ELEVATIONS OF GASTROINTESTINAL HORMONES
IN CHRONIC RENAL FAILURE

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Summary

Fasting levels of 5 gut hormones were studied in 30 patients with advanced uraemia (CRF), 40 undergoing regular dialysis (RD) and 55 renal transplant patients (RT). Mean values of gastrin and total glucagon were markedly elevated in CRF and RD patients compared with 20 normal subjects; there were lesser elevations in pancreatic glucagon, insulin and vasoactive intestinal peptide (VIP). Secretin levels were unchanged. In RT patients, fasting levels of VIP and pancreatic glucagon had returned to normal, while levels of gastrin, total glucagon and insulin remained slightly elevated compared with controls.

Food stimulated hormone levels were measured in 18 RD patients and compared with 18 controls. After eating, RD patients failed to show the late increase in total glucagon, or the suppression of VIP and secretin seen in normal subjects; the pattern of gastrin and insulin response was similar to controls, but after the initial increase plasma levels in RD patients tended to show a slower decline.

Thus involvement of the gastrointestinal tract in uraemia is associated with functional disturbance of the endocrine system of the gut.

Introduction

The kidney produces hormones such as renin and erythropoietin but its role in removing hormones from the circulation may be equally important. The list of hormones removed from the circulation by the kidney is continually growing, and now includes hormones of gastrointestinal origin, such as gastrin [1] and gastric inhibitory polypeptide [2]. The purpose of this study was to examine fasting and food-stimulated levels of gut-hormones in patients with advanced uraemia and to compare them with normal subjects.

Patients and Methods

Patients  Fasting hormone concentrations were measured in four groups of
patients; 20 normal subjects without known renal/gastrointestinal disease or diabetes; 30 patients with advanced uraemia (creatinine clearance <10 ml/min); 40 patients undergoing regular haemodialysis, and 55 renal transplant patients. In 40 of the 55 transplant patients, prednisolone dosage and serum creatinine were recorded at the time of testing. In dialysis patients samples were taken on the morning of a dialysis day (i.e. 48–72 hours after the previous haemodialysis).

Food stimulated hormone concentrations were measured in 18 normal subjects and 18 regular dialysis patients. Informed consent was obtained from all subjects involved.

Methods Hormone radioimmunoassays were carried out by conventional methods with antisera raised to synthetic human gastrin I [3], purified natural secretin [4], porcine insulin [5], natural vasoactive intestinal peptide (VIP) [6]; two different antibodies were used in the glucagon assay, both raised to porcine glucagon [4]. Antibody YY 57 reacts with the N-terminal sequence of the glucagon molecule and detects all known forms of glucagon-like immunoreactivity (GLI), whether originating from pancreas or gut. Glucagon measured by this antibody is referred to as total glucagon or N-GLI. Antibody YY 89 reacts with the C-terminal sequence of the glucagon molecule (C-GLI) but not with most forms of gut GLI, and it is therefore often regarded as measuring ‘pancreatic glucagon’. For practical purposes, change in N-GLI (total glucagon) alone suggests that the origin is gut, while simultaneous change recorded in both N-GLI and C-GLI implies that the alteration is in ‘pancreatic’ glucagon.

The assays could detect the following plasma changes with 95% confidence: gastrin 5 ng/L; secretin 6 ng/L; insulin 0.5 mu/L; VIP 5 ng/L; glucagon 8 ng/L. No significant cross-reaction was detectable between any of the peptides assayed.

Blood samples for hormone assays were taken into heparinised tubes with 400 KI units of aprotinin (“Trasylol”) per ml added, and were placed immediately in an ice bath. For glucagon, secretin and VIP assays, the plasma was extracted by the method of Heding [7] and stored as a dried extract; samples for insulin and gastrin were stored as frozen plasma. Fasting samples were obtained after a 12 hour overnight fast, and in 20 RD patients, the effect of a single haemodialysis on fasting levels was studied. In this group, fasting samples were taken before the 14-hour dialysis period, and again the following morning. The release of hormones in response to food was measured using a standard test meal which contained 50 grams carbohydrate, 18 grams protein and 20 grams fat. This meal consisted of orange juice with added glucose, three sandwiches of lean cooked ham, and a cup of tea (with 30 ml of milk), sweetened, if necessary, with Saxin and not sugar. The meal test was performed in the morning after an overnight fast. The subject reclined at 45° on a couch throughout the test and blood samples were removed through an indwelling needle in a forearm vein.

The significance of differences was calculated using the Student t and paired t tests as appropriate. In renal transplant patients, linear regression analysis was used to determine if a correlation existed between (a) prednisolone
Figure 1. Basal hormone levels ± SEM (expressed as percentage of normal) in patients with chronic uraemia (CRF), undergoing regular dialysis therapy (RDT), and after renal transplantation (RT). P values refer to comparison with group of 20 normal subjects.
dosage, (b) serum creatinine and basal levels of gastrin, insulin, N-GLI. ‘Normal’ in Figure 1 refers to the mean basal level in the group of 20 normal subjects.

Results

Fasting Hormone Levels

The basal plasma concentrations of the 5 hormones in each group of patients are shown in Figure 1. There were marked elevations in gastrin and N-GLI levels in both CRF and RD patients, lesser elevations of insulin, C-GLI and VIP. Secretin levels were unchanged. The only difference between the CRF and RD groups was that the slight elevation in insulin in the RD group did not attain statistical significance. After renal transplantation, VIP and C-GLI had returned to normal, gastrin, N-GLI and insulin remained slightly elevated compared to controls. There was a positive correlation in transplant patients between basal gastrin and serum creatinine ($r=0.4243$, $P < 0.01$) but no correlation with prednisolone dosage. The mean serum creatinine in the RT group was 131 µmol/L (upper limit of normal 110 µmol/L). For basal insulin and N-GLI there was no correlation with either serum creatinine or prednisolone dosage.

Effect of Haemodialysis

The effect of a single haemodialysis on fasting hormone concentrations in 20 RD patients is shown in Table I. The mean values of insulin, N-GLI and secretin were unchanged after a single haemodialysis; the mean values of gastrin and C-GLI were decreased, while the mean plasma VIP level was increased.

TABLE I. Effect of a Single Haemodialysis on Basal Hormone Levels

<table>
<thead>
<tr>
<th></th>
<th>INSULIN mlU/l</th>
<th>GASTRIN ng/l</th>
<th>SECRETIN ng/l</th>
<th>VIP ng/l</th>
<th>GLUCAGON N-GLI ng/l</th>
<th>C-GLI ng/l</th>
</tr>
</thead>
<tbody>
<tr>
<td>PRE-DIALYSIS</td>
<td>10.3 ± 5.8</td>
<td>492 ± 947</td>
<td>31.3 ± 32</td>
<td>121 ± 74.3</td>
<td>742 ± 472</td>
<td>176 ± 53.8</td>
</tr>
<tr>
<td>POST-DIALYSIS</td>
<td>11.6 ± 6.8</td>
<td>419 ± 792</td>
<td>24.3 ± 14</td>
<td>164 ± 93.8</td>
<td>708 ± 340</td>
<td>155 ± 55.5</td>
</tr>
<tr>
<td>$P$ VALUE (Paired t)</td>
<td>&lt; 0.2</td>
<td>&lt; 0.05</td>
<td>&lt; 0.15</td>
<td>&lt; 0.05</td>
<td>&lt; 0.35</td>
<td>&lt; 0.0125</td>
</tr>
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</table>

Test Meal

After ingestion of the test meal, normal subjects showed an increase in plasma levels of gastrin, insulin and N-GLI, and suppression of plasma VIP and secretin levels. C-GLI remained unchanged. For the purpose of clarity, only those
hormones which showed a markedly different response in RD patients are illustrated. In the RD group, the pattern of plasma gastrin response was similar to normal subjects, but the peak value was reached earlier and plasma levels tended to decline more slowly (Figure 2). Plasma insulin response in the two

![Graph](image)

**Figure 2.** Plasma gastrin response to test meal in regular dialysis patients and controls

groups differed only in the 2 hour value, this being significantly higher in the RD patients. The late rise in N-GLI observed in normals was not seen in the RD patients (Figure 3), and similarly the suppression of VIP (Figure 4) and secretin observed after food in normals was not apparent in the RD group.

**Discussion**

The study has shown that the fasting ‘gut-hormone profile’ is altered in chronic uraemia; plasma levels of gastrin and total glucagon (N-GLI) are markedly elevated; insulin, VIP and pancreatic glucagon (C-GLI) are elevated to a lesser degree, while secretin levels are unchanged. The only difference between CRF and RD groups was that the elevation in basal insulin did not attain statistical significance in the RD patients.
Figure 3. Total glucagon (N-GLI) response to test meal in regular dialysis patients and controls. (P values indicate controls show significant rise at 90 and 120 minutes compared with baseline values; RD patients do not show any increase)

Figure 4. Plasma VIP response to test meal in regular dialysis patients and controls. (P values indicate points at which control values differ significantly from baseline; RD patients do not show significant suppression at any point)
Fasting hyperinsulinaemia has been reported in uraemic patients both before and after adequate haemodialysis [8], and factors thought to be responsible include impaired insulin degradation [9] and insensitivity of the peripheral tissues to insulin [10]. The hypergastrinaemia and hyperglucagonaemia recorded in uraemic patients in this study is in keeping with the findings of previous similar investigations [11,12]; the slight increase in basal gastrin observed in RT patients compared with normal subjects probably reflects the small differences in serum creatinine, in view of the correlation between these two variables and the difference in mean serum creatinine between the two groups. The cause of the slight elevation in insulin and total glucagon after transplant is not clear, as no relationship could be shown with either serum creatinine or prednisolone dosage. The finding that plasma VIP levels are elevated in uraemic subjects and returned to normal after transplantation suggests that the kidney may also be involved in degradation of this hormone. In contrast, the lack of any change in secretin levels suggests that the kidney is unlikely to be involved in catabolism, and this is contrary to conclusions reached from recent studies of secretin metabolism in animal models [13].

The study has also shown that haemodialysis may exert a short-term effect on some basal hormone levels, increasing VIP and decreasing gastrin and C-GLI. There was however, no difference in basal hormone levels between CRF patients and RD patients tested 48–72 hours after dialysis, suggesting that the elimination capacity of regular haemodialysis does not exceed the secretory capacity of the endocrine cells.

In addition to basal hormone concentrations, there were also differences in the hormonal response after eating when RD patients were compared with normal subjects. Plasma gastrin reached an earlier peak in RD patients and tended to decline more slowly (Figure 2); this suggests that increased and prolonged gastric acid secretion may occur after a meal in such patients. Plasma insulin response in RD patients also tended to decline more slowly, a finding in keeping with the important role of the kidney in metabolism of circulating insulin. Gut GLI is produced mainly in the lower small intestine and thus, after eating, a late increase is observed in normal subjects [14]. This response was not observed in RD patients (Figure 3) and could reflect slower intestinal transit-time.

In normal subjects, the suppression of plasma VIP and secretin levels after eating suggest that these hormones might act as physiological inhibitors of gut secretory activity. In RD patients, plasma VIP (Figure 4) and secretin levels failed to suppress after eating and the reason for this is not clear.

It may be concluded that both the fasting and food-stimulated gut hormone profile is abnormal in patients with advanced uraemia, and in those undergoing regular dialysis. There is evidence that hypergastrinaemia is implicated in acid hypersecretion and frequent duodenal ulceration occurring in RD patients [15], but the role of the other gut hormones studied here is less clear. N-GLI is thought to influence mucosal growth [16] and it is therefore conceivable that hyperglucagonaemia may be related to hyper-regenerative changes which have been described in the small bowel mucosa of uraemic patients [17]. Glucagon also has a hypocalcaemic effect and thus might (in part at least) contribute to the low serum calcium levels of chronic renal failure. The significance of elevated VIP
levels in uraemic patients is also a matter of speculation at present, but it is interesting to note that elevated levels are associated with the watery diarrhoea syndrome [18], and the known effects of the hormone include abnormalities of lipid and carbohydrate metabolism, and impairment of platelet aggregation [19], features common in advanced uraemia.

Caution is necessary in interpreting the results of this study, based as they are on radioimmunoassay measurements. Immunoreactivity does not necessarily reflect biological activity and it is obviously important to know more about the nature of what the assay measures as ‘immunoreactive hormone’. For example, the majority of circulating insulin in uraemic patients is pro insulin, which has very little hypoglycaemic activity [20]. Analysis of the various circulating forms of gut hormones in uraemic patients is therefore necessary in order to establish the significance or otherwise of elevations in ‘total’ immunoreactivity. Other problems of radioimmunoassay include the influence of inter hormonal relationships on blood levels, and the relative lack of information on hormone catabolism.

In conclusion, it appears from these preliminary studies that involvement of the gastrointestinal tract in chronic uraemia is not only histological, but is associated with functional disturbance of the endocrine system of the gut. The implications of this merit further attention.

Acknowledgments

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Open Discussion

KLINKMANN (Rostock) Did you take any biopsies from the gut? I wonder if the influence of regular dialysis may be due to the treatment itself or just associated with the change in the overall uraemic intoxication. Are the changes related to the degree of uraemic intoxication or to an effect of the dialysis treatment?

DOHERTY We have no biopsy material from small bowel mucosa in these patients. I agree with you that it would be extremely interesting to have this. Glucagon for example might be responsible for hyper-regenerative changes in the small bowel. We have no information relating to the degree of uraemia because all our patients were similar in having virtually end stage renal failure.

ROODVOETS (Haarlem) If the lack of removal by the kidneys is the reason for the elevation of plasma levels of these hormones, one would expect, because of the feedback, that usually the basal levels would be back to normal again unless there are many stimuli during the 24 hours. Another possibility of raised levels may be diminished end organ sensitivity to hormones like PTH for instance.

DOHERTY Yes, impaired end organ sensitivity is relevant, not only to parathyroid hormone but also to insulin. Elevated insulin levels reflect both impaired degradation by the kidney and peripheral tissue resistance to the effect of the hormone. I think the mechanisms underlying elevations in circulating hormones are complex. At any point in time, a single plasma level is the net effect of secretion, degradation or excretion, tissue uptake and possibly other factors about which we know very little.

KOPP (Munich) I think your studies have very important implications in particular for acute renal failure. They support our observations that one has to withhold oral alimentation in these patients until or nearly until normal kidney function has returned because oral feeding may provoke diffuse gastrointestinal bleeding.

DOHERTY I am not sure you can attribute gastrointestinal bleeding in acute renal failure to oral feeding. It has been suggested that hypergastrinaemia in acute uraemia may cause GI bleeding, but I wonder whether this is cause or effect — if a patient with acute renal failure develops gastric erosions, back diffusion of hydrogen ion will cause a drop in titratable gastric acidity and this reduction in feedback inhibition of gastrin release may produce massive
gastrin elevation, when coupled with impaired renal removal. I often wonder whether massive hypergastrinaemia is the cause of or the effect of gastrointestinal bleeding in acute renal failure.

RITZ (Heidelberg) Nutrition is known to affect the serum concentration of many of these hormones. Do you have information on a comparable group with malnutrition due to diseases other than chronic renal failure?

DOHERTY I agree that malnutrition of itself may alter circulating levels of these hormones, particularly secretin which has been shown to rise with starvation. I agree that a control group with chronic diseases other than renal failure would be desirable. We do not at present have such data.