenous glucose R₈ was suppressed in the two groups to ~15% of basal levels in response to hyperglycemia and hyperinsulinemia, and the rates did not differ between groups at any time. Glucose R₄ increased nearly fivefold between the basal period and the end of study in both groups. R₄ tended to be higher in Sal-INS during both P1 and P2 (*P = 0.4 and 0.2, respectively; Table 2).

Lactate metabolism, net hepatic carbon retention, and hepatic glycogen synthesis. The arterial blood lactate concentrations increased significantly during P1 in all groups. The levels in Sal-Ins and Fluv-Ins did not differ at any time, nor did the concentrations in Sal-INS and Fluv-INS differ from each other (Table 3). All groups exhibited net hepatic lactate uptake in the basal period, shifting to net hepatic lactate output with the onset of the experimental period. The rates did not differ at any time in the two Ins groups or in the two INS groups.

Net hepatic carbon retention did not differ in Sal-Ins and Fluv-Ins groups at any time, but it was enhanced (*P < 0.05) in Fluv-INS vs. Sal-INS during P2 (18.5 ± 2.7 vs. 12.2 ± 1.9 μmol·kg⁻¹·min⁻¹; Fig. 3). Net hepatic glycogen synthesis over the 4-h experimental period, calculated from the hepatic glycogen concentrations, was greater in Fluv-INS than in Sal-INS (19.9 ± 6.3 and 15.4 ± 6.3 mg/g liver, respectively), although this did not reach statistical significance (*P = 0.2).

Glycerol and NEFA metabolism. Arterial blood glycerol concentrations were reduced ~15–40% by hyperglycemia and 50% by the combination of hyperglycemia and hyperinsulinemia, and net hepatic glycerol uptake was reduced ~25–45% by hyperglycemia and ~50% by hyperglycemia plus hyperinsulinemia (Table 3). Fluvoxamine had no effect on the degree of suppression in either the presence or absence of hyperinsulinemia. Arterial NEFA concentrations and net hepatic NEFA uptake changed in a manner similar to glycerol, and there were no differences between Sal-Ins and Fluv-Ins or between Sal-INS and Fluv-INS (Table 3).

DISCUSSION

Under conditions of basal insulin and hyperglycemia, intraportal fluvoxamine infusion had no significant impact on NHGU. The GIRs, glucose R₄, non-HGUs, and net hepatic carbon retention were similarly unaffected by fluvoxamine in the presence of basal insulin. However, in the presence of hyperinsulinemia, fluvoxamine was associated with rates of NHGU and net hepatic fractional extraction of glucose >50% greater than in the saline-infused control animals (*P < 0.05 for both parameters). We were unable in our previous study to determine the time course of the response to fluvoxamine because that study employed three successive infusion rates of the SSRI (20). However, the current data indicate that the effect is relatively rapid, with a significant enhancement of NHGU evident within 45 min (Fig. 3).

The interaction between insulin and fluvoxamine also resulted in enhancement of hepatic glycogen storage, as evidenced by the significant enhancement of net hepatic carbon retention and the tendency toward greater net hepatic glycogen synthesis. Although the enhancement of net hepatic glycogen synthesis was similar in magnitude to the enhancement of net hepatic carbon retention in Fluv-INS vs. Sal-INS, it reflected glycogen accretion during both P1 and P2 rather than P2 alone. The 150-min fluvoxamine infusion period was apparently not long enough to allow the SSRI to bring about a significant increase in the hepatic glycogen measurement.

There are several possible ways in which fluvoxamine (i.e., 5-HT) could enhance NHGU and hepatic glucose storage. First, it could act independently of insulin. Because that would imply that fluvoxamine could promote NHGU in the absence of insulin, and we failed to observe enhanced NHGU at basal insulinemia, this possibility seems unlikely. Second, fluvoxamine could enhance hepatic insulin sensitivity via a direct action. In this regard, more than one type of 5-HT receptor is known to be expressed in the liver (10, 16), providing a target through which fluvoxamine might act. Third, fluvoxamine might act indirectly on the liver by targeting nonhepatic tissues, bringing about an alteration in substrate supply from peripheral tissues or a change in neural or chemical signaling that in turn altered hepatic metabolism. Although we attempted to target the SSRI to the liver by choosing an agent with a high first-pass extraction and infusing it directly into the portal circulation, it is possible that sufficient fluvoxamine escaped the splanchic bed to exert effects on remote sites. There were
those previously treated with streptozotocin to create insulin deficiency (39, 40). Moreover, subcutaneous insulin (1 U/kg) significantly enhanced the leptin response of normal mice given an intraperitoneal 5-HTP injection (38). Both the insulin and the leptin responses could be blocked by pretreatment with an intraperitoneal injection of benserazide, which prevents the formation of 5-HT from 5-HTP (40, 41). Leptin receptors are expressed in the liver (13, 15, 28), and 2 days of intraperitoneal leptin administration increased hepatic insulin sensitivity and insulin receptor activation in vivo in mice (15). Thus stimulation of leptin release in response to fluvoxamine is a possible mechanism explaining our findings, but we cannot reach any conclusion about this from our current work.

Interestingly, glucagon concentrations did not decline during the experimental period in the groups receiving fluvoxamine (Table 1). We have consistently observed that plasma glucagon concentrations decline ~15% during an infusion of a few hours duration (see, for example, Refs. 12, 20, 33), presumably because of the tendency of molecules of the hormone to aggregate in the infusate and become less available (18). We postulate that some chemical or neural signal induced by 5-HT enhanced hepatic insulin sensitivity.

With regard to the last possibility, intraperitoneal injection of 5-hydroxytryptophan (5-HTP), the immediate precursor of 5-HT, brought about hyperleptinemia in normal mice but not in those previously treated with streptozotocin to create insulin deficiency (39, 40). Moreover, subcutaneous insulin (1 U/kg) significantly enhanced the leptin response of normal mice given an intraperitoneal 5-HTP injection (38). Both the insulin and the leptin responses could be blocked by pretreatment with an intraperitoneal injection of benserazide, which prevents the formation of 5-HT from 5-HTP (40, 41). Leptin receptors are expressed in the liver (13, 15, 28), and 2 days of intraperitoneal leptin administration increased hepatic insulin sensitivity and insulin receptor activation in vivo in mice (15). Thus stimulation of leptin release in response to fluvoxamine is a possible mechanism explaining our findings, but we cannot reach any conclusion about this from our current work.

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Table 3. Arterial lactate, glycerol, and NEFA concentrations and net hepatic balances

<table>
<thead>
<tr>
<th>Parameter and Group</th>
<th>Basal</th>
<th>P1</th>
<th>P2</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>90 min</td>
<td>120 min</td>
</tr>
<tr>
<td>Arterial blood lactate, μmol/l</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sal-Ins</td>
<td>441±67</td>
<td>933±112</td>
<td>1,062±127</td>
</tr>
<tr>
<td>Fluv-Ins</td>
<td>669±107</td>
<td>976±93</td>
<td>1,069±60</td>
</tr>
<tr>
<td>Sal-INS</td>
<td>448±53</td>
<td>922±100</td>
<td>886±84</td>
</tr>
<tr>
<td>Fluv-INS</td>
<td>542±132</td>
<td>1,093±126</td>
<td>984±122</td>
</tr>
<tr>
<td>Net hepatic lactate balance, μmol/kg/min</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sal-Ins</td>
<td>−7.3±0.9</td>
<td>5.7±1.8</td>
<td>5.0±1.2</td>
</tr>
<tr>
<td>Fluv-Ins</td>
<td>−9.7±2.1</td>
<td>2.4±3.2</td>
<td>1.8±2.6</td>
</tr>
<tr>
<td>Sal-INS</td>
<td>−7.7±1.5</td>
<td>4.2±1.6</td>
<td>3.2±1.5</td>
</tr>
<tr>
<td>Fluv-INS</td>
<td>−5.2±1.6</td>
<td>6.1±0.5</td>
<td>4.1±1.0</td>
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<td>Arterial blood glycerol, μmol/l</td>
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<tr>
<td>Sal-Ins</td>
<td>86.1±6.2</td>
<td>50.3±9.8</td>
<td>44.8±6.4</td>
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<tr>
<td>Fluv-Ins</td>
<td>81.6±6.0</td>
<td>69.4±13.3</td>
<td>59.5±11.1</td>
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<tr>
<td>Sal-INS</td>
<td>89.4±10.9</td>
<td>44.3±9.3</td>
<td>39.8±8.2</td>
</tr>
<tr>
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<td>81.2±4.2</td>
<td>39.9±9.2</td>
<td>39.2±13.0</td>
</tr>
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<td>Net hepatic glycerol uptake, μmol/kg/min</td>
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<td></td>
<td></td>
</tr>
<tr>
<td>Sal-Ins</td>
<td>1.9±0.2</td>
<td>1.0±0.2</td>
<td>0.8±0.1</td>
</tr>
<tr>
<td>Fluv-Ins</td>
<td>2.2±0.2</td>
<td>1.6±0.2</td>
<td>0.9±0.4</td>
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<tr>
<td>Sal-INS</td>
<td>1.8±0.4</td>
<td>0.8±0.3</td>
<td>0.6±0.2</td>
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<tr>
<td>Fluv-INS</td>
<td>2.1±0.3</td>
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<td>Arterial plasma NEFA, μmol/l</td>
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<td></td>
</tr>
<tr>
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<td>916±46</td>
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<td>353±58</td>
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<td>796±76</td>
<td>555±128</td>
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<td>Sal-INS</td>
<td>820±174</td>
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<tr>
<td>Fluv-INS</td>
<td>1,020±133</td>
<td>246±61</td>
<td>212±55</td>
</tr>
<tr>
<td>Net hepatic NEFA uptake, μmol/kg/min</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sal-Ins</td>
<td>2.9±0.2</td>
<td>1.0±0.2</td>
<td>1.0±0.2</td>
</tr>
<tr>
<td>Fluv-Ins</td>
<td>3.0±0.5</td>
<td>2.0±0.7</td>
<td>1.5±0.5</td>
</tr>
<tr>
<td>Sal-INS</td>
<td>2.9±0.6</td>
<td>0.1±0.3</td>
<td>0.1±0.2</td>
</tr>
<tr>
<td>Fluv-INS</td>
<td>3.1±0.3</td>
<td>0.7±0.3</td>
<td>0.5±0.2</td>
</tr>
</tbody>
</table>

Values are means ± SE; n = 9, 7, 8, and 6 for Sal-Ins, Fluv-Ins, Sal-INS, and Fluv-INS, respectively, except for nonesterified fatty acid (NEFA) concentrations and net hepatic uptake, where n = 5 for Sal-INS. For group descriptions, see legend to Table 1. Negative values indicate net hepatic uptake. There were no significant differences between Ins groups or between INS groups.
seen only with longer infusions achieving higher levels of 5-HT, since it did appear to be present during intraportal infusion of 5-HT over 3 h (22). It is unlikely that fluvoxamine/5-HT stimulates glucagon secretion, both because we used somatostatin at a rate that has proven to control pancreatic endocrine secretion under a variety of stimulatory conditions (e.g., Refs. 7, 21) and because most data indicate that 5-HT and 5-HT receptor agonists have no effect (36) or an inhibitory effect (1, 4, 17) on glucagon release in normal humans or animals. Thus it appears that fluvoxamine or endogenous 5-HT might slow the clearance of glucagon. Nevertheless, the failure of glucagon concentrations to decline in Fluv-INS in parallel with the levels in Sal-INS could not have been responsible for the enhancement of NHGU in the Fluv-INS group. If anything, a relative elevation of glucagon should reduce NHGU (9).

Non-HGU (primarily skeletal muscle glucose uptake under hyperinsulinemic conditions; Ref. 6) was significantly reduced during P2 in Fluv-INS vs. Sal-INS, but it also tended to be lower in the Fluv-INS group during P1. The GIR and glucose Rg also tended to be lower in Fluv-INS than in Sal-INS throughout P1 and P2. The reduction in non-HGU and the tendency toward reduction in the GIR and glucose Rg in P2 may have been related directly or indirectly to administration of fluvoxamine (20). We have previously shown that there is substantial reciprocity between NHGU and non-HGU such that, under a variety of circumstances, when NHGU increases there is a compensatory decrease in non-HGU (e.g., Refs. 2, 6, 20, 23). The current data are consistent in this respect with our previous findings. However, because of the tendency of nonhepatic and whole body glucose metabolism (i.e., non-HGU, GIR, glucose Rg) to differ during P1, we cannot rule out a slight preexisting peripheral insulin resistance in Fluv-INS relative to the Sal-INS group. There is no evidence that the possible insulin resistance affected the liver, however, and therefore it had minimal impact on our findings.

In conclusion, intraportal infusion of fluvoxamine enhanced NHGU under hyperglycemic hyperinsulinemic conditions but not under euglycemic conditions. Enhancement of NHGU was accompanied by stimulation of net hepatic carbon retention and also by a tendency toward an offsetting decrease in non-HGU. Thus total body glucose Rg was not significantly different in saline- and fluvoxamine-infused dogs. The apparent interaction between insulin and fluvoxamine suggests that SSRIs or related compounds that could be targeted to the hepatocyte might have a role in reducing postprandial hyperglycemia in individuals with impaired glucose tolerance or type 2 diabetes.

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