Nephrology Dialysis Transplantation. 2008; 23(11): 3696-3703

Short-term regulation of peptide YY secretion by a mixed meal or peritoneal glucose-based dialysate in patients with chronic renal failure

Miguel Pérez-Fontán[1,2], Fernando Cordido[1,3], Ana Rodríguez-Carmona[2], Manuel Penín[3], Helena Díaz-Cambre[2], Andrés López-Muñiz[2], Susana Sangiao-Alvarellos1[,3], Jesús García-Buela[4]

[1] Department of Medicine, University of A Coruña, [2] Divisions of Nephrology, [3] Endocrinology and [4] Laboratory, University Hospital Juan Canalejo, Spain

Abstract

*Background***.** Malnutrition is very prevalent among patients with chronic renal failure. The role of derangements in the gut–brain axis for regulation of appetite in the genesis of anorexia of these patients has not been adequately investigated.

*Design***.** Following a randomized, crossover design, we analysed plasma levels of peptide YY (PYY) $_{1–36}$ and PYY $_{3–36}$ both fasting and after a standardized oral mixed meal or intraperitoneal glucose infusion in 10 stable uraemic patients undergoing peritoneal dialysis and 8 healthy controls, matched for age, gender and body mass index.

*Main results***.** Median baseline plasma levels of PYY $_{1–36}$ in the different provocation tests oscillated between 406 and 460 pg/mL in patients, as compared with 73 and 100 pg/mL in controls ($P < 0.001$). Corresponding values for PYY $_{3–36}$ oscillated between 235 and 267 pg/mL in patients, versus 56 and 70 pg/mL in controls ($P < 0.001$). The association of high levels of PYY $_{3–36}$ and normal levels of acylated ghrelin (when compared with healthy controls) configurated a markedly pro-anorexigenic pattern in patients. Neither oral intake nor intraperitoneal glucose resulted in significant changes in plasma levels of PYY $_{1–36}$ or PYY $_{3–36}$ in subjects with renal failure, in contrast with the expected postprandial rise observed in healthy controls (41% for PYY $_{1–36}$, $P = 0.04$ and 32% for PYY $_{3–36}$, $P = 0.02$, median values).

*Conclusions***.** Baseline plasma levels of PYY $_{1–36}$ or PYY $_{3–36}$ are markedly elevated in patients with renal failure undergoing peritoneal dialysis. Provocation studies disclose a marked disregulation in the postprandial secretion of these anorexigenic peptides, when compared with healthy controls. These findings may contribute to clarify the complex pathogenesis of anorexia of chronic renal failure.

Keywords

Anorexia; Ghrelin; Peritoneal dialysis; PYY; Renal failure

Introduction

Malnutrition is highly prevalent among patients with chronic renal failure (CRF), and represents a consistent marker of poor survival in this population [1]. Many factors beyond inadequate nutrient intake contribute to the pathogenesis of this complication [2], but anorexia is a prominent feature of the uraemic syndrome. Dialysis therapy is often unable to correct, and may even worsen, this symptom [3–5]. During the last decades, considerable attention has been devoted to the potential role of derangements in the mechanisms that regulate short- and long-term nutrient intake and utilization [6] in the pathogenesis of the nutritional disorders of CRF. Resistance to the anabolic effects of insulin is a well-known contributor to malnutrition, in this setting [2]. Also, distorted secretion of adipocytokines may compromise the nutritional status of these patients [7]. Plasma leptin levels are consistently elevated in patients with CRF, particularly in those treated with peritoneal dialysis (PD) [8], but the actual contribution of this disorder to anorexia and malnutrition of uraemia is controversial [9,10].

The role of potential derangements of the gut–brain axis in the genesis of disorders of appetite of patients with CRF has not been established. Plasma levels of total (but not acylated) ghrelin have been found to be increased in CRF [11], and a subnormal suppression of the secretion of this orexigenic peptide by oral intake has also been observed in these patients [12]. On the other hand, exogenous administration of ghrelin is able to increase appetite in anorexic patients with CRF [13]. Other gut peptides implicated in the regulation of appetite have not been adequately studied in CRF, so far.

Peptide YY (PYY) is released by intestinal L cells in response to meals, both as the full 36 amino acid molecule ($PYY_{1–36}$) and as the 3–36 fragment ($PYY_{3–36}$), which seemingly brings most of the biologic activity of the hormone. PYY appears to play a significant role in the short-term regulation of appetite [14,15], acting as a break to oral intake by complex mechanisms, which include stimulation of vagal afferents and a direct interaction with the receptor Y2 (Y2R) of the hypothalamic arcuate nucleus [16]. Exogenous administration of PYY reduces oral intake in rodents [17] and obese humans [18], and plasma PYY levels are elevated under several low-appetite conditions, including anorexia nervosa [19]. Altered PYY secretion patterns could play a role in the pathogenesis of anorexia of CRF, but this possibility has not been adequately investigated. We have performed a crossover randomized trial, with the aim of disclosing potential derangements in the response pattern of PYY to a standardized mixed meal and to hypertonic glucose-based PD-dialysate, in a group of uraemic patients undergoing chronic PD.

Population and method

General design

Following a crossover design, 10 non-diabetic patients undergoing chronic PD therapy and 8 healthy controls, matched for age, sex and body mass index, were scrutinized for baseline $PYY_{(1–36)}$ and $PYY_{(3–36)}$ levels, as also for the reponses of these peptides to a standardized oral feeding, hypertonic glucose-based dialysate and orally administered placebo. Thus, every patient underwent three tests, and every control two tests (oral feeding and placebo), with a time span of 40 days for the whole sample retrieval period. Tests were carried out in a randomized order. The study protocol fulfilled the requirements of the ethical committee of our centre, and a written informed consent was obtained from both patients and controls.

Subjects

Patients were randomly selected from our population undergoing chronic PD therapy, after applying the following exclusion criteria: diabetes mellitus, age <30 or >70 years, significant clinical events during the previous 3 months and unwillingness or inability to cooperate. Controls were selected from a pool of healthy volunteers available to the endocrinology unit of our centre. Both groups were comparable in regard to age [median 56 years (range 36–65) for patients versus 53 (36–61) for controls], gender (50% males in each group) and body mass index [24.7 kg/m^2 (20.7–32.0)

for patients versus 25.7 (20.1–31.3) for controls]. Patients had been treated with PD for a median of 11 months (3–63), preserving a median glomerular filtration rate (GFR) (as estimated from the mean of urea and creatinine clearances) was 4.2 mL/min (0.0–8.8) in patients. Three patients were oligoanuric at the time of the study. No patient reported overt anorexia or suffered a significant clinical event during the study period.

Protocol

Patients were instructed to perform a single nighttime PD exchange using either 1.36% glucose- or icodextrin-based dialysate (Baxter®, Deerfield, IL, USA), the day before the test. After an overnight fast, all tests started between 8:30 and 9:00 a.m. Patients and controls were at rest in a sitting position, and a peripheral venous line was obtained. Thirty minutes later, both received the following on three different days.

- A standardized oral feeding, consisting of 400 mL of Isosource Energy (Novartis®, Zurich, Switzerland), containing 5.7 g of protein, 6.2 g of lipid and 20 g of carbohydrate per 100 mL, for a total caloric content of 158.8 kcal/100 mL (14.4% protein, 35.1% lipid, 50.4% carbohydrate). The first PD exchange of the day was delayed until the end of the test, in patients. All the patients and controls were able to eat the whole test meal.
- A PD exchange consisting of 2 L of dialysate (Baxter ®, Deerfield, IL, USA) with a glucose concentration of 3.86 g/dL (patients). Patients were kept fasting until the end of the test.
- Orally administered placebo (400 mL of tap water). Patients/controls were kept fasting and, again, the first PD exchange of the day was delayed until the end of the test in patients.

We obtained blood samples for $PYY_{(1–36)}$, $PYY_{(3–36)}$, total and acylated ghelin, leptin, insulin and growth hormone (GH) at baseline and then at 30, 45, 60 and 120 min. Data concerning the responses of total and acylated ghrelin, leptin, insulin and GH during these tests have been previously published[12]. Baseline hormonal values (except PYY levels) have also been presented in the aforementioned article.

Sample management and laboratory methods

All blood samples were immediately centrifuged, separated and frozen at −80°C. Samples destined for determination of ghrelin and PYY levels were specifically retrieved to chilled tubes containing aprotinin and EDTA-Na, and then immediately centrifuged at 4°C, separated to aliquots and frozen at −80°C.

For estimation of the main scrutinized hormones, we used the following specific commercial assays:

- $PYY_{(1–36)}$ (RIA, Linco, St Charles, MO, USA) (intra- and inter-assay variation coefficients 2.9% and 5.5%, respectively; lower detection limit 10 pg/mL)
- $PYY_{(3–36)}$ (RIA, Linco, St Charles, MO, USA) (intra- and inter-assay variation coefficients 6.4% and 7.7%, respectively; lower detection limit 20 pg/mL)
- Total ghrelin (RIA, Phoenix Pharmaceuticals, Belmont, CA, USA) (intra- and inter-assay variation coefficients 5.3% and 13.6%, respectively; lower detection limit 10 pg/mL)
- Acylated ghrelin (RIA, Linco, St Charles, MO, USA) (intra- and inter-assay variation coefficients 6.5% and 9.6%, respectively; lower detection limit 10 pg/mL).

Other hormonal laboratory estimations included leptin (RIA, Mediagnost, Tübingen, Germany), insulin (RIA, CIS Bio International, Cedex, France), GH and IGF1 (RIA, Nicholls Inst. Diagnost., San Juan Capistrano, CA, USA) and cortisol (Chemoluminiscence, ADVIA Centaur System, Bayer, Leverkusen, Germany). We also estimated baseline (placebo test) plasma levels of interleukin-6 (ELISA, R&D Systems, Minneapolis, MN, USA) and high-sensitivity C-reactive protein (hsCRP) (Immunoturbidimetry, Roche Diag., Mannheim, Germany). Plasma levels of albumin, prealbumin, cholesterol, triglycerides and glucose (as also dialysate glucose levels) were estimated using a standard autoanalyzer.

Secondary calculations

The time courses of plasma PYY $_{(1–36)}$ and PYY $_{(3–36)}$ levels after the different tests were analysed in absolute terms and also as a percent change versus baseline, the latter to overcome potential biases induced by potentially different baseline plasma levels of both peptides in patients and controls. Areas under the curve were estimated using the trapezoidal method.

Glucose absorption during the hypertonic PD exchange test was estimated by simple mass balance. Resistance to insulin was estimated indirectly, according to the Homeostasis Model Assessment (HOMA) score [plasma insulin (mcUI/mL) * plasma glucose (mM/L)/22.5] [20].

Statistics

All comparisons were based on univariate, non-parametric tests. The degree of concordance between baseline estimations was analysed using Spearman's correlation coefficient and the *t* -test for paired data (systematic biases). Intragroup comparisons (changes versus baseline and oral versus placebo) were based on Wilcoxon's test. Comparisons between patients and controls were based on Fisher's (categorical) and Mann–Whitney *U* (numerical) tests. Numerical correlations were analysed using Spearman's correlation test. *P* -values ≤ 0.05 were considered to be significant. For graphic representation we used mean values ± SEM. The SPSS software 14.0 was used to produce statistical analysis.

Results

Baseline estimations

Main baseline hormonal estimations in the three tests are presented in Table 1. Plasma levels of both PYY $_{(1–36)}$ and PYY $_{(3–36)}$ were significantly higher in patients than in controls in each test. In patients, baseline PYY $_{(1–36)}$ and PYY $_{(3–36)}$ levels showed a good concordance between the three estimations performed, with correlation coefficients higher than 0.70 ($P < 0.02$) in all cases (Spearman), and no evidence of systematic biases. Also, the correlation coefficients between PYY $_{(1–36)}$ and PYY $_{(3–36)}$ levels in each test were high: 0.95 for the oral test, 0.90 for the placebo test and 0.92 for the intraperitoneal glucose test ($P < 0.001$ any). In reference to healthy controls, the correlation between PYY $_{(1–36)}$ and PYY $_{(3–36)}$ was also good. However, baseline plasma PYY $_{(3–36)}$ levels came out to be significantly lower during the placebo test than during the oral feeding test (Table 1). There was also more variability in the correlations between baseline plasma levels of PYY $_{(1–36)}$ and PYY $_{(3–36)}$ [$r = 0.38$ ($P = 0.31$) in the oral test, $r = 0.78$ ($P = 0.03$) in the placebo test], in the control group.

Table 1. Baseline plasma hormone levels

	Patients	Controls	*P*
PYY $_{(1–36)}$ (pg/mL)			
Oral feeding test	406 (237–1244)	100 (61–179)	0.001
Placebo test	432 (312–1247)	73 (52–164)	0.001
Hypertonic glucose-based dialysate test	460 (265–910)		
PYY $_{(3–36)}$ (pg/mL)			
Oral feeding test	267 (161–521)	70 (51–89)	0.001
Placebo test	244 (153–358)	56 (37–71) *	0.001
Hypertonic glucose-based dialysate test	235 (138–542)		
Total ghrelin (pg/mL)			
Oral feeding test	960 (728–1302)	731 (399–1016)	0.06
Oral placebo test	933 (784–1252)	669 (411–1023)	0.02
Hypertonic glucose-based dialysate test	1022 (696–1238)		
Acylated ghrelin (pg/mL)			
Oral feeding test	97 (35–185)	140 (43–193)	0.07
Oral placebo test	106 (19–188)	112 (39–227)	0.97
Hypertonic glucose-based dialysate test	128 (28–211)		
Quotient total ghrelin/PYY $_{(1–36)}$			
Oral feeding test	1.90 (0.90–4.57)	7.44 (3.64–14.56)	0.001
Oral placebo test	2.31 (0.97–4.01)	9.84 (2.51–20.95)	0.001
Hypertonic glucose-based dialysate test	1.61 (1.19–3.78)		
Quotient acylated ghrelin /PYY $_{(3–36)}$			
Oral feeding test	0.45 (0.10–0.91)	2.03 (0.92–3.00)	0.001
Oral placebo test	0.46 (0.09–0.76)	2.14 (0.74–3.85)	0.001
Hypertonic glucose-based dialysate test	0.52 (0.23–0.93)		
Leptin (ng/mL)			
Oral test	41.4 (3.6–170.8)	9.3 (3.0–24.1)	0.04
Placebo test	36.0 (0.7–103.6)	11.1 (2.7–30.5)	0.08
Hypertonic glucose-based dialysate test	36.2 (3.3–94.9)	–	
Insulin (mcUI/mL)			
Oral test	14.5 (5.1–25.3)	9.5 (3.7–16.1)	0.04
Placebo test	13.1 (6.9–20.4)	9,7 (2.8–15.0)	0.09
Hypertonic glucose-based dialysate test	13.5 (6.7–22.5)	–	
HOMA score			
Oral test	3.03 (1.70, 7.81)	2.26 (0.75, 4.05)	0.12
Placebo test	3.02 (1.89, 4.55)	2.61 (0.57, 3.83)	0.24
Hypertonic glucose-based dialysate test	2.52 (1.58, 5.71)	–	
Plasma growth hormone (ng/mL)			
Oral	3.52 (0.18, 9.50)	0.72 (0.01, 2.20)	0.08
Placebo	2.51 (0.19, 5.80)	1.35 (0.01, 6.53)	0.17
Hypertonic glucose-based dialysate test	2.19 (0.16, 5.30)	–	
Plasma cortisol (mcgr/dL)			
Oral	14.1 (5.0, 21.9)	12.3 (10.0, 16.2)	0.51
Placebo	16.3 (7.1, 20.0)	13.3 (9.4, 18.8)	0.41
Hypertonic glucose-based dialysate test	14.4 (5.4, 22.9)	–	

Figures denote median values (range) at baseline of each test. Comparisons by the Mann–Whitney *U* -test. $^{*}P = 0.04$ versus oral feeding test. Other intragroup differences not significant.

We observed no clear correlation between baseline plasma levels of PYY $_{(1–36)}$ or PYY $_{(3–36)}$ on one side, and of total or acylated ghrelin levels on the other. Quotients between baseline values of ghrelin and PYY are presented in Table 1. Values proved consistent in the three tests performed, depicting a markedly lower orexigenic pattern in patients than in controls.

In patients, we observed a significant direct correlation between baseline plasma levels of cortisol on one side, and PYY $_{(1–36)}$ (r = 0.87, P = 0.002) and PYY $_{(3–36)}$ (r = 0.88, P = 0.001) on the other, while this correlation was nonsignificant in controls. Baseline PYY $_{(1–36)}$ or PYY $_{(3–36)}$ plasma levels showed no apparent correlation with leptin, insulin or GH levels or the HOMA score. Nor did we detect any correlation between PYY $_{(1–36)}$ and PYY $_{(3–36)}$ levels and the main demographic variables scrutinized (age, gender, GFR, time on dialysis, body mass index).

All patients were apparently well nourished, as estimated from subjective global assessment. Also, as previously stated, no patient reported anorexia during the study period, although we did not apply any normalized anorexia score. One patient and one control were obese (body mass index >30). Table 2 displays the selected biochemical inflammatory and nutritional markers. None of them showed a correlation with plasma levels of PYY $_{(1–36)}$ or PYY $_{(3–36)}$, either in patients or healthy controls.

Table 2. Inflammatory and nutritional markers

	Patients	Controls	*P*
Interleukin-6 (pg/mL)	7.4 (0.0–32.9)	0.0 (0.0–8.0)	0.006
C-reactive protein (mg/L)	2.1 (0.2–13.8)	0.7 (0.2–1.4)	0.08
Albumin (g/L)	41 (36–46)	45 (40–49)	0.08
Prealbumin (mg/dL)	37 (29–41)	41 (32–48)	0.22
Cholesterol (mg/dL)	183 (119–201)	195 (130–241)	0.35
Triglycerides (mg/dL)	165 (84–460)	102 (54–276)	0.15
IGF-1 (ng/mL)	140 (111–289)	79 (63–118)	0.01

Figures denote median values (range) at baseline of each test. Comparisons by the Mann–Whitney *U* -test.

In patients, median glucose absorption 120 min after the hypertonic glucose peritoneal exchange was 36.9 g (range 29.3–56.8).

PYY $_{(1–36)}$

Plasma levels of PYY $_{(1–36)}$ in response to the mixed meal, placebo and intraperitoneal glucose are presented in Figures 1 a, b and 3 a, respectively. Only healthy controls showed a response to oral intake (Figure 1 b), while there was no apparent change of PYY $_{(1–36)}$ levels after oral feeding (Figure 1 a) or intraperitoneal glucose (Figure 3 a), in patients.

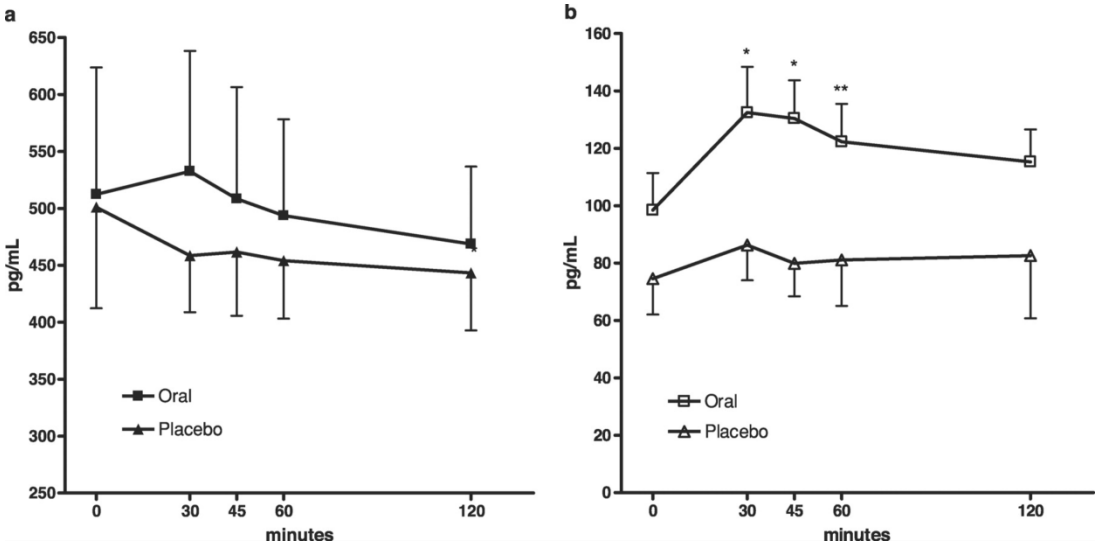

Fig. 1. **(a)** Plasma levels of PYY $_{(1–36)}$ after placebo or a mixed meal in patients. Values denote mean ± SEM. $^{*}P < 0.05$ versus baseline. Placebo versus oral, $P = 0.028$ at 30 min; other differences NS. **(b)** Plasma levels of PYY $_{(1–36)}$ after placebo or a mixed meal in controls. Values denote mean ± SEM. $^{*}P < 0.05$ versus baseline. $^{**}P = 0.09$ versus baseline. Placebo versus oral, $P < 0.05$ at 30 and 45 min; other differences NS.

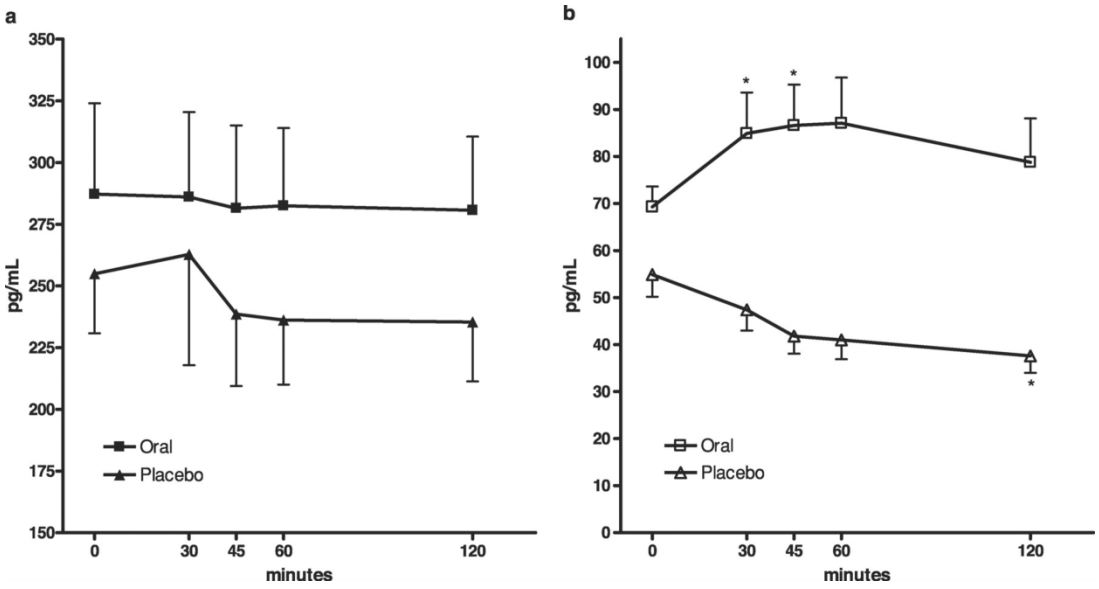

Fig. 2. **(a)** Plasma levels of PPY $_{(3–36)}$ after placebo or a mixed meal in patients. Values denote mean ± SEM. Differences NS. **(b)** Plasma levels of PYY $_{(3–36)}$ after placebo or a mixed meal in controls. Values denote mean ± SEM. $^{*}P < 0.05$ versus baseline. Placebo versus oral, $P < 0.02$ at any point beyond baseline.

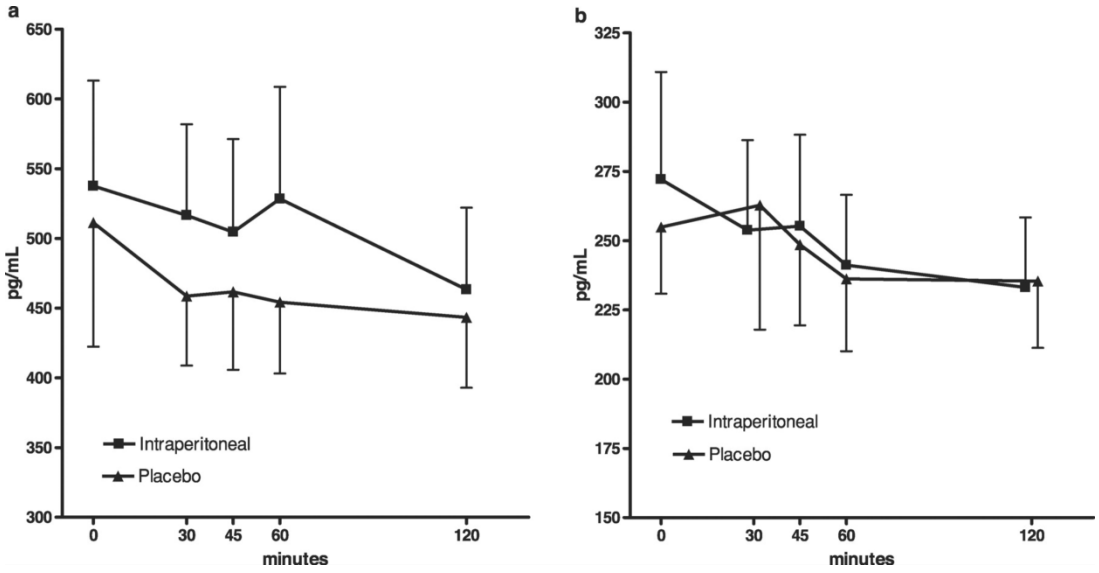

Fig. 3. **(a)** Plasma levels of PPY $_{(1–36)}$ after placebo or intraperitoneal glucose in patients. Values denote mean ± SEM. Differences NS. **(b)** Plasma levels of PYY $_{(3–36)}$ after placebo or intraperitoneal glucose in patients. Values denote mean ± SEM. Differences NS.

In patients, peak plasma levels of PYY $_{(1–36)}$ were 400 (272–1269) (meal), 462 (277–894) (placebo) and 480 pg/mL (321–950) (intraperitoneal glucose) (NS). In controls, the observed peak values were 141.5 (81–198) (meal) and 92.5 pg/mL (46–219) (placebo) (P = 0.04) (patients versus controls $P < 0.001$ any comparison).

In patients, the AUC values were 43560 (29715–128227) (meal), 50430 (32685–103477) (placebo) and 51142 pg/mL min (32025–100080) (intraperitoneal glucose) (NS). In controls, corresponding values were 14865 (8032–19822) (meal) and 8992 pg/mL min (4455–21382) (placebo) (P = 0.06). Differences between patients and controls were significant ($P < 0.001$) for each test.

PYY $_{(3–36)}$

Plasma levels of PYY $_{(3–36)}$ in response to the mixed meal, placebo and intraperitoneal glucose are presented in Figures 2 a, b and 3 b, respectively. In patients, we observed no significant change in PYY $_{(3–36)}$ levels after oral intake or intraperitoneal glucose. In contrast, controls displayed a clear rise of PYY $_{(3–36)}$ levels after oral intake (Figure 2 b).

In patients, peak plasma levels of PYY $_{(3–36)}$ were 271 (191–534) (meal), 234 (138–541) (placebo) and 247 pg/mL (130–483) (intraperitoneal glucose) (NS). In controls, the observed peak values were 92.5 (59–135) (meal) and 46.5 pg/mL (30–66) (placebo) (P = 0.02) (patients versus controls $P <$ 0.001 any comparison).

In patients, the AUC values for PYY $_{(3–36)}$ were 33982 (21277–60195) (oral), 28785 (17040–41182) (placebo) and 29 659 pg/mL min (14542–47707) (intraperitoneal glucose) (NS). In controls, corresponding values were 9086 (6412–14970) (oral) and 4984 pg/mL min (3142–6772) (placebo) (P = 0.012) (patients versus controls $P < 0.001$ any comparison).

Discussion

CRF is frequently complicated by a catabolic and inflammatory state, resulting in progressive wasting and significant mortality rates [2]. Many factors (related to renal insufficiency itself, dialysis therapies and associated comorbidities) contribute to this complex condition, but malnutrition is a cardinal feature of the syndrome [1,3]. Anorexia is a prominent factor leading to malnutrition in CRF [3], and PD bears some specific features that may aggravate this symptom [4], including a significant risk of inadequacy of dialysis once residual renal function declines [21], pressure-related satiation secondary to the presence of intra-abdominal dialysate [22] and a direct inhibition of appetite by glucose- and amino acid-based dialysis solutions [22–24]. In fact, low appetite is an almost universal feature in patients undergoing this therapy [25].

In recent years, a considerable interest has been raised on the role of humoral mediators of appetite in the pathogenesis of anorexia of CRF [5]. However, the available evidence is fragmentary and unconclusive, and often based on cross-sectional screening of plasma levels of different mediators of appetite. Leptin and ghrelin have received most attention, although other orexigenic and anorexigenic factors have also been scrutinized [5]. Leptin levels are commonly increased in patients with CRF, keeping an inverse correlation with GFR, although the basic relationship with body fat mass is also preserved, in this setting. Patients undergoing PD display particularly elevated leptin levels, as a consequence of the increased fat mass and hyperinsulinism frequently observed in these individuals [8] and, probably, of a direct stimulatory effect of intraperitoneal glucose on adipocytes [26]. Similar to normal individuals, leptin secretion is not affected by oral intake in the short term [12]. The actual role of this adipokine as a mediator of anorexia and malnutrition in patients on PD is still a matter of controversy [9,10,27]. On the other hand, plasma levels of total, but not acylated ghrelin, are frankly elevated in CRF [11], without significant differences according to the mode of dialysis therapy [28]. The role of ghrelin in the pathogenesis of anorexia of CRF has not been established, but anorexic PD patients have been claimed to present relatively low plasma ghrelin levels [29], and the exogenous administration of ghrelin has been proved to increase appetite, in this setting [13].

L cells in the distal intestine are seemingly the main source of PYY secretion. The peptide is secreted postprandially in proportion to the caloric load delivered, with a macronutrient potency of lipids being greater than that of carbohydrates, which is greater than that of proteins [30]. Secretion decays to reach nadir plasma levels during fasting periods, rising significantly after meals [31]. Plasma PYY levels start to increase within 15 min of a meal, peak at ~90 min and then remain elevated for up to 6 h [32]. A significant fraction of the full peptide ($PYY_{1–36}$) is rapidly proteolyzed by DPP4, and the cleaved product, $PYY_{3–36}$, represents the bioactive fraction of the hormone [15]. The ability of $PYY_{3–36}$ to induce satiation has been established in several recent studies, supporting a role for this peptide as a promising therapy for obesity. Peripheral $PYY_{3–36}$ administration, at doses generating physiologic postprandial blood excursions, reduces food intake and body weight in rats [16]. In obese humans, intravenous infusion of $PYY_{3–36}$ replicating postprandial concentrations inhibits appetite, decreasing buffet-meal intake by one-third, without causing nausea, affecting food palatability or altering fluid intake [18]. This is in contrast to the well-known inability of leptin and insulin to induce anorexia, in this subset. Furthermore, baseline levels of $PYY_{3–36}$ are lower in obese than in lean individuals, lending further support to the potential role for this peptide in the pathogenesis of obesity [18,33,34]. The initial reports on the appetite-inhibiting effects of $PYY_{3–36}$ surprised some investigators, because central administration of either $PYY_{1–36}$ or $PYY_{3–36}$ had been previously demonstrated to stimulate food intake [35]. The explanation to this apparent paradox appears to respond to Y receptor (YR) subtype selectivity and accessibility issues [15,16]. As such, the orexigenic effects of $PYY_{1–36}$ could be mediated by the interaction of this peptide with receptors Y1R and Y5R, which are expressed in the hypothalamic paraventricular nucleus and appear to mediate NPY-induced feeding. $PYY_{3–36}$ selectively activates Y2R and Y5R, and central administration of this peptide might increase food intake through the latter receptor. On the other hand, circulating $PYY_{3–36}$ may interact selectively with Y2R in the hypothalamic arcuate nucleus, an area believed by some to be accessible to blood-borne factors. In the hypothalamus, Y2R is a presynaptic autoinhibitory receptor of orexigenic neurons expressing both NPY and agouti-related protein (AGRP), so-named NPY/AGRP neurons. Thus, according to this hypothesis, circulating $PYY_{3–36}$ reduces food intake by inhibiting NPY/AGRP neurons through Y2R, thereby depressing adjacent anorectic melanocortin-producing cells, which are inhibited by NPY/AGRP neurons [16]. Consistent

with this, the effects of circulating PYY $_{3–36}$ on feeding behaviour are abolished by pharmacologic or genetic blockade of Y2R [16,36].

Our results show that fasting plasma levels of PYY $_{1–36}$ and PYY $_{3–36}$ are markedly increased in patients with CRF treated with PD, when compared with matched healthy controls. Moreover, plasma levels of PYY $_{1–36}$ and PYY $_{3–36}$ do not appear to change after a standardized meal or intraperitoneal glucose infusion in these patients, as opposed to the observed postprandial increase observed in controls. To our knowledge, this is the first study estimating plasma levels of PYY $_{1–36}$ and PYY $_{3–36}$ both fasting and after a mixed meal in patients with CRF treated with dialysis. Our results show some similarities with those observed in patients with anorexia nervosa [19,37], suggesting that PYY may play some role in the anorexia of CRF. The strong negative association between fasting PYY levels and fat intake observed in girls with anorexia nervosa [19] may explain the marked reductions in fat intake observed in these subjects [19,38]. Similar to obese individuals, and again in contrast to the cases of leptin and ghrelin [39,40], the nutritional drive of patients with anorexia nervosa appears to be responsive to changes in plasma PYY levels. In patients with CRF, plasma levels of PYY (both PYY $_{1–36}$ and PYY $_{3–36}$) and total ghrelin [28] are increased, while those of acylated ghrelin are not [11,12]. This association of markedly increased levels of PYY $_{3–36}$ with normal levels of acylated ghrelin sets a potentially anorexigenic scenery, in these patients.

We have previously shown that plasma total ghrelin levels decreased modestly, and plasma acyl-ghrelin more markedly, after a standardized oral feeding in patients with CRF treated with PD [12]. However, these changes were significantly attenuated, when compared with healthy controls, indicating that ghrelin secretion is partially refractory to the acute inhibitory effect of oral intake, in these patients. In a similar way, neither PYY $_{1–36}$ nor PYY $_{3–36}$ levels responded to oral intake, indicating a marked disruption in the normal secretory patterns of this peptide. Again, the combination of an attenuated but still significant decay of acylated ghrelin secretion with persistently elevated PYY $_{3–36}$ levels in the postprandial period could contribute to early satiety and malnutrition, in these patients.

Intravenous infusion of PYY to normal subjects has been found to cause a significant decrease in GFR [41]. This could give a role for increased PYY levels in the decline of residual renal function of CRF patients. On the other hand, in the control group, baseline serum PYY $_{(3–36)}$ levels were moderately but significantly higher before meal ingestion than before placebo ingestion. This difference could be fortuitous, but could also follow increased vagal activity due to an anticipation of food, in a similar way as insulin secretion [42]. The relevance of cephalic–vagal stimulation in the control of gastrointestinal peptides has been demonstrated for other peptides, including ghrelin [43], using the technique of modified sham feeding or the ‘chew and spit’ technique, in which foods are smelled, chewed and tasted, but not swallowed.

In summary, we have found that plasma levels of PYY $_{1–36}$ and PYY $_{3–36}$ are markedly increased, both fasting and after a standardized oral feeding, in patients with CRF treated with dialysis, when compared with healthy controls. Plasma PYY $_{1–36}$ and PYY $_{3–36}$ do not show the expected increase after a standardized oral feeding in patients with CRF treated with dialysis, indicating a severe disruption in the physiologic mechanisms that regulate appetite in these patients. Increased plasma levels of PYY could contribute to the anorexia and malnutrition of CRF.

Acnowledgements
The author thanks Mr Ramón Pensado and Ms Rosa Nemiña for technical assistance. This work was supported by grants PI051024 and PI070413 from Instituto de Salud Carlos III, and PS07/12 and 2006/27 from Xunta de Galicia.

Conflict of interest statement.
We declare no conflict of interest for this manuscript. We have no involvement that might rise the question of bias in the work reported or in the conclusions, implications or opinions exposed. We also state that the data presented in the manuscript are original. Also, the main data have not been published elsewhere in whole or part, except in abstract form. We specifically state that part of the crude data on baseline hormone levels (but not those subject of the present study) depicted in Table 1 were presented in the journal *Kidney International* in 2005, as stated in the Method description (page 6, Section Population and Method-Protocol).

References

1. Fouque D, Kopple JD. Malnutrition and dialysis. In: Hörl WH, Koch, KM, Lindsay RM, Ronco C and Winchester JF (eds). Replacement of Renal Function by Dialysis. Dordrecht: Kluwer, 2004, 1013–1039
2. Mitch WE. Malnutrition: a frequent misdiagnosis for hemodialysis patients. J Clin Invest 2002; 11: 437–439
3. Bergstrom J. Nutrition and mortality in hemodialysis. J Am Soc Nephrol 1995; 6: 1329–1341
4. Wang T, Heimburger O, Bergstrom J et al. Nutritional problems in peritoneal dialysis: an overview. Perit Dial Int 1999; 19: S297–S303
5. Bossola M, Tazza L, Giungi S et al. Anorexia in haemodialysis patients: an update. Kidney Int 2006; 70: 417–422
6. Coll AP, Farooqi IS, O'Rahilly S. The hormonal control of food intake. Cell 2007; 129: 251–262
7. Heimbürger O, Stenvinkel P. Adipokines in chronic kidney disease—fat tissue gives nephrologists a message. Perit Dial Int 2005; 25: 340–342
8. Pérez Fontan M, Rodríguez-Carmona A, Cordido F et al. Hyperleptinemia in uremic patients undergoing conservative treatment, peritoneal dialysis and hemodialysis: a comparative analysis. Am J Kidney Dis 1999; 34: 824–831
9. Stenvinkel P, Lindholm B, Lohnqvist F et al. Increases in serum leptin during peritoneal dialysis are associated with inflammation and a decrease in lean body mass. J Am Soc Nephrol 2000; 11: 1303–1309
10. Rodríguez-Carmona A, Pérez-Fontán M, Cordido F et al. Hyperleptinemia is not correlated with markers of protein malnutrition in chronic renal failure. A cross-sectional study in predialysis, peritoneal dialysis and hemodialysis patients. Nephron 2000; 86: 274–280
11. Yoshimoto A, Mori K, Sugawara A et al. Plasma ghrelin and desacyl ghrelin concentratins in renal failure. J Am Soc Nephrol 2002; 13: 2748–2752
12. Pérez Fontán M, Cordido F, Rodríguez-Carmona A et al. Acute plasma ghrelin and leptin responses to oral feeding or intraperitoneal hypertonic glucose-based dialysate in patients with chronic renal failure. Kidney Int 2005; 68: 2877–2885
13. Wynne K, Bloom SR. The role of oxyntomodulin and peptide tyrosine-tyrosine (PYY) in appettite control. Nat Clin Pract Endocrino Metabol 2006; 2: 612–620
14. Murphy KG, Dhillo WS, Bloom SR. Gut peptides in the regulation of food intake and energy homeostasis. Endocr Rev 2006; 27: 719–727
15. Cummings DE, Overduin J. Gastrointestinal regulation of food intake. J Clin Invest 2007; 117: 13–23
16. Batterham RL, Cowley MA, Small CJ et al. Gut hormone PYY (3–36) physiologically inhibits food intake. Nature 2002; 418: 650–654
17. Chaudhri OB, Field BCT, Bloom S. From gut to mind—hormonal satiety signals and anorexia nervosa. J Clin Endocrinol Metab 2006; 91: 797–798
18. Batterham RL, Cohen MA, Ellis SM et al . Inhibition of food intake in obese subjects by peptide YY 3–36. New Engl J Med 2003; 349: 941–948
19. Misra M, Miller KK, Tsai P et al. Elevated peptide YY levels in adolescent girls with anorexia nervosa. J Clin Endocrinol Metab 2006; 91: 1027–1033
20. Matthews DR, Hosker HP, Rudenski AS et al. Homeostasis model assessment: insulin resistance and beta-cell function from fasting plasma glucose and insulin concentrations in man. Diabetologia 1985; 28: 412–419
21. Szeto CC, Lai KN, Wong TYH et al. Independent effects of residual renal function and dialysis adequacy on nutritional status and patients outcome in continuous ambulatory peritoneal dialysis. Am J Kidney Dis 1999; 34: 1056–1064
22. Van Vlem BA, Schoonjans RS, Struijk DG et al. Influence of dialysate on gastric emptying time in peritoneal dialysis patients. Perit Dial Int 2002; 22: 32–38
23. Mamoun AH, Anderstam B, Södersten P et al. Influence of peritoneal dialysis solutions with glucose and aminoacids on ingestive behavior in rats. Kidney Int 1996; 49: 1276–1282
24. Zheng ZH, Sederholm F, Anderstam B et al. Acute effects of peritoneal dialysis solutions on appetite in non-uremic rats. Kidney Int 2001; 60: 2392–2398
25. Sutton D, Higgins B, Stevens JM. Continuous ambulatory peritoneal dialysis patients are unable to increase dietary intake to recommended levels. J Ren Nutr 2007; 17: 329–335
26. Teta D, Maillard M, Tedjani A et al. The effect of pH-neutral peritoneal dialysis fluids on adipokine secretion from cultured adipocytes. Nephrol Dial Transplant 2007; 22: 862–869
27. Bossola M, Muscaritoli M, Valenza V et al. Anorexia and serum leptin levels in hemodialysis patients. Nephron Clin Pract 2004; 97: c76–c82
28. Pérez Fontán M, Cordido F, Rodríguez-Carmona A et al. Plasma ghrelin levels in patients undergoing haemodialysis and pertitoneal dialysis. Nephrol Dial Transplant 2004; 19: 2095–2100
29. Aguilera A, Cirugeda A, Amair R et al. Ghrelin plasma levels and appetite in peritoneal dialysis patients. Adv Perit Dial 2004; 20: 194–199
30. Degen L, Oesch S, Casanova M et al. Effect of peptide YY 3–36 on food intake in humans. Gastroenterology 2005; 129: 1430–1436
31. Soffer EE, Adrian TE, Launspach J et al. Meal-induced secretion of gastrointestinal regulatory peptides is not affected by sleep. Neurogastroenterol Motil 1997; 9: 7–12
32. Adrian TE, Ferri GL, Bacarese-Hamilton AJ et al. Human distribution and release of a putative new gut hormone, peptide YY. Gastroenterology 1985; 89: 1070–1077

33. Korner J, Leibel RL. To eat or not to eat—how the gut talks to the brain. New Engl J Med 2003; 349: 926–928
34. Álvarez-Bartolomé M, Borque M, Martínez-Sarmiento J et al. Peptide YY secretion in morbidly obese patients before and after vertical banded gastroplasty. Obes Surg 2002; 12: 324–327
35. Hagan M. Peptide YY: a key mediator of orexigenic behavior. Peptides 2002; 23: 377–382
36. Scott V, Kimura N, Stark JA et al . Intravenous peptide YY 3–36 and Y2 receptor antagonism in the rat: effects on feeding behaviour. J Neuroendocrinol 2005; 17: 452–457
37. Pfluger PT, Kampe J, Castaneda TR et al. Effect of human body weight changes on circulating levels of peptide YY and peptide YY 3–36. J Clin Endocrinol Metab 2007; 92: 583–588
38. Affenito SG, Dohm FA, Crawford PB et al. Macronutrient intake in anorexia nervosa: the national heart, lung, and blood institute growth and health study. J Pediatr 2002; 141: 701–705
39. Nedvidkova J, Krykorkova I, Bartak V et al. Loss of meal-induced decrease in plasma ghrelin levels in patients with anorexia nervosa. J Clin Endocrinol Metab 2003; 88: 1678–1682
40. Misra M, Miller KK, Kuo K et al. Secretory dynamics of leptin in adolescent girls with anorexia nervosa and healthy adolescents. Am J Physiol Endocrinol 2005; 289: E373–E381
41. Playford RJ, Mehta S, Upton P et al. Effect of peptide YY on human renal function. Am J Physiol 1995; 268: F754–F759
42. Hojlund K, Wildner-Christensen M, Eshoj O et al. Reference intervals for glucose, β-cell polypeptides and counterregulatory factors during prolonged fasting. Am J Physiol Endocrinol Metab 2001; 280: E50–E58
43. Arosio M, Ronchi CL, Beck-Peccoz P et al. Effects of modified sham feeding on ghrelin levels in healthy human subjects. J Clin Endocrinol Metab 2004; 89: 5101–5104