Nephrol Dial Transplant (2010) 25: 985–992
doi: 10.1093/ndt/gfp566
Advance Access publication 23 October 2009

Glucose tolerance before and after renal transplantation

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Abstract

Background. Renal insufficiency predisposes to insulin resistance, hyperparathyroidism and derangements in calcium phosphate and nitrogenous compound balance, leading to pre-transplant hyperglycaemia. These metabolic risk factors are not fully corrected after renal transplantation. The present study aimed to assess the role of pre-transplant glycaemia and the named metabolic risk factors in post-transplant hyperglycaemia [PHYG; impaired fasting glucose (IFG), impaired glucose tolerance (IGT) or diabetes mellitus (DM)].

Methods. This is a retrospective cohort study involving 301 patients without pre-transplant DM. Measurements included a pre- and post-transplant oral glucose tolerance test (OGTT) as well as glomerular filtration rate (GFR), parathyroid hormone (PTH), phosphate, calcium and urea measured 10 weeks post-transplant. The risk of PHYG at 10 weeks post-transplant was analysed using multiple logistic regression.

Results. Ninety-three patients (31%) had PHYG (two IFG, 52 IGT, 39 DM). Variables associated with PHYG included pre-transplant 2-h glycaemia [OR 1.26, 95% CI (1.09, 1.46)] and post-transplant urea levels [OR 1.14, 95% CI (1.02, 1.27)]. Older age, non-Caucasian ethnicity, previous transplants, ≥3 HLA class 1 mismatches and high prednisolone doses were likewise associated with an increased PHYG risk (all $P < 0.05$).

Conclusions. Pre-transplant glycaemia and high post-transplant levels of urea were associated with a greater risk of PHYG. This seemed to be independent of GFR, PTH, phosphate, calcium and traditional risk factors such as age and glucocorticoid load.

Keywords: hyperglycaemia; multiple imputation; oral glucose tolerance test; renal transplantation; urea

Introduction

New-onset diabetes mellitus after transplantation (NODAT) is promoted by old age, non-Caucasian ethnicity and immunosuppressive drugs [1–4]. NODAT develops in 10–20% of patients after renal transplantation (RTx) and is known to reduce both patient and graft survival. In the general population [5–7] and RTx patients [8,9], even non-diabetic hyperglycaemia appears to predict mortality and cardiovascular events. Since both diabetic [diabetes mellitus (DM)] and non-diabetic hyperglycaemia [impaired fasting glucose (IFG) or impaired glucose tolerance (IGT)] are amenable to intervention post-transplant [10], early identification of patients at risk of developing any form of post-transplant hyperglycaemia is of clinical importance to prevent long-term complications.

Post-transplant hyperglycaemia (PHYG) reflects both pre-transplant and transplant-induced abnormalities in glucose metabolism. In non-diabetic patients with advanced chronic kidney disease (CKD), insulin metabolism is often abnormal [11]. Impairments in insulin secretion have been linked to calcium and parathyroid hormone (PTH) levels [12,13]. Insulin resistance appears to be associated with the retention of nitrogenous compounds such as urea, since both dialysis and low-protein diets improve insulin sensitivity in terminal CKD [14]. These metabolic derangements have received surprisingly little attention post-transplant [1–4].

In this study, pre-transplant glycaemia assessed by a standard oral glucose tolerance test (OGTT) was included in a model to predict PHYG 10 weeks post-transplant. The model also included metabolites related to post-transplant renal impairment, in addition to classical NODAT risk factors.

Materials and methods

Study population and design

This is a retrospective cohort study using PHYG as the outcome of interest. All patients who underwent RTx in Norway between October 2003 and October 2005 were considered ($n = 500$). All were transplanted at our centre. The following were exclusion criteria: overt pre-transplant DM (diabetic nephropathy or DM as a secondary diagnosis defined prior to the RTx work-up by fasting plasma glucose ≥7.0 mmol/L, random plasma glucose ≥11.1 mmol/L or the use of anti-diabetic medication) [15], age <18 years and loss to follow-up. A total of 301 patients were included; the disposition of patients is shown in Figure 1. Main primary renal diagnoses among in-

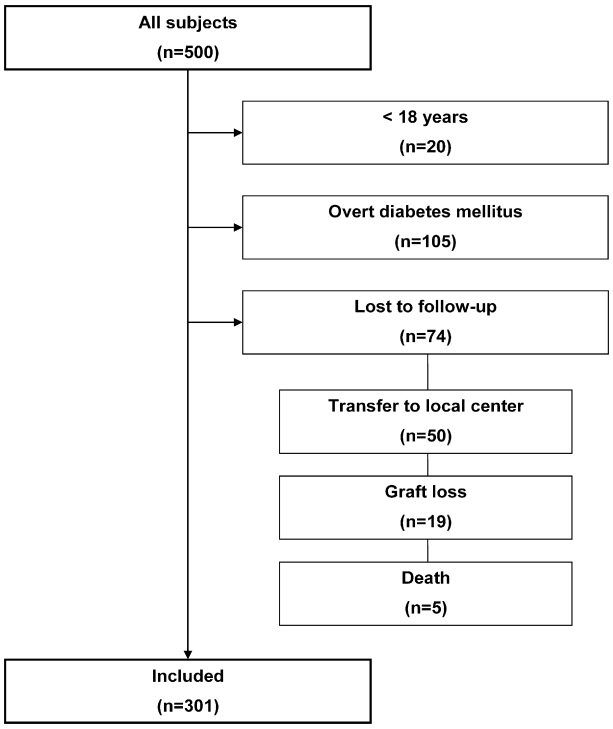

Fig. 1. Patient disposition. Patients with overt diabetes mellitus pre-transplant were excluded. Fifty patients were lost to follow-up due to transfer to hospitals in other parts of the country; loss to follow-up was otherwise defined by graft loss or death.

cluded patients were primary glomerulonephritis (37%), nephrosclerosis (19%), autosomal dominant polycystic kidney disease (17%) and other interstitial nephropathies (14%). All patients gave their written informed consent for the use of their data. The project was approved by the local Data Inspectorate and performed in accordance with the Declaration of Helsinki.

Data collection and laboratory methods

Human leukocyte antigen (HLA) was typed using immunomagnetic methods [16]. With the exception of glucose, other laboratory data were collected in the fasting state at 10 weeks post-transplant. Plasma clearance of ^{51}Cr-EDTA provided the glomerular filtration rate (GFR) estimate. Phosphate was measured by ammonium molybdate methods, intact PTH by chemiluminescence immunoassays, and ionized calcium (iCa) by potentiometric methods. Reported iCa values were measured at the actual pH of each patient; phosphate was measured at a precision level of one decimal place.

Patients of European ancestry were defined as Caucasians, while non-Caucasians included patients with ethnic origins in Central Africa, the Middle East and Central or South-East Asia. None of the patients was subjected to dietary restrictions post-transplant.

Pre-transplant glucose tolerance

Non-diabetic patients awaiting RTx in Norway perform a mandatory standard 75-g OGTT at local centres. Numeric OGTT results are enclosed with the referral for RTx and specify whether plasma or whole blood was used. Two hundred and thirty-nine patients completed the OGTT, at a median of 50 (32–82) weeks pre-transplant. Results were reported in plasma for all but two patients and therefore considered plasma equivalent. Sixty-two patients had only fasting glucose (n = 38) or no glucose measured (n = 24). Despite scrutiny of clinical records, including complete referrals, no pre-transplant DM or other likely explanation could be found for the omission of these data. Missing observations were thus assumed to be missing at random (MAR; detailed below), and the patients were included in the analysis. Consequently, a total of 301 patients qualified for participation in the study. Fasting (FPG) and 2-h post-challenge plasma glucose (2h-PG) are reported.

Post-transplant glucose tolerance

A repeat OGTT was performed 10 weeks post-transplant in all patients, except if NODAT had been diagnosed between RTx and the 10-week data collection [by fasting plasma glucose ≥7.0 mmol/L (≥126 mg/dL), random plasma glucose ≥11.1 mmol/L (≥200 mg/dL) or requirement of anti-diabetic medication] [15]. This OGTT was analysed in venous whole blood by a modified glucose dehydrogenase method. Using the World Health Organization criteria for venous whole blood samples, the primary endpoint, PHYG, was defined either as NODAT diagnosed ahead of the OGTT or as an abnormal post-transplant OGTT equivalent to DM [fasting glucose ≥6.1 mmol/L (≥110 mg/dL) and/or 2-h glucose ≥10.0 mmol/L (≥180 mg/dL)], IGT [fasting glucose <6.1 mmol/L (<110 mg/dL) and 2-h glucose 6.7–9.9 mmol/L (120–179 mg/dL)] or IFG [fasting glucose 5.6–6.0 mmol/L (100–109 mg/dL) and 2-h glucose <6.7 mmol/L (<120 mg/dL)] [15]. Glycaemia below these levels was termed normal glucose metabolism (NGM). Although PHYG was diagnosed using whole blood, glucose is reported as plasma equivalent (FPG and 2h-PG) in accordance with international recommendations (mmol/L plasma = 1.11 * mmol/L whole blood) [17].

Immunosuppressive therapy

Methylprednisolone was administered intravenously on Days 0 and 1 (500 and 80 mg, respectively). Thereafter, oral prednisolone was tapered from 80 (Day 2) to 10 mg/day (>8 weeks). Mycophenolate was given as 1000 mg twice daily. Cyclosporine A or tacrolimus were given to obtain 8-week trough levels of about 150–200 or 5–8 μg/L, respectively. Tacrolimus was primarily used if cyclosporine A was withdrawn due to rejection, toxicity or side effects. Acute rejections (biopsy-proven or >20% increase in creatinine) were treated with intravenous methylprednisolone (Day 0: 500 mg; Days 1–3: 250 mg/day), and subsequent increments in oral prednisolone. Cumulative glucocorticoid doses were calculated from standard regimens for the duration of follow-up, with protocol increments for acute rejections, and reported as prednisolone-equivalent. Methylprednisolone doses were multiplied by 1.25 to achieve prednisolone-equivalent doses [18].

Missing data

Eighty-four patients (28%) had ≥1 missing data entry (Table 1). Pre-transplant 2h-PG was the most frequently missing entry (n = 62, 21%). Patients with incomplete data were leaner and less often first-kidney recipients compared to complete cases. Since no patients had known pre-transplant DM, unmeasured OGTT results were likely to be similar to those that were measured, when controlling for previous transplants, body mass index (BMI) and other factors. Data were therefore analysed using the MAR assumption, which states that missing observations may depend on observed quantities but not on the missing values themselves [19]. Multiple imputation (MI) was used to generate 10 different data set copies, each containing 301 complete cases. Complete and incomplete variables were used as predictors during imputation. All missing observations were imputed, except for post-transplant OGTT results (missing only in NODAT patients, i.e. not MAR). Variables used for MI included: PHYG (yes), pre- and post-transplant fasting and 2-h glucose, pre-dialytic pre-transplant OGTT (yes), age, gender, ethnicity, donor type, previous transplants (yes), time on dialysis, HLA-1 mismatch, BMI pre- and post-transplant, GFR, phosphate, ionized calcium, PTH, urea, haemoglobin, prednisolone dose, furosemide (yes), beta blocker (yes) and missing ≥1 observation (yes). Statistical analyses were first performed using complete data, then repeated on each imputed data set, and finally pooled to achieve single parameter estimates.

Statistical analysis

Data are reported as mean (SD), median (interquartile range, IQR) or frequencies (%). Groups were compared using independent samples t-test, Mann–Whitney or χ^2 test, with s Fisher's exact test for sparse 2 × 2 tables (expected cell frequency <5). Continuous variables were compared using

Table 1. Comparison of subjects with zero vs. ≥1 missing observation[a]

	Complete (n = 217)	#	≥1 missing (n = 84)[b]	#	P
Patient characteristics					
Age (years)	50.9 (14.6)	0	50.5 (14.4)	0	0.850
Female gender (yes)	72 (33%)	0	35 (42%)	0	0.168
Non-Caucasian ethnicity (yes)	8 (4%)	0	5 (6%)	0	0.362
Pre-transplant BMI (kg/m^2)[c]	25.4 (4.0)	0	24.5 (4.1)	27	0.119
Post-transplant BMI (kg/m^2)[c]	24.9 (3.8)	0	23.9 (3.5)	6	0.047
Previous renal transplants (yes)	23 (11%)	0	21 (25%)	0	0.002
Time since transplantation (days)	71 (8)	0	71 (10)	0	0.695
Time on dialysis (weeks)	40 (7–78)	0	42 (0–83)	0	0.861
Living donor (yes)	99 (46%)	0	28 (33%)	0	0.053
≥3 HLA class 1 mismatches (yes)	71 (33%)	0	32 (38%)	0	0.378
≥1 HLA class 2 mismatches (yes)	132 (61%)	0	46 (51%)	0	0.337
Cold ischaemic time (h)	6 (3–14)	0	10 (4–15)	0	0.125
Acute rejection <10 weeks (yes)	77 (36%)	0	30 (36%)	0	0.970
Medication at 10 weeks post-transplant					
Prednisolone (mg/kg/day)	0.16 (0.13–0.21)	0	0.17 (0.13–0.23)	0	0.274
CSD (mg/kg)	20.8 (17.3–40.9)	0	25.2 (17.6–44.4)	0	0.267
Cyclosporine A (yes)	175 (81%)	0	65 (77%)	0	0.527
Tacrolimus (yes)	31 (14%)	0	18 (21%)	0	0.132
Furosemide (yes)	76 (35%)	0	32 (38%)	7	0.618
Beta blocker (yes)	110 (51%)	0	45 (54%)	5	0.654
Pre-transplant laboratory results					
Pre-transplant FPG (mmol/L)	5.0 (4.6–5.4)	0	4.9 (4.5–5.4)	24	0.503
Pre-transplant 2h-PG (mmol/L)	6.6 (5.4–8.3)	0	6.4 (5.1–7.9)	62	0.222
Post-transplant laboratory results					
Phosphate (mmol/L)	0.84 (0.18)	0	0.88 (0.26)	0	0.218
iCa (mmol/L)	1.33 (0.09)	0	1.34 (0.08)	9	0.544
PTH (pmol/L)	11.1 (7.6–16.6)	0	10.4 (7.8–15.4)	7	0.697
Urea (mmol/L)	9.6 (3.4)	0	10.9 (6.7)	0	0.078
Creatinine (μmol/L)	124 (36)	0	123 (48)	0	0.803
GFR (ml/min/1.73 m^2)	54 (14)	0	54 (16)	12	0.813
Haemoglobin (g/dL)	11.8 (1.5)	0	11.8 (1.5)	0	0.796
FPG (mmol/L)[d]	4.5 (3.9–4.9)	0	4.5 (3.9–4.9)	0	0.993
2h-PG (mmol/L)[d]	5.2 (4.1–6.8)	0	5.2 (4.3–6.6)	0	0.877
PHYG (yes)	64 (31%)	0	29 (35%)	0	0.397

[a]Mean (SD), median (interquartile range) or frequencies (%), with comparisons by independent samples t-test, Mann–Whitney or χ^2 test as appropriate.
[b]Pooled results for all 84 subjects, i.e. after imputation of missing observations whenever present.
#, number of missing observations imputed prior to pooling.
iCa, ionized calcium; CSD, cumulative steroid dose; FPG, fasting plasma glucose; 2h-PG, 2-h post-challenge plasma glucose.

parametric (Pearson's r) or non-parametric correlations (Spearman's Rho) and repeated measurements using paired samples or Wilcoxon signed rank test.

The outcome (PHYG) was assessed using simple multiple logistic regression, followed by backward and forward stepwise regressions to confirm results using −2 log likelihood (LL) cut-offs of $P < 0.20$ and >0.25 for entry and removal, respectively. Explanatory variables [age, gender, ethnicity, donor type, previous transplants, BMI, prednisolone, GFR, urea, PTH, phosphate, iCa and pre-transplant glucose (FPG or 2h-PG)] were included a priori. Four additional variables were included as confounders for donor type (time on dialysis, HLA mismatch), age and laboratory results (beta blocker, furosemide). This produced >5 events per variable (EPV), which has been shown to produce similar results as compared to the commonly employed but probably overly strict 10 EPV rule of thumb [20]. Urea and pre-transplant 2h-PG displayed near-linear increments in the log odds of PHYG with increasing categories (Figures 2A and B) and were included as continuous covariates; other continuous variables were categorized for logistic analyses (Table 2). The lowest categories (tertile 1) were used as reference. Model fit was confirmed in all models (Hosmer–Lemeshow test $P > 0.2$). Multicollinearity (r or Rho > 0.7) was observed [cold ischaemic time (donor type); acute rejection (cumulative glucocorticoid dose); GFR (plasma creatinine); all r or Rho > 0.7] but not among included variables. Models with and without glucose measurements were compared using the LL method. Two-tailed $P < 0.05$ was considered statistically significant. Analyses were implemented using SPSS 17.0 (SPSS, Chicago, IL).

Results

Multiple imputation (MI, n = 301) allowed for inclusion of 84 subjects who would have been lost in complete case (CC, n = 217) analysis. Subjects with zero vs. ≥1 missing observation are compared in Table 1. Table 2 displays the categories of continuous variables used in logistic regression. With two notable exceptions (null effect relating to age and ethnicity in CC analysis), CC and MI data produced similar overall results. For the purpose of clarity, only MI results are presented; CC results are available in the online Appendix.

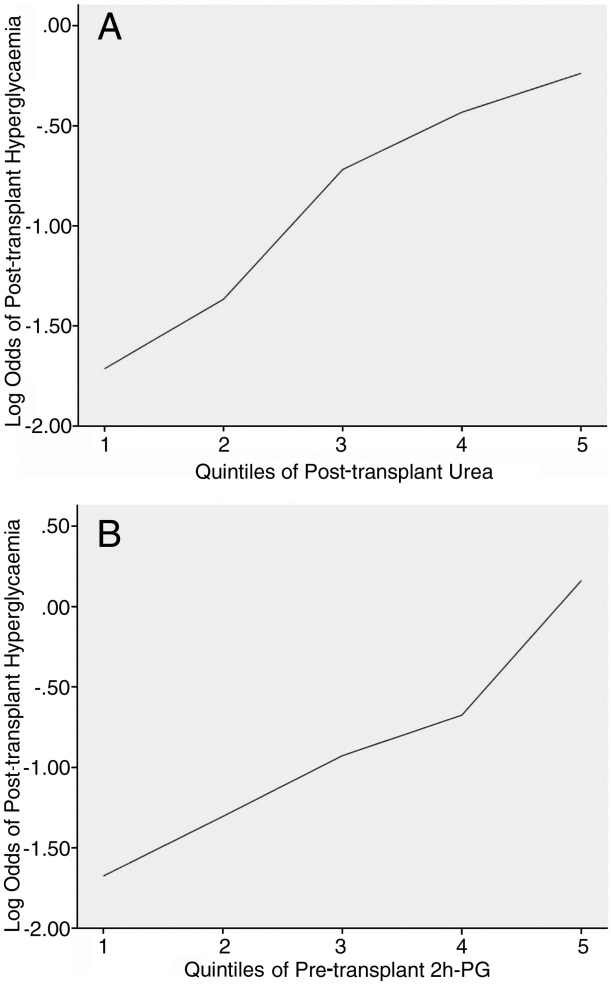

Fig. 2. Assumption of linearity. The use of continuous covariates in logistic regression assumes that the log odds of the outcome progresses linearly across categories of a continuous covariate. Only urea and pre-transplant 2-h plasma glucose (2h-PG) met this assumption. The figures show the approximately linear increase in the log odds of post-transplant hyperglycaemia across categories (quintiles) of urea (**A**) or pre-transplant 2h-PG (**B**).

Patient characteristics and drug use are presented in Tables 3 and 4. PHYG was observed in 93 patients (31%; two IFG, 52 IGT, 39 DM). Twenty of 39 patients with DM (51%) were diagnosed prior to 10 weeks and did not undertake the post-transplant OGTT. Among the other 73 patients diagnosed with PHYG at 10 weeks, only 11 patients (15%) were diagnosed using fasting glucose alone (nine DM, two IFG); the remaining proportion [n = 62 (85%); 52 IGT, 10 DM] was diagnosed by the 2-h glucose. Compared to NGM patients, PHYG patients were older, had longer pre-transplant dialysis times, fewer living donations, longer cold ischaemic times and poorer HLA class 1 matching and received higher glucocorticoid doses. BMI dropped between the pre- and post-transplant recording in both PHYG and NGM patients (both $P = 0.002$); the magnitude of the BMI reduction was similar (0.8 vs. 0.4 kg/m^2, $P = 0.203$).

Patients diagnosed with PHYG had higher glucose levels pre-transplant compared to patients with NGM (Table 5). A 1-mmol/L increment in pre-transplant FPG and 2h-PG, respectively, predicted a 122% and 27% increase in the risk of PHYG [crude OR 2.22 (95% CI 1.46–3.39) and 1.27 (95% CI 1.12–1.44)]. Pre-transplant glycaemia correlated with post-transplant glucose and urea levels but not with other post-transplant laboratory results and was comparable whether patients were pre-dialytic (62%) or not at the time of investigation (FPG 5.0 vs. 4.9, $P = 0.194$; 2h-PG 6.6 vs. 6.7 mmol/L; $P = 0.430$). In patients with PHYG, FPG and 2h-PG were significantly higher post- as compared to pre-transplant. In NGM patients, the opposite result was observed for both parameters (all $P < 0.001$) (Table 5).

PHYG patients had higher post-transplant levels of urea compared to NGM patients, while PTH, phosphate, ionized calcium, creatinine, GFR and haemoglobin (Hb) were similar (Table 5). A 1-mmol/L increment in plasma urea conferred a 14% increase in the crude risk of PHYG [crude OR 1.14 (95% CI 1.07–1.21)]. A similar result was observed in each category of GFR [OR 1.17 (1.05–1.31), 1.19 (1.03–1.37) and 1.35 (1.06–1.71), tertile 1–3, respectively]. Urea levels correlated with pre-transplant FPG,

Table 2. Tertiles of continuous variables[a]

	Tertile 1			Tertile 2			Tertile 3			Total		
	M	Range	*n*	*M*	Range	*n*	*M*	Range	*n*	*M*	Range	*n*
Age at transplant (years)[b]	34.0	(18.1–45.2)	102	52.0	(45.5–58.3)	98	66.5	(58.3–78.8)	101	50.8	(18.1–78.8)	301
BMI post-transplant (kg/m^2)[b]	20.8	(16.3–22.6)	100	24.3	(22.8–25.9)	101	28.7	(25.9–38.7)	100	28.7	(16.3–38.7)	301
Time on dialysis (weeks)[c]	0	(0–0)	74	33	(3–60)	116	94	(61–696)	111	58	(0–696)	301
Prednisolone (mg/kg)[c]	0.13	(0.09–0.14)	101	0.16	(0.15–0.20)	100	0.25	(0.21–0.68)	100	0.16	(0.09–0.68)	301
GFR (ml/min/1.73 m^2)[b]	39	(8–47)	103	54	(48–60)	100	70	(61–108)	98	54	(8–108)	301
iCa (mmol/L)[b]	1.24	(1.04–1.29)	94	1.32	(1.30–1.35)	97	1.42	(1.36–1.70)	110	1.33	(1.04–1.70)	301
Phosphate (mmol/L)[b,d]	0.7	(0.3–0.8)	159	0.9	(0.9–0.9)	64	1.1	(1.0–2.4)	78	0.9	(0.3–2.4)	301
PTH (pmol/L)[c]	6.5	(1.1–8.4)	100	10.9	(8.4–14.5)	101	18.7	(14.6–155.7)	100	10.9	(1.1–155.7)	301

[a]Within each tertile, *M* represents either the mean or the median value. Range indicates the highest and lowest value in each tertile. Prednisolone dose and laboratory values assessed at 10 weeks post-transplant.
[b]*M* represents mean.
[c]*M* represents median.
[d]Phosphate tertiles are skewed due to a large number of identical phosphate concentrations contained in the lower tertile.
iCa, ionized calcium.

Table 3. Patient characteristics[a]

	All (n = 301)	NGM (n = 208)	PHYG (n = 93)	P[b]
Age (years)	50.8 (14.5)	49.3 (13.8)	54.2 (15.6)	0.007
Female gender (yes)	107 (36%)	75 (36%)	32 (34%)	0.782
Non-Caucasian ethnicity (yes)	13 (4%)	5 (2%)	8 (9%)	0.027
Pre-transplant BMI (kg/m^2)[c]	25.1 (4.0)	24.9 (3.9)	25.6 (4.4)	0.185
Post-transplant BMI (kg/m^2)[c]	24.6 (3.7)	24.5 (3.5)	24.8 (4.1)	0.518
Previous renal transplants (yes)	44 (15%)	26 (13%)	18 (19%)	0.120
Time since transplantation (days)	71 (9)	72 (9)	71 (8)	0.403
Time on dialysis pre-transplant (weeks)	41 (4–81)	34 (0–72)	50 (21–94)	0.006
Living donor (yes)	127 (42%)	98 (47%)	29 (31%)	0.010
≥3 HLA class 1 mismatches (yes)	103 (34%)	63 (30%)	40 (43%)	0.032
≥1 HLA class 2 mismatches (yes)	178 (59%)	123 (59%)	55 (59%)	0.999
Cold ischaemic time (h)	7 (3–15)	6 (3–14)	11 (4–16)	0.046
Acute rejection <10 weeks (yes)	107 (36%)	63 (30%)	44 (47%)	0.004

[a]Mean (SD), median (interquartile range) or frequencies. Pooled mean with mean standard deviations are given for data containing imputed values.
[b]PHYG vs. NGM by independent samples t-test, Mann–Whitney or χ^2 test as appropriate.
[c]Pre- to post-transplant BMI dropped significantly in both groups (P = 0.002 by paired samples t-test).
NGM, normal glucose metabolism; PHYG, post-transplant hyperglycaemia.

post-transplant 2h-PG, age, male gender, the occurrence of acute rejections or presence of ≥3 HLA class 1 mismatches, plasma phosphate, prednisolone dose (mg/kg), beta blocker or furosemide use and the time spent on dialysis pre-transplant (all $P < 0.05$). An inverse correlation was observed between urea and GFR (r −0.59), albumin and Hb (all $P \leq 0.001$).

Crude results were adjusted by multiple regression as shown in Table 6. Results of stepwise procedures were almost identical to those presented (not shown). A 1-mmol/L increment in pre-transplant 2h-PG predicted a PHYG risk increase of 26% (adjusted OR 1.26, 95% CI 1.09–1.46). A similar result was achieved using FPG rather than 2h-PG (adjusted OR 2.20, 95% CI 1.29–3.75). As in univariable analysis, a 1-mmol/L increment in urea was associated with a 14% increment in the risk of PHYG (adjusted OR 1.14, 95% CI 1.02–1.27).

The inclusion of either FPG or 2h-PG each increased the discriminative ability of the model as compared to omitting pre-transplant glucose entirely [Nagelkerke R^2 0.30, 0.32 and 0.26 for FPG, 2h-PG and omission of glucose, respectively; LL statistic 8.80 (P = 0.003) and 16.26 ($P < 0.001$) for FPG or 2h-PG, each compared to omission]. Addition of 2h-PG to a model already containing FPG significantly improved the model (R^2 increased from 0.30 to 0.33; LL statistic 11.60, $P < 0.001$), while adding FPG to a model already containing 2h-PG did not, statistically, add significant information (R^2 increased from 0.32 to 0.33; LL statistic 3.39, P = 0.066). Pre-transplant 2h-PG was therefore more strongly associated with PHYG than was pre-transplant FPG.

In addition to pre-transplant glycaemia and high post-transplant urea levels, older age, non-Caucasian ethnicity, previous transplants, ≥3 HLA class 1 mismatches and high prednisolone doses were associated with a higher risk of PHYG. No significant associations were observed between PHYG and PTH, phosphate or iCa, and no significant impact of gender or BMI could be demonstrated. Patients with PHYG had a slightly elevated GFR compared to NGM patients having the same age and urea level (difference of 3 ml/min/1.73 m^2, result not shown). A missing data variable (MI performed = yes) was created in each MI data set to test whether results were confounded by the imputation itself. This variable was statistically insignificant (P = 0.832), left other results unaltered and was discarded.

Discussion

The main findings of this study were the direct associations between pre- and post-transplant glycaemia and also

Table 4. Medication at 10 weeks after transplantation[a]

	All (n = 301)	NGM (n = 208)	PHYG (n = 93)	P[b]
Prednisolone (mg/kg/day)	0.16 (0.13–0.21)	0.16 (0.13–0.20)	0.20 (0.13–0.28)	0.003
CSD (mg/kg)	21.6 (17.4–42.8)	21.0 (17.2–38.2)	26.4 (17–47.8)	0.037
Cyclosporine A (yes)	240 (80%)	166 (80%)	74 (80%)	0.962
Tacrolimus (yes)	49 (16%)	33 (16%)	16 (17%)	0.771
Furosemide (yes)	108 (36%)	63 (30%)	45 (48%)	0.003
Beta blocker (yes)	155 (51%)	97 (47%)	58 (62%)	0.011

[a]Median (interquartile range) or frequencies.
[b]PHYG vs. NGM by Mann–Whitney or χ^2 test as appropriate.
CSD, cumulative steroid dose; NGM, normal glucose metabolism; PHYG, post-transplant hyperglycaemia.

Table 5. Laboratory results[a]

	All (n = 301)	NGM (n = 208)	PHYG (n = 93)	P[b]
10 weeks after renal transplantation				
Phosphate (mmol/L)	0.8 (0.20)	0.8 (0.18)	0.9 (0.24)	0.059
iCa (mmol/L)	1.33 (0.09)	1.34 (0.09)	1.33 (0.10)	0.463
PTH (pmol/L)	10.9 (7.7–16.2)	10.9 (7.7–15.4)	10.8 (7.6–17.3)	0.849
Urea (mmol/L)	9.9 (4.6)	9.2 (3.4)	11.7 (6.2)	<0.001
Creatinine (μmol/L)	124 (40)	122 (36)	126 (47)	0.447
GFR (ml/min/1.73 m^2)	54 (15)	54 (15)	53 (15)	0.478
Haemoglobin (g/dL)	11.8 (1.5)	11.8 (1.5)	11.8 (1.5)	0.948
FPG (mmol/L)[c]	5.0 (4.3–5.4)	4.8 (4.2–5.1)[d]**	5.8 (5.2–6.3)[e]**	<0.001[c]
2h-PG (mmol/L)[c]	5.8 (4.7–7.4)[d]*	5.2 (4.4–6.0)[d]**	8.8 (8.0–10.7)[e]**	<0.001[c]
Before transplantation				
FPG (mmol/L)	5.0 (4.6–5.4)	4.9 (4.5–5.3)	5.2 (4.9–5.6)	<0.001
2h-PG (mmol/L)	6.5 (5.3–8.2)	6.2 (5.1–7.5)	7.4 (6.0–9.0)	<0.001

[a]Mean (SD) or median (interquartile range). For normally distributed data containing imputed values, pooled means and mean standard deviations are reported. For data with non-normal distribution containing imputed values, medians and interquartile ranges represent the medians among imputed datasets.
[b]PHYG vs. NGM by independent samples t-test or Mann–Whitney U-test as appropriate.
[c]n = 281; oral glucose challenge was not performed in 20 patients who had been diagnosed with diabetes mellitus <10 weeks post-transplant. Their post-transplant glucose levels were not missing at random and therefore not imputed. Plasma values are reported as achieved through multiplication of the original whole blood measurements by a factor of 1.11 [17].
[d]Significant decrease compared to the corresponding pre-transplant result (*P < 0.005 or **P < 0.001 by Wilcoxon signed rank test).
[e]Significant increase compared to the corresponding pre-transplant result (*P < 0.005 or **P < 0.001 by Wilcoxon signed rank test).
iCa, ionized calcium; FPG, fasting plasma glucose; NGM, normal glucose metabolism; PHYG, post-transplant hyperglycaemia; 2h-PG, 2 h post-challenge plasma glucose.

the association between post-transplant urea levels and PHYG. These relationships appeared to be continuous and independent of GFR, glucocorticoids and other well-known confounders. There were no similar associations for PTH, phosphate or calcium. In line with previous observations [1–4], older age, non-Caucasian ethnicity and high glucocorticoid doses were also independently associated with PHYG.

Post-transplant hyperglycaemia probably develops as a combined result of pre-existing and transplant-related factors. A pre-existing disposition for hyperglycaemia is not necessarily evident before surgery, since insulin clearance is impaired and the requirement for insulin accordingly reduced in CKD. Mild cases of pre-existing hyperglycaemia can remain undetected until insulin requirements rebound post-transplant, preventing predisposed individuals from being identified ahead of transplant unless specifically tested. Impairments in pre-transplant insulin sensitivity [21], and also insulin secretion [22,23], have been reported in patients who later developed PHYG. Our findings fall in line with these reports but, to the best of our knowledge, have not been reported in a Caucasian RTx population of the current size. The results extend previous findings by indicating that PHYG may be more effectively predicted through pre-transplant evaluation of 2h-PG rather than FPG. This supports the use of the OGTT in the pre-transplant setting, as is increasingly advocated [24]. It remains to be studied what discrete levels of pre-transplant glycaemia predict PHYG and whether PHYG can be prevented by interventions among patients awaiting transplantation.

Despite their steroid use and other transplant-related factors, NGM patients displayed a reduction in FPG and 2h-PG from pre- to post-transplant measurements. This may have been related to the lowering of BMI, although the reduction in BMI did not correlate with reductions in FPG [Rho 0.085 (NGM) and −0.142 (PHYG)] or 2h-PG [0.065 (NGM) and 0.008 (PHYG); all P > 0.235]. Whether RTx can improve glucose metabolism in selected patients, possibly by attenuating uraemia, is an intriguing question that demands further studies.

Renal transplantation alleviates the accumulation of urea by improving renal elimination. At the same time, transplantation is accompanied by processes that promote urea appearance. The observed results suggest that PHYG was

Table 6. Multiple regression vs. post-transplant hyperglycaemia

	OR[a]	(95% CI)[a]	P[b]
Age (tertiles)	2.40	(1.02, 5.65)	0.047
Male gender (yes)	0.69	(0.35, 1.37)	0.536
Non-Caucasian (yes)	4.44	(1.08, 18.3)	0.021
BMI (tertiles)	1.31	(0.58, 2.95)	0.419
Previous transplants (yes)	2.42	(1.04, 5.64)	0.030
Time on dialysis (tertiles)[c]	1.88	(0.77, 4.56)	0.169
Living donor (yes)	0.91	(0.45, 1.83)	0.999
≥3 HLA-1 mismatches (yes)	2.24	(1.17, 4.28)	0.031
Use of furosemide (yes)	1.42	(0.70, 2.88)	0.318
Use of beta blocker (yes)	1.72	(0.89, 3.30)	0.130
Prednisolone (tertiles)	2.65	(1.16, 6.05)	0.009
Urea (mmol/L)	1.14	(1.02, 1.27)	0.014
GFR (tertiles)	3.07	(1.20, 7.85)	0.012
iCa (tertiles)	1.43	(0.66, 3.10)	0.391
Phosphate (tertiles)	1.56	(0.71, 3.40)	0.278
PTH (tertiles)	0.73	(0.34, 1.59)	0.464
Pre-transplant 2h-PG (mmol/L)	1.26	(1.09, 1.46)	0.001

[a]Categorized continuous variables: OR for the highest vs. lowest tertile (reference).
[b]Linear trend across categories.
[c]Pre-emptive transplantation is reference tertile.

indirectly related to mechanisms either impairing urea elimination or promoting urea appearance or possibly directly related to the actual plasma urea levels. In support of mechanisms not related to urea elimination, PHYG was associated with increments in urea across each category of GFR.

Endogenous protein catabolism is the main contributor to the appearance of urea and is also associated with insulin resistance in CKD and haemodialysis patients [25, 26]. On the one hand, transplant-related catabolic processes, promoted by surgery, glucocorticoids and chronic inflammation, may thus lead to PHYG by compromising insulin sensitivity. We adjusted for glucocorticoids but had no specific data to reflect protein turnover or inflammation. On the other hand, protein breakdown may also be secondary to insulin resistance, rather than strictly vice versa [26], and we observed a crude association between pre-transplant FPG and post-transplant urea levels. Markers of insulin sensitivity, such as the homeostasis model assessment (HOMA) index, may increase our understanding of the interplay between glucose and protein metabolism and should be considered in future studies on this topic in transplant patients. Gastrointestinal bleeding increases urea levels but is unlikely to have contributed significantly to PHYG in our study, since Hb was similar in NGM and PHYG patients.

There are several indications that urea may interfere directly with glucose metabolism. First, dialysis temporarily normalizes glucose tolerance, suggesting that uraemic hyperglycaemia involves a dialyzable solute [27–30]. Second, urea levels have been found to correlate with insulin resistance in CKD patients [31] and are lowered by low-protein diets, which have insulin-sensitizing effects in terminal uraemia [32]. Third, oral urea loading has been reported to reduce glucose disposal in healthy males [33] and stable CKD patients [34]. In apparent contrast, dialysis against a high urea gradient does not seem to yield any evident glycaemic changes [28,35]. It is conceivable, however, that glycaemic effects of high urea levels were diluted by dialysis itself in these reports, whereby the removal of alternative diabetogenic solutes was comparatively unaffected. Such solutes could include dialyzable by-products of urea generated through carbamylation, such as carbamylated amino acids [36].

Plasma calcium, phosphate and PTH were not associated with PHYG in our study. Even if they correlate with insulin metabolism in the absence of renal failure [37–41], the role of calcium and phosphate may be confounded by internal phosphate redistribution [42] or persistently altered vitamin D or fibroblast growth factor 23 levels after RTx [43]. Parathyroidectomy has led to increased [13], unchanged [12] or even reduced [29] insulin secretion, and the insignificant role of PTH in our study falls in line with these inconsistencies.

The present study has certain limitations. First, since regression models are sensitive to correlations between explanatory variables, results relating urea to PHYG may have been affected by residual confounding due to GFR. Nonetheless, we considered it mandatory to adjust for GFR due to its key role as confounder. Univariable and confirmatory regressions yielded urea a significant contributor, regardless of whether GFR was entered into the model or not (not shown), and the PHYG risk associated with increments in urea was significantly higher than null in each category of GFR. Second, missing data were present for 28% of patients. However, MI serves to maintain power and lower the risk of bias and is the preferred method of handling missing data when their prevalence exceeds 10% [44]. Our MI results retained all significant contributors from CC analysis and also revealed associations with PHYG for age and ethnicity, two well-established risk factors. By rendering these important factors insignificant, CC results are likely to have been biased as compared to our MI results. Third, information on diet or family history of DM was unavailable. However, high protein intake can either improve or worsen glycaemia, respectively, depending on whether renal function is normal [45] or terminally impaired [32]. The role of a family history of DM can also be questioned, due to information bias [46]. Finally, PTH, phosphate and calcium levels do not stabilize as swiftly as does the renal function post-transplant, and interpretation of their plasma levels requires caution.

In conclusion, elevated pre-transplant glycaemia and post-transplant urea levels were independently associated with PHYG in the early post-transplant period. These results were independent of GFR, PTH, phosphate and calcium. It remains to be determined which cut-off points of pre-transplant glycaemia best describe the risk of PHYG and whether PHYG is associated with the elimination, appearance or actual plasma levels of urea.

Acknowledgements. This project was financed by grants from the Norwegian Foundation for Health and Rehabilitation.

Conflict of interest statement. The results presented in this paper have not been published previously in whole or part, except in abstract format.

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Received for publication: 8.7.09; Accepted in revised form: 28.9.09