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Research Article

The hepatic and renal metabolism of somatostatin-like immunoreactivity (SLI) was assessed simultaneously in an in vivo dog model. The hepatic extraction of this peptide was 29.4 +/- 2.3% and was similar for endogenous and infused exogenous SLI. The renal extraction was 62.3 +/- 5%. The renal clearance of SLI was significantly greater than that of inulin indicating that the peptide is handled by peritubular uptake from postglomerular blood in addition to glomerular filtration. In both organs SLI extraction was not saturable even at arterial concentrations in excess of 100 times physiological range. The overall metabolic clearance rate of SLI was 19.7 +/- 1.6 ml/kg per minute of which 32.7 +/- 4.6% was contributed by hepatic and 37 +/- 4.9% by renal uptake mechanisms. The plasma half disappearance time of exogenously infused SLI was 1.9 +/- 0.3 min. The studies indicate that in the dog, the liver and kidney are both major sites of SLI metabolism, together accounting for 70.0 +/- 8.7% of the metabolic clearance of the peptide.

Hepatic and Renal Metabolism of Somatostatin-like Immunoreactivity

SIMULTANEOUS ASSESSMENT IN THE DOG

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ABSTRACT The hepatic and renal metabolism of somatostatin-like immunoreactivity (SLI) was assessed simultaneously in an in vivo dog model. The hepatic extraction of this peptide was 29.4±2.3% and was similar for endogenous and infused exogenous SLI. The renal extraction was 62.3±5%. The renal clearance of SLI was significantly greater than that of inulin indicating that the peptide is handled by peritubular uptake from postglomerular blood in addition to glomerular filtration. In both organs SLI extraction was not saturable even at arterial concentrations in excess of 100 times physiological range. The overall metabolic clearance rate of SLI was 19.7±1.6 ml/kg per minute of which 32.7±4.6% was contributed by hepatic and 37±4.9% by renal uptake mechanisms. The plasma half disappearance time of exogenously infused SLI was 1.9±0.3 min.

The studies indicate that in the dog, the liver and kidney are both major sites of SLI metabolism, together accounting for 70.0±8.7% of the metabolic clearance of the peptide.

INTRODUCTION

Since its discovery in 1973 (1) somatostatin has been the object of intense scientific interest and study. As a result of these investigations a great deal of information is available regarding the distribution of this peptide in various parts of the nervous system (1–4), gut, and pancreas (5), as well as its effect on the secretion of various hormones (1, 6, 7). Exogenous somatostatin is rapidly metabolized in man, the metabolic clearance rate and plasma half life of infused somatostatin-like immunoreactivity (SLI)[1] being 28.7 ml/kg per min and 1.9 min, respectively (8). The sites of SLI metabolism, however, have not been studied in detail, although there is some evidence that both liver and kidney may be important in this regard. Shapiro et al. (9) measured a significant renal arteriovenous difference in SLI in the rat and suggested that the kidney is an important site of its degradation. This conclusion was further supported by the observation that the metabolic clearance rate of SLI was significantly prolonged in uremia (8). A number of workers (10–12) have observed portal peripheral SLI gradients, a finding that suggests that the liver may be an important site of SLI metabolism.

We have described (13) an in vivo dog model for the estimation of peptide hormone metabolism. The present experiments were designed to study the in vivo metabolism of SLI and to assess the relative contribution of the liver and kidney to its metabolic clearance. The results indicate that both the liver and kidney are major sites of SLI degradation, together accounting for 70% of its overall metabolic clearance.

METHODS

18 mongrel dogs (15–28 kg) of both sexes were studied. Animals were observed for 1 wk before surgery to evaluate their general health status. After an overnight fast, anesthesia was induced with 30 mg/kg i.v. sodium pentobarbital (Diabutal, Diamond Laboratories Inc., Des Moines, Iowa) and maintained with boluses of 30–60 mg when necessary. The animals were intubated and artificially ventilated with a Bird pressure cycled ventilator with 40% oxygen supplementation.

Surgical procedures. The portal vein and hepatic artery were exposed, and electromagnetic flow probes (Gould Statham SP 2202) were placed around each vessel (Fig. 1). The portal vein probe was placed 2 cm proximal to the bifurcation of this vessel into right and left portal trunks. The hepatic artery probe was positioned 2 cm distal to the origin of this vessel from the celiac trunk. Probes of appropriate size were chosen in each experiment to produce ~30 and 15% constriction of the portal vein and hepatic artery, respectively, as recommended by Gordon et al. (14). Before each experiment the flow probes were calibrated in saline and re-

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[1] *Abbreviations used in this paper*: BSA, bovine serum albumin; MCR, metabolic clearance rate; PAH, para-aminohippuric acid; SLI, somatostatin-like immunoreactivity.

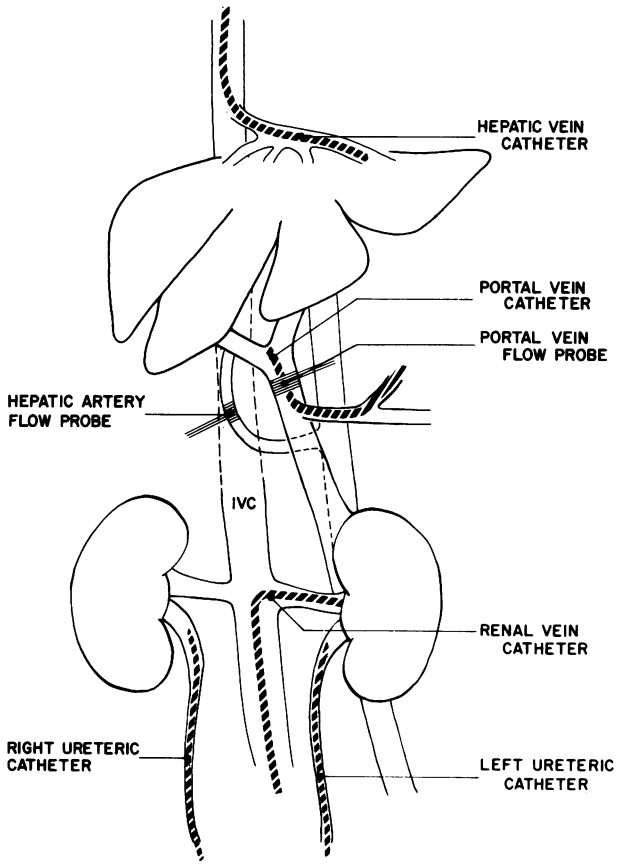

FIGURE 1 Schematic representation of the position of the sampling catheters and flow probes.

checked after their placement to ensure that occlusion of the vessels recorded zero flows. The gastroduodenal branch of the hepatic artery was identified and ligated because this artery contributes to hepatic artery blood flow, while bypassing the liver. The gastroduodenal vein was also ligated as it entered the portal vein above the position where portal blood flow is measured. Bilateral ureteric catheters were inserted to ensure complete and accurate urine collection. End sampling catheters were placed in the portal vein, hepatic vein, left renal vein, and femoral artery in 10 animals (Fig. 1). In eight additional animals only portal and hepatic vein and femoral artery catheters were placed. The portal vein catheter was positioned so that its tip lay at the bifurcation of this vessel in the porta hepatis. The peripheral arterial catheter was inserted through a small branch of the left femoral artery into the common femoral artery. The hepatic vein catheter was passed through the right jugular vein into the left common hepatic vein 1–2 cm from the wedged position. The renal vein catheter was advanced through the left femoral vein into the left renal vein so that its tip lay as close to the renal hilum as possible. Two catheters were inserted into the left jugular vein, one for monitoring central venous pressure, and the other for infusion of somatostatin, insulin, glucagon, para-aminohippuric acid (PAH), and inulin.

The catheter positions were verified by a number of independent methods. The portal and renal catheters were inserted under direct vision and their position checked by palpation. The position of the hepatic vein catheter was confirmed radiologically by injection of 5 ml renografin. Because all dogs were studied in the fasted state, the presence of a 10–20 mg/100 ml glucose gradient between portal and hepatic veins provided further confirmation of the location of the catheter in the hepatic vein. In addition all catheter positions were verified at autopsy on completion of each experiment.

Physiological monitoring. Special care was taken to maintain the vital functions of the dog within normal limits for the duration of the experiment. Arterial pulse and blood pressure were monitored continuously via the femoral artery catheter by means of a "slope mobile" monitor (Honeywell, Inc., Test Instruments Div., Denver, Colo.). Intravenous fluid (0.45% saline) was infused to maintain a stable pulse and blood pressure, central venous pressure of 4–8 cm water, and urine output of at least 20 ml/h. The rectal temperature was monitored by means of a Tele-thermometer (Yellow Springs Instrument Co., Yellow Springs, Ohio) and maintained within normal limits with the aid of a heating blanket. Arterial blood gases were measured at the end of the surgical procedure and at least twice during the subsequent experiment (Radiometer ABL, Copenhagen). The ventilator settings were adjusted to maintain normal arterial pH. The hematocrit was measured at the start of surgery and during each infusion period.

Preparation of infusates. A solution of cyclic somatostatin (Bachem, Inc., Torrance, Calif.) was prepared in a solution containing 0.9% saline and 3% bovine serum albumin (BSA) immediately before each experiment and kept at 4°C until used. Glucagon (Novo, Inc., Copenhagen) was diluted in 3% BSA saline immediately before each experiment and kept at 4°C until used. Insulin (Pork regular U 100, Eli Lilly & Co., Indianapolis, Ind.) was infused together with the glucagon. 5 ml Trasylol (FBA Pharmaceuticals, Inc., New York) was added to each 100 ml of infusate (500 kallikrein inhibitory units/ml).

PAH (Sigma Chemical Co., St. Louis, Mo.) and inulin (Fisher Scientific Co., Fair Lawn, N. J.) were dissolved in 0.9% saline before each experiment.

Experimental protocol. (Fig. 2) 18 animals were studied. Metabolism of endogenously secreted SLI as well as infused somatostatin was measured in nine dogs. In nine additional animals the metabolism of SLI was assessed during constant infusion of exogenous somatostatin. After catheter placement, animals were allowed to stabilize for 30 min. During this stabilization period, following priming injections, PAH and inulin were administered at constant rates (constant infusion pump, model 975, Harvard Apparatus Co., Inc., S. Natick, Mass.) to achieve arterial plasma concentrations of ~1.5 and 50 μg%, respectively. Thereafter there was a 45-min period for assessment of endogenous SLI metabolism. Blood samples were drawn at 35, 40, and 45 min. Flow probe readings were taken at each sampling time. After the period of evaluation of endogenous metabolism, infusions of exogenous somatostatin were begun via the left jugular venous catheter. At the start of each infusion period an appropriate priming dose of somatostatin (1–100 μg) was administered intravenously. Samples for SLI were drawn from all four vessels at the 35-, 40-, and 45-min time points, and urine was collected during this 10-min period. Samples for PAH and inulin were drawn at 35 and 45 min. Samples for insulin and glucagon were drawn at each time from the femoral artery. A second exogenous infusion period was carried out in all experiments, and in 12 dogs a third infusion period was added. The dose of somatostatin infused varied from 1 to 800 ng/kg per min. When somatostatin doses > 10 ng/kg per min were used,

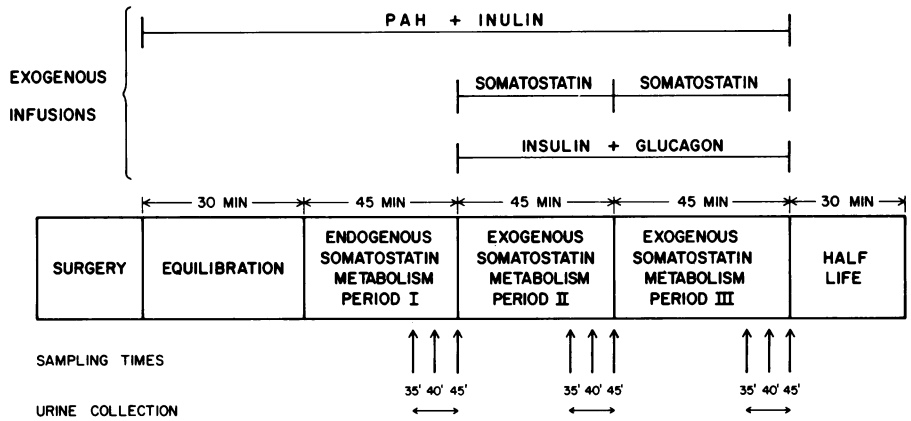

FIGURE 2 The experimental protocol used in this study.

insulin and glucagon were simultaneously infused at rates to replace basal levels of these hormones (insulin 0.2 mU/kg per min, glucagon 1 ng/kg per min). The sampling time points were chosen on the basis of preliminary experiments, which revealed that consistent, steady-state SLI levels had been obtained 35 min into each period. An infusate sample was taken from the infusion catheter at the point where it entered the animal at the start and end of each infusion period, and these two values were used for calculation of the infusion rate. The plasma half disappearance time ($t_{1/2}$) was calculated from the rate of fall of plasma SLI after stopping the somatostatin infusion.

Sample collection. The SLI and glucagon samples were collected in chilled tubes at 4°C containing 500 kallikrein inhibitory units of Trasylol (FBA Pharmaceuticals) and 1.2 mg/ml EDTA. Plasma was separated at 4°C and stored at −20°C until assayed. Insulin samples were allowed to clot at room temperature and after centrifugation at 4°C, the serum was frozen as for glucagon. Urine was collected in chilled plastic tubes at 4°C, frozen immediately, and stored at −20°C until assayed.

Assay techniques. SLI radioimmunoassay was performed as described (15) in 0.1 M phosphate-0.15 M saline buffer containing 0.05 M disodium EDTA and 0.5% crystalline BSA (final pH 5.6). The antibody (rabbit antisomatostatin-hemocyanin; final concentration 1:75,000) is directed towards the central portion of somatostatin, and recognizes molecules with N-terminal modifications with equal affinity to that of somatostatin (12). [^{125}I-Tyr^1]somatostatin was purified by CM52 (Whatman, Inc., Clifton, N. J.) ion exchange chromatography. A double antibody technique was used to separate bound from free peptide. The sensitivity of the assay as assessed by the least amount of somatostatin causing significant displacement ($P < 0.05$) of [^{125}I-Tyr^1]somatostatin from antibody was 15–20 pg/ml (1.5–2 pg/tube). The interassay variation was 12% and the intraassay variation was 9%. Urinary SLI was measured as outlined (16).

The potency of Trasylol and EDTA in overcoming the degradation of [^{125}I-Tyr^1]somatostatin in plasma has been documented (17). Tracer degradation in this radioimmunoassay is prevented by the combined effects of the acid pH of the assay buffer plus trasylol and EDTA [0.05 m] (18). In these studies, incubation damage to [^{125}I-Tyr^1]somatostatin by plasma was assessed by paper chromatoelectrophoresis (15) and immunoprecipitation (17). Under the assay conditions described there was no evidence of tracer degradation in plasma in excess of that seen in the presence of buffer alone, which was negligible. Recovery of cyclic somatostatin added to dog plasma and incubated at 4°C overnight was >85%.

The glucagon radioimmunoassay was performed with 30K antiserum (obtained from Dr. Unger, University of Texas, Southwestern Medical School, Dallas, Tex.) as described (19) using charcoal for separation of bound and free hormone. Insulin radioimmunoassay was performed as described (20). PAH and inulin concentrations in blood and urine were measured by a Technicon autoanalyzer (Technicon Instruments Corp., Tarrytown, N. Y.).

Gel filtration. Representative plasma samples obtained from all vessels during infusion of exogenous somatostatin as well as portal venous plasma samples obtained before somatostatin infusion were applied to a 50 × 1-cm Biogel P-10 column (Bio-Rad Laboratories, Richmond, Calif.) and eluted under gravity at 4°C with the radioimmunoassay buffer. The elution patterns of [^{125}I]gammaglobulin (void volume), Na^{125}I (salt peak), and synthetic somatostatin were used to calibrate the column.

Data analysis. The hepatic extraction of SLI was calculated according to the formula:

$$\text{Hepatic extraction \%} = \frac{\text{hepatic delivery rate} - \text{hepatic removal rate}}{\text{hepatic delivery rate}} \times 100.$$

Hepatic delivery rate (μg/min) a + b: a = hepatic artery plasma flow × femoral artery SLI; b = portal vein plasma flow × portal vein SLI. Hepatic removal rate (μg/min) = hepatic vein plasma flow × hepatic vein SLI. Hepatic vein plasma flow portal vein + hepatic artery plasma flows. Hepatic uptake (μg/min) = hepatic delivery − hepatic removal. Hepatic clearance (ml/kg/min) = hepatic extraction × hepatic plasma flow × 1/body wt (kg).

Metabolic clearance rates (MCR) were measured by the constant infusion technique and calculated from the following formula:

$$\text{MCR}_{(\text{SLI})}(\text{ml/kg/min}) = \frac{\text{SLI infusion rate (ng/min)}}{\text{steady-state SLI level (ng/ml)} - \text{preinfusion SLI (ng/ml)}} \times \frac{1}{\text{wt(kg)}}.$$

Renal extraction and clearance rates and renal function data were calculated as reported (21). SLI extraction and clearance rates in each experimental period were calculated from the values at the individual sampling times. The data in different experimental periods were used to analyze relationships between hepatic and renal metabolism saturability and differences in the handling of endogenous and exogenously infused hormone. In addition a single value for organ extraction, clearance, and MCR was derived for each animal from the value in the individual experimental periods. These data were used to calculate overall rates of SLI extraction, clearance, and MCR.

Statistical methods. All results are expressed as mean ±SEM. Statistical significance of differences was assessed by paired or nonpaired two-tailed Student's t test where applicable, P values <0.05 being considered significant.

RESULTS

Hepatic metabolism of SLI. Table I shows the hepatic extraction of SLI in the exogenous and endogenous periods in each of the 18 dogs studied. Hepatic extraction of SLI during exogenous somatostatin infusion (29.3±2.1% [n = 18]) was similar to that of endogenous secreted peptide (28.0±4.9% [n = 9]). The overall hepatic extraction of endogenous and exogenous SLI was 29.4±2.3% (n = 18). A significant linear relationship (Fig. 3) was observed between the amount of SLI reaching the liver and the amount extracted over a wide range of arterial SLI concentrations (0.185–43.510 ng/ml). The percentage of hepatic extraction of SLI was similar at physiologic and pharmacologic levels. These data indicate that the hepatic extraction of SLI is nonsaturable at levels extending well beyond the physiological range. The hepatic plasma flow was 400±26 ml/min (n = 18). The hepatic clearance was 5.4±0.6 ml/kg per min (n = 18), with no difference between the hepatic clearance of exogenous (5.2±0.5 ml/kg per min; n = 18) and endogenous (5.4±1.2 ml/kg per min; n = 9) SLI.

TABLE I
Hepatic Extraction of Endogenous and Infused Exogenous SLI in 18 Dogs

Dog No.	Hepatic extraction	
	Exogenous SLI	Endogenous SLI
	%	
1	26.6	23.6
2	24.2	21.2
3	28.2	16.3
4	18.8	22.6
5	17.2	12.9
6	27.4	30.6
7	41.4	20.1
8	30.2	49.9
9	35.5	54.4
10	24.9	
11	39.1	
12	24.7	
13	18.1	
14	49.5	
15	30.2	
16	39.1	
17	33.3	
18	18.9	

In dogs 1–9 the hepatic extraction of SLI after exogenous somatostatin infusion (27.7±2.5%) and endogenously secreted SLI (28.0±4.9%) was measured. In dogs 10–18 hepatic extraction was measured only after exogenous infusion (30.9 ±3.5%). The overall hepatic extraction in all 18 dogs after exogenous somatostatin infusion was 29.3±2.1%. None of these differences was statistically significant.

Renal metabolism of SLI. The renal extraction of exogenously infused SLI was 62.3±5% (n = 10) and remained stable over a wide range of circulating levels of the peptide. The amount of SLI extracted by the kidney also increased in a linear fashion as the amount delivered to the kidney increased (Fig. 4). Renal extraction of SLI is therefore also nonsaturable over a wide range of arterial SLI levels.

The renal plasma flow was 240±21 ml/min. The clearance of inulin was 3.0±0.5 ml/kg per min, and its renal extraction was 26.7±3.6% (n = 10). The renal organ clearance of SLI was 7.0±1.1 ml/kg per min (n = 10). This was significantly greater than inulin clearance ($P < 0.002$). The urinary clearance of SLI was assessed in six experimental periods in three dogs. The levels were 26±7 pg/ml, which are negligible, representing <1% of the renal organ clearance of the peptide. These data suggest that renal extraction of SLI involves, in addition to filtration, uptake of the peptide from postglomerular peritubular blood. Furthermore, the negligible urinary excretion of intact SLI indicates that hormone removed from the renal circulation is degraded by the kidney.

MCR of SLI. The MCR, 19.7±1.6 ml/kg per min (n = 17), was unchanged over the above range in arterial concentrations. In 10 animals in which metabolic, hepatic, and renal clearance rates were simultaneously measured, the contributions of the liver and the kidney to the overall metabolic clearance of SLI were calculated. In these animals, MCR was 20.7±2.5, hepatic clearance 6.1±0.7, and renal clearance 7.0±1.1 ml/kg per min. Thus hepatic clearance constituted 32.7±4.6% and renal clearance 37.4±4.9% of the MCR. The combined hepatic and renal contribution to MCR was 70.0±8.7%. The plasma $t_{1/2}$ of SLI was 1.9±0.3 min.

Chromatographic profiles of SLI. The pattern of SLI in the column fractions was similar in all four ves-

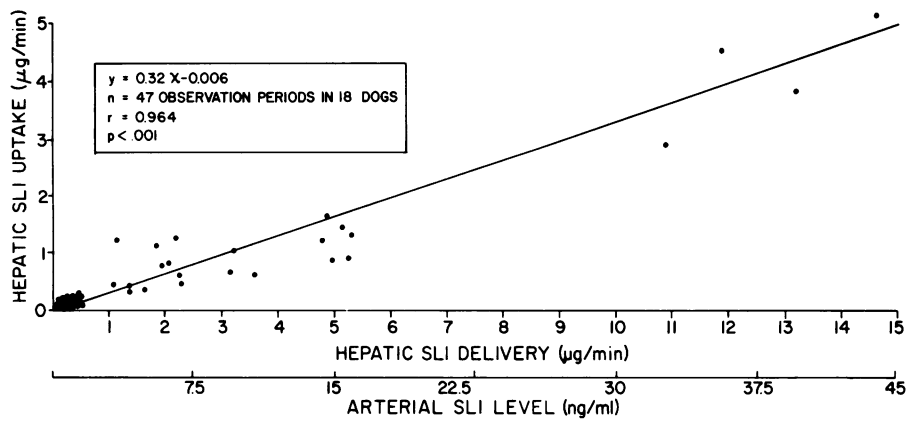

FIGURE 3 The relationship between hepatic SLI delivery (μg/min) and hepatic SLI uptake (μg/min). Hepatic plasma flow is variable and therefore similar delivery rates can occur at different arterial levels. The approximate arterial level corresponding to a given delivery is shown below the abscissa for reference.

sels both before and after somatostatin infusion. Only a small fraction of the immunoreactivity (1.2±0.2%) eluted in the void volume of the column, the rest coeluting with synthetic somatostatin. Chromatographic profiles of plasma samples at pH 7.8 and pH 5.6 were very similar with virtually all the SLI coeluting with synthetic somatostatin.

DISCUSSION

The animal model used in the present experiments has been used in our laboratory for the study of insulin and glucagon metabolism (13). In these studies the hepatic extraction of insulin was found to be 46.8±4.1% and its renal extraction 34.0±4.9%. These values are

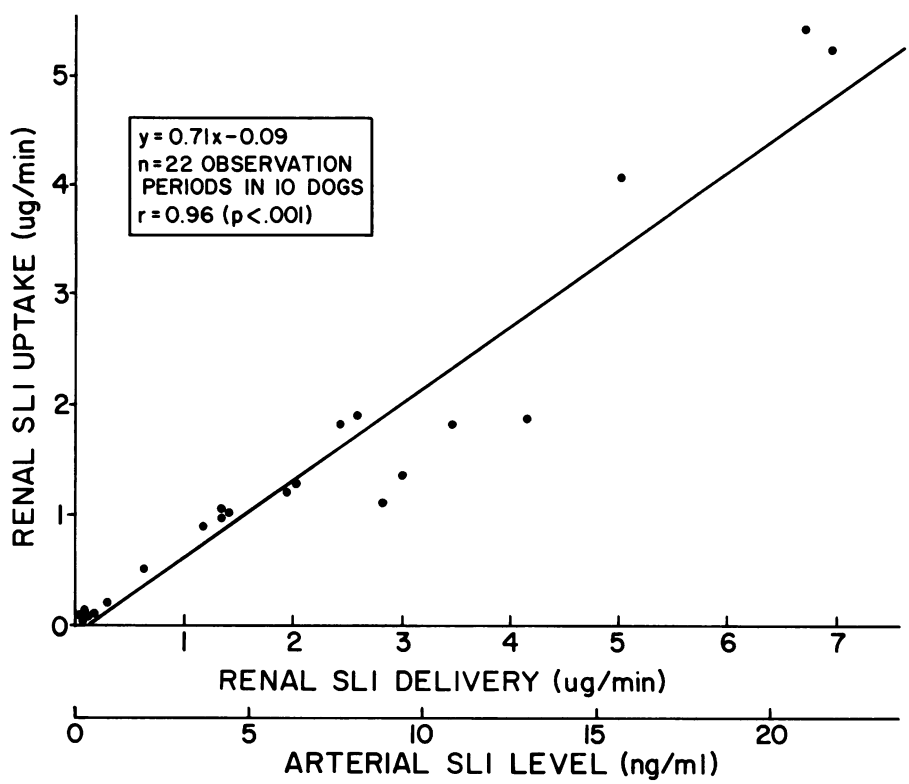

FIGURE 4 The relationship between renal SLI delivery (μg/min) and renal SLI uptake (μg/min). The approximate arterial level corresponding to a given delivery is again shown below the abscissa for references as in Fig. 3.

in close agreement with results obtained by other workers (22–25), confirming the validity of our experimental model.

All hormonal measurements were made under steady-state conditions, eliminating the variability inherent in an experimental design in which hormonal levels are changing rapidly. Furthermore, the peripheral infusion of exogenous somatostatin ensured complete mixing of the peptide in the portal vein, thus eliminating the possibility that portal streaming of newly secreted endogenous SLI may result in nonrepresentative portal concentrations. In the accurate assessment of SLI metabolism two particular considerations require attention, namely validity of the radioimmunoassay of plasma SLI and the possible influence of the heterogeneity of plasma SLI on measured organ extraction.

One of the major problems encountered in the radioimmunoassay of plasma SLI is degradation of [^{125}I-Tyr^{1}]-somatostatin (10). There is a marked species difference in this regard: human, rat, and baboon plasma show a much greater propensity for degradation of [^{125}I-Tyr^{1}]-somatostatin than dog plasma (26). In fact under the acid conditions of our assay and in the presence of Trasylol there is no evidence of tracer degradation after incubation with dog plasma for 24 h as judged by both immunoprecipitation and paper chromatoelectrophoresis. These findings are similar to those of Boldt et al. (26). We have therefore found it unnecessary to use extraction techniques as described by others to overcome degradation of [^{125}I-Tyr^{1}]somatostatin (10, 27).

There is much controversy surrounding the molecular forms of circulating SLI. Conlon et al. (28) found that at pH 8.8, endogenous as well as synthetic somatostatin added to dog plasma in small amounts eluted in the void volume of a Sephadex G-25 column. If synthetic somatostatin was added in concentrations >5 ng/ml, a proportion coeluted with synthetic somatostatin. Under acid conditions SLI eluted with synthetic somatostatin, suggesting that somatostatin had been dissociated from a plasma binder. Patel et al. (10) found that rat portal plasma SLI contained equal amounts of immunoreactivity corresponding to the tetradecapeptide as well as material of molecular weight >5,000. The nature of this big SLI is unknown. Utsumi et al. (11) found that most SLI in rat plasma eluted with synthetic somatostatin, although a small amount was present in the void volume. Insignificant levels of void volume SLI were found under both acid and neutral chromatographic conditions.

This study was not designed to identify the predominant form of circulating SLI normally present. Because our antibody recognizes the central portion of the somatostatin molecule it cannot distinguish between the intact tetradecapeptide and somatostatin molecules that have undergone N-terminal modifications. Thus the radioimmunoassay measures plasma somatostatin-like immunoreactivity and not strictly plasma somatostatin. After infusion of 1,600-dalton exogenous somatostatin, virtually all the somatostatin immunoreactivity eluted from the columns in the same position as synthetic somatostatin. The data presented relates to the metabolism of this plasma fraction. Furthermore the hepatic handling of endogenous SLI was the same as that of the SLI present after infusion of exogenous somatostatin, irrespective of the plasma level to which it was infused. Thus, even if somatostatin infusion alters the relative proportions of plasma SLI fractions, this did not affect the hepatic handling of the peptide.

The reasons for the apparent difference between the chromatographic profiles of plasma SLI in this study and others is uncertain. It may be due to differences in species, assay conditions, pH of buffer used to elute SLI from the columns, or different antibody specificities. In this regard we have found that different glucagon antibodies that show equivalent immunoreactivity with 3,500-dalton glucagon, have very different specificities with respect to void volume glucagon immunoreactivity (29). This is a possible explanation for the low levels of big SLI measured in our assay. The data presented therefore represent the metabolism of an ~1,600-mol wt SLI fraction only.

These studies indicate that in the dog, the liver and kidney are major sites of SLI metabolism. The liver extracted 29.4±2.3% of the SLI delivered to it and hepatic SLI clearance was 5.2±0.5 ml/kg per min. Renal SLI extraction was 62.3±5% and renal clearance was 7.0±1.1 ml/kg per min. The extraction process in both liver and kidney was shown to be linear over a wide concentration range from physiological (<200 pg/ml) to high pharmacological levels. Therefore both organs exhibit an extremely large capacity for SLI extraction. No difference in the hepatic handling of endogenous and exogenous SLI was shown. The MCR of infused SLI was 19.7±1.6 ml/kg per min. The hepatic contribution to overall MCR was 32.7±4.6%, and the simultaneously measured renal contribution was 37±4.9%.

The site of metabolism of the 30% of infused SLI not accounted for by hepatic and renal metabolism is unknown. Because somatostatin is secreted by the gastro-intestinal tract (30) as well as by the pancreas, arterial portal differences represent net somatostatin fluxes across the gut and not simply extraction. Thus it is not possible to measure accurately bowel metabolism of the peptide in a model such as ours. In the experiments in which somatostatin was infused to high levels, secreted SLI probably contributes only a small percentage of the total portal SLI level. This is particularly likely because under the conditions of our ex-

periment no stimulus to somatostatin secretion was present. Furthermore there is evidence that infusion of somatostatin may suppress endogenous secretion of the peptide (31). Thus in dogs in which the plasma SLI level was >5 ng/ml we noted a consistently large arterial-portal SLI gradient. The mean gut extraction calculated from portal vein plasma flow and arterial and portal SLI concentrations, was 41.8±3.6%, representing 29.4±2.3% of overall metabolic clearance.

There are a number of similarities between the in vivo handling of somatostatin, insulin, and glucagon. The major sites of metabolism of all three peptides are kidney and liver. The renal handling of all three peptides is characterized by high extraction ratios and negligible urinary excretion of the intact molecule, suggesting that hormone removed by the kidney is degraded by this organ. In addition renal disposal of all three peptides involves both glomerular filtration and uptake from peritubular blood (21, 32). Lastly, the renal metabolism of all three peptides in nonsaturable at very high plasma levels. All three peptides have a rapid half life when injected intravenously, although the half life of somatostatin (1.9 min) is faster than that of either insulin (4.5 min) or glucagon (4.2 min) (13).

There are, however, certain differences in the metabolism of these three peptides. Insulin is degraded in the liver after initially binding to cell surface receptors (33). Hepatic insulin extraction is saturable at high delivery rates that accompany high serum insulin levels (34). This may be due to saturation of hepatic binding sites for insulin which have been suggested to mediate the hepatic metabolism of this hormone (33). Sonksen et al. (35) showed that in man, increasing peripheral insulin from basal levels of 16–216, μU/ml by exogenous insulin infusion lead to a fall in the MCR of insulin from 34 to 11.4 ml/kg per min. These authors suggested that since the major site of insulin metabolism is the liver, progressive saturation of the degradative process was presumably occurring largely in the liver. This interpretation is consistent with the observations of numerous workers that in the kidney, the other major site of insulin metabolism, extraction is nonsaturable. Experiments performed on the dog have provided data that confirm the impression of saturability of hepatic insulin metabolism[2]. Hepatic glucagon extraction, on the other hand, is nonsaturable until high pharmacologic plasma glucagon levels have been reached, and the metabolic clearance rate for glucagon is nonsaturable at very high infusion rates of exogenous hormone (13). In these studies we were unable to show saturation of SLI removal by the liver despite extremely elevated arterial levels of the peptide. This is in agreement with the findings of Sacks et al. (36) in an isolated perfused rat liver system. Metabolic clearance rate for SLI was also nonsaturable at these extremely high infusion rates. These differences may have some relevance to the mechanisms for hepatic peptide hormone metabolism. Binding of hormones to receptors is recognized to be a saturable process and in this light, the differences between the saturability of hepatic insulin extraction and nonsaturability of hepatic glucagon extraction are noteworthy. These observations might therefore be taken to support the suggestion (33) that hepatic insulin extraction is dependent on initial receptor binding. However, hepatic glucagon extraction may not be dependent, or alternatively, depend only partly on receptor binding. Hepatic receptors for somatostatin have not been demonstrated and in this context our observations of nonsaturability of hepatic SLI metabolism may have similar implications.

A number of studies have suggested that the hepatic metabolism of peptide hormones may be related to their biological effects on that organ. Proinsulin, which has ~5–10% of the intrinsic activity of insulin (34) is also extracted by the liver to a much lesser extent than insulin. Hruska et al. (37) have shown that intact parathyroid hormone increases hepatic glucose output and is avidly extracted by the liver. The amino terminal fragment of parathyroid hormone, however, has no effect on hepatic glucose output and is not extracted to any significant degree. We have shown that the metabolically active 3,500 mol wt plasma glucagon component is extracted by the liver, whereas other molecular species of immunoreactive glucagon that do not have known metabolic actions are not extracted by the liver (12, 38). The question of whether somatostatin has a role in regulating hepatic function is controversial. Although some investigators have suggested somatostatin directly affects hepatic gluconeogenesis and glycogenolysis (39), others have argued that its effect on the liver is purely secondary to the suppressive action on insulin and glucagon secretion (40). The fact that somatostatin is extracted by the liver to such a significant extent raises the possibility that it may have a physiological action on the liver.

In addition to the theoretical issues raised by the in vivo metabolism of somatostatin a number of practical implications arise. Because the percentage of hepatic extraction of the peptide varies considerably from animal to animal, the magnitude of hepatic extraction will not be known in any given animal. As a result of this, portal somatostatin levels cannot be accurately predicted from peripheral levels. It is also possible that in any given animal hepatic extraction of somatostatin may vary under different physiological conditions. Furthermore as a result of significant

[2] Polonsky, K. Unpublished observations.

hepatic extraction, changes in portal somatostatin levels may not be appreciated in the periphery. This must be taken into consideration in studies attempting to elucidate the physiological importance of the peptide.

It has been suggested that somatostatin may be important in the treatment of diabetes (41), gastrointestinal hemorrhage (42), and acute pancreatitis (43). Any therapeutic potential that it may have in these conditions would be severely limited by its rapid metabolic disposal. It is possible, however, that analogues less avidly degraded at these sites, may demonstrate greater therapeutic benefit.

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