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Cross-Sectional Assessment of the Effect of Kidney and Kidney-Pancreas Transplantation on Resting Left Ventricular Energy Metabolism in Type 1 Diabetic-Uremic Patients

A Phosphorous-31 Magnetic Resonance Spectroscopy Study

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OBJECTIVES To test whether left ventricular (LV) dysfunction affecting type 1 diabetic-uremic patients was associated with abnormal heart high-energy phosphates (HEPs) and to ascertain whether these alterations were also present in recipients of kidney or kidney-pancreas transplantation.

BACKGROUND Heart failure is the major determinant of mortality in patients with diabetic uremia. Both uremia and diabetes induce alterations of cardiac HEPs metabolism.

METHODS Magnetic resonance imaging and phosphorous magnetic resonance spectroscopy of the LV were performed in the resting state by means of a 1.5-T clinical scanner. Eleven diabetic-uremic patients, 5 nondiabetic patients with uremia, 11 diabetic recipients of kidney transplantation, and 16 diabetic recipients of combined kidney-pancreas transplantation were studied in a cross-sectional fashion. Eleven nondiabetic recipients of kidney-only transplant and 13 healthy subjects served as control groups.

RESULTS Uremic patients had higher LV mass, diastolic dysfunction, and lower phosphocreatine (PCr)/adenosine triphosphate (ATP) ratio in comparison with recipients of kidney-pancreas or nondiabetic recipients of kidney transplant. In diabetic recipients of kidney transplant the PCr/ATP ratio was higher than in uremic patients but was lower than in the controls. Recipients of combined kidney-pancreas transplant had a higher ratio than uremic patients but no difference was found in comparison with controls.

CONCLUSIONS Altered resting myocardial HEPs metabolism may contribute to LV dysfunction in diabetic-uremic patients. In diabetic recipients of kidney transplantation, a certain degree of LV metabolic and functional impairment was found. In combined kidney-pancreas recipients the resting LV metabolism and function were not different than in controls. (J Am Coll Cardiol 2005;46:1085–92) © 2005 by the American College of Cardiology Foundation

More than a century ago heart failure was regarded as a "frequent and noteworthy complication of diabetes mellitus" (1). Epidemiologic evidence sustaining this observation in patients with type 1 diabetes became available in the 1970s (2). It is now recognized that heart failure is the major cause of death (~65%) among patients with type 1 diabetes and this risk is high in those with concomitant uremia (3). Mechanisms for myocardial dysfunction include coronary macro- and microvascular angiopathy and hypertension (4). However, a specific cardiomyopathy was suggested as a causal factor producing high cardiac mortality and morbidity independent of the aforementioned conditions (5,6), and it was hypothesized that lipo- and gluco-toxicity may induce altered cardiac function in patients with diabetes (7).

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Localized phosphorous-31 magnetic resonance spectroscopy (^{31}P-MRS) has been used to study the left ventricular (LV) high-energy phosphates (HEPs) in vivo in humans (8,9): The derived phosphocreatine (PCr)/adenosine triphosphate (ATP) ratio is considered the index of energy metabolism and the phosphate potential (energy charge) of the myocardium (10).

We hypothesized that the alterations of cardiac function in patients with type 1 diabetes and uremia might be due to impaired cardiac energy metabolism and that the cure of chronic hyperglycemia and uremia might induce improvements of these abnormalities. This study was undertaken: 1) to test whether the abnormal LV diastolic function of type 1 diabetic-uremic patients was associated with abnormal PCr/ATP ratio, and 2) to ascertain the effects of kidney transplantation alone and/or combined kidney-pancreas transplantation on heart function and metabolism using noninvasive cardiac magnetic resonance imaging (MRI) and ^{31}P-MRS.

METHODS

Subjects. **STUDY SUBJECTS.** Study subjects were recruited within the Section of Organ Transplantation of the Istituto

Abbreviations and Acronyms

E/A ratio = E-peak filling rate/A-peak filling rate ratio
ECG = electrocardiogram/electrocardiographic
HEPs = high-energy phosphates
LV = left ventricle
MRI = magnetic resonance imaging
NYHA = New York Heart Association
PCr/ATP = phosphocreatine/adenosine triphosphate
^{31}P-MRS = phosphorous-31 magnetic resonance spectra/spectroscopy
rCRSD = relative Cramer-Rao standard deviation

Scientifico H San Raffaele. All patients were in stable clinical and nutritional conditions. The main criteria for exclusion from the study were: 1) history of coronary, cerebral, or peripheral vascular events; 2) history of dilated cardiomyopathy; 3) previous knowledge of a pathologic ejection fraction; 4) previous knowledge of resting electrocardiographic (ECG) markers of cardiac ischemia; and 5) features compatible with the New York Heart Association (NYHA) functional classes for heart failure. To assess the combined effects of type 1 diabetes and kidney failure on cardiac HEPs metabolism and the effect of kidney failure alone, 11 type 1 diabetic-uremic patients and 5 nondiabetic uremic patients were selected. They were on the waiting list of our institute for receiving combined kidney-pancreas transplantation (diabetic patients) or kidney transplantation alone (nondiabetic patients). Eleven diabetic recipients of kidney transplantation only (owing to macroscopic damage of the pancreas at the time of organ harvest) and 16 diabetic recipients of combined kidney-pancreas transplantation also were studied to assess cardiac HEPs metabolism in transplanted patients.

CONTROL SUBJECTS. To control the effects of immunosuppression and of diabetes, an additional group of 10 nondiabetic patients with uremia were studied after receiving kidney transplantation alone and 13 healthy individuals matched for age and body mass index with the other study groups served as controls without history of diabetes and uremia. Finally, to compare the severity of the LV abnormalities detected in our study groups with that of a group of individuals with overt systolic dysfunction, 11 individuals with known heart failure determined by means of standard echocardiography were recruited. Characteristics of study groups are summarized in Table 1. In all transplanted patients, immunosuppression was maintained using cyclosporine, mycophenolate mofetil, or azathioprine. Recipients of combined kidney-pancreas transplantation were insulin independent, whereas diabetic-uremic patients and diabetic recipients of kidney transplantation were on subcutaneous insulin therapy. Blood pressure was taken twice in the sitting position and after 10 min rest with a sphyngomanometer. Informed consent was obtained from all subjects after explanation of purposes, nature, and potential risks of the study. The protocol was approved by the ethical committee of the Istituto Scientifico H San Raffaele.

Experimental protocol. Patients were instructed to consume an isocaloric diet and to abstain from exercise activity for two weeks before the study. Patients took all of their medications and subcutaneous insulin according to the regular schedule. All patients underwent the ^{31}P-MRS

Table 1. Anthropometric and Laboratory Parameters of Study Groups

	Diabetic-Uremic Patients	Nondiabetic Uremic Patients	Diabetic Recipients of Kidney Transplantation	Recipients of Combined Kidney-Pancreas Transplantation	Nondiabetic Recipients of Kidney Transplantation	Normal
Number of patients (F/M)	11 (3/8)	5 (0/5)	11 (3/8)	16 (6/10)	11 (3/8)	13 (2/11)
Age (yrs)	44 ± 2	38 ± 5	48 ± 3	39 ± 2	46 ± 4	42 ± 5
Body mass index (kg/m^2)	23.3 ± 1.2	21.2 ± 1.6	23.9 ± 0.8	22.4 ± 0.7	22.4 ± 0.7	23.8 ± 0.5
Surface area (m^2)	1.75 ± 0.26	1.75 ± 0.05	1.76 ± 0.19	1.79 ± 0.21	1.72 ± 0.08	1.89 ± 0.22
Duration of dialysis (yrs)	2.2 ± 0.6	2.1 ± 0.9	3.2 ± 0.9	2.9 ± 0.5	1.3 ± 0.3	—
Transplant age (yrs)	—	—	3.2 ± 1.2	3.5 ± 0.7	4.0 ± 0.6	—
Duration of diabetes (yrs)	30 ± 3†	—	31 ± 2	25 ± 2	—	—
Glycated hemoglobin (%)	9.0 ± 0.6*†‡‖	4.1 ± 0.3	8.6 ± 0.5*†‡‖	5.6 ± 0.2	5.6 ± 0.2	5.0 ± 0.3
Creatinine (mg/dl)	7.2 ± 0.5*†§‖	6.3 ± 0.9*†§‖	1.0 ± 0.2	1.4 ± 0.2‖	1.5 ± 0.2‖	0.9 ± 0.2
Microalbuminuria	nd	nd	18 ± 7	15 ± 5	15 ± 2	nd
Total cholesterol (mg/dl)	206 ± 15	176 ± 28	206 ± 15	177 ± 9	172 ± 12	181 ± 4
Triglycerides (mg/dl)	121 ± 22	187 ± 22	141 ± 18	92 ± 7†‡§	211 ± 48	83 ± 5
Systolic blood pressure (mm Hg)	133 ± 6	140 ± 12	141 ± 6	128 ± 5	133 ± 3	125 ± 3
Diastolic blood pressure (mm Hg)	76 ± 7	85 ± 6	83 ± 2	79 ± 2§	81 ± 2	81 ± 2
Patients on antihypertensive drugs (%)	65%	100%	91%	73%	82%	0%
Retinopathy (%)	100%	—	100%	100%	—	—
Neuropathy (%)	100%	—	100%	100%	—	—

One-way analysis of variance and Tukey's post-hoc test. *$p < 0.01$ vs. recipients of combined kidney-pancreas transplantation. †$p < 0.01$ vs. non-diabetic recipients of kidney transplantation. ‡$p < 0.05$ vs. non-diabetic uremic patients. §$p < 0.05$ vs. diabetic recipients of kidney transplantation. ‖$p < 0.05$ vs. normals.

protocol at 4:00 PM on Thursdays. Seven diabetic-uremic patients, four nondiabetic uremic patients, seven diabetic recipients of kidney transplantation, nine recipients of combined kidney-pancreas transplantation, and nine nondiabetic uremic recipients of kidney-only transplantation performed the MRI protocol in the same session or the following Thursday. Uremic patients performed the studies the day after the dialysis session.

^{31}P-MRS PROTOCOL. Cardiac ^{31}P-MRS and MRI were performed at rest with the use of a 1.5-T whole-body scanner (Gyroscan Intera Master 1.5 MR System; Philips Medical Systems, Best, the Netherlands). ^{31}P spectra were obtained by means of a 10-cm-diameter surface coil used for transmission and detection of radio frequency signals at the resonance frequency of ^{31}P (at 1.5 T, 25.85 MHz) as described by Lamb et al. (11). The surface coil was secured in place with a Velcro band around the chest. The ECG-triggered MRI was performed to acquire scout images, to establish the exact position of the ^{31}P surface coil, and eventually to reposition the coil center just below the mitral valve level of the heart. Localized homogeneity adjustment was performed using the body coil and ECG-triggering by optimizing the ^{1}H-MRS water signal. Shim volumes were planned on the transverse and sagittal scout images to include the entire LV. The transmitter-receiver was then switched without time delay to the ^{31}P frequency. Manual tuning and matching of the ^{31}P surface coil was performed to adjust for different coil loading. The radio frequency level was adjusted to obtain a 180° pulse of 40 μs for the reference sample at the center of the ^{31}P surface coil. The acquisition of ^{31}P-MR spectra was triggered to the R-wave of the ECG, with a trigger delay time of 200 ms and a recycle time of 3.6 s. The image-selective in vivo spectroscopy (ISIS) volume selection in three dimensions (3D-ISIS) based on 192 averaged free induction decays was employed. The volume of interest was oriented avoiding inclusion of chest wall muscle and diaphragm muscle. The volume size was typically 6 (caudo-cranial) $\times$ 7 $\times$ 7 cm^3. Acquisition time was 10 min. Adiabatic frequency-modulated hyperbolic secant pulses and adiabatic half-passage detection pulses were used to achieve inversion and excitation over the entire volume of interest. Examination time was 40 to 45 min. Three-dimensional ISIS was employed after testing that the PCr/ATP ratios were in close agreement using higher spatial resolution (two-dimensional ISIS + one-dimensional spectroscopic imaging using a one-dimensional phase encoding bar with 32 rows of 1 cm thickness).

MRI PROTOCOL. MRI studies were performed with the scanner just described using an enhanced gradient system with a maximum gradient strength of 30 mT/m and a maximum gradient slew rate of 150 mT $\cdot$ m^{-1} $\cdot$ s^{-1}. The Cardiac Research software patch (operating system 9) was used. The examination was performed using a five-element cardiac phased-array coil (SENSE-cardiac) and retrospective ECG-triggering obtained with the vectorcardiogram system (12) using standard MRI methodology. Briefly, cine long-axis, four-chamber, and two-chamber views were obtained using balanced fast field echo breath-hold sequence (bFFE/BH), and a volumetric evaluation in a three-dimensional fashion was obtained (13). Flow mapping of the transmitral flow was performed with an ECG-gated, quantitative flow measurement, two-dimensional, phase-contrast, fast field echo sequence oriented perpendicular to the transmitral flow, and parameters of diastolic function were calculated (14). The entire MRI protocol lasted 30 min.

Analytical high-density lipoprotein cholesterol, and triglycerides were measured as previously described (15). Low-density lipoprotein cholesterol was calculated using the Friedwald formula (16).

Calculation. ^{31}P-MRS ANALYSIS. The ^{31}P-MRS were transferred to a remote SUN-SPARC workstation (SUN Microsystems Inc., Santa Clara, California) for analysis. The spectra (Fig. 1) were quantified automatically in the time domain, using Fitmasters. The ATP level was corrected for the ATP contribution from blood in the cardiac chambers based on a previous study (17). Depending on the repetition time (TR), PCr/ATP ratios had to be corrected for partial saturation effects and T1 values obtained from inversion recovery experiments on the human LV were used. Based on these data and a repetition time of 3.6 s, a saturation correction factor of 1.35 was obtained and applied to all "blood corrected" myocardial PCr/ATP ratios (11,18). An estimate of the signal-to-noise-ratio of each spectrum was obtained from the relative Cramer-Rao standard deviation (rCRSD) calculated for the PCr/ATP, which is a commonly reported index of accuracy of the spectral quantification (11). The ^{31}P-MRS with an rCRSD $>$20% were excluded and the experiment was repeated to obtain a better spectrum.

MRI ANALYSIS. Image analysis was performed by an image-processing workstation (EasyVision; Philips Medical Systems) using the cardiac analysis software package. The endocardial contours of the LV were traced manually by an experienced cardioradiologist on all cardiac phases of the 10 short-axis orientation slices, and epicardial contours were traced on telediastolic phases. The surface areas of the endocardial tracings in end-diastole and end-systole were automatically summed up and multiplied by parameters relative to the section thickness to produce the end-diastolic and end-systolic volumes. From this analysis the stroke volume, cardiac output, and ejection fraction were automatically measured (Table 2). The LV mass was calculated multiplying the wall volume by the specific density of cardiac muscle (1.05 g/cm^3) (19). For the assessment of the diastolic function the mitral orifice area was manually traced on the magnitude images and automatically transferred to the velocity phase images by A. E. and F. D. C. The morphology and the size of each contour were adjusted according to the cardiac phase. The mean flow (ml/s) was

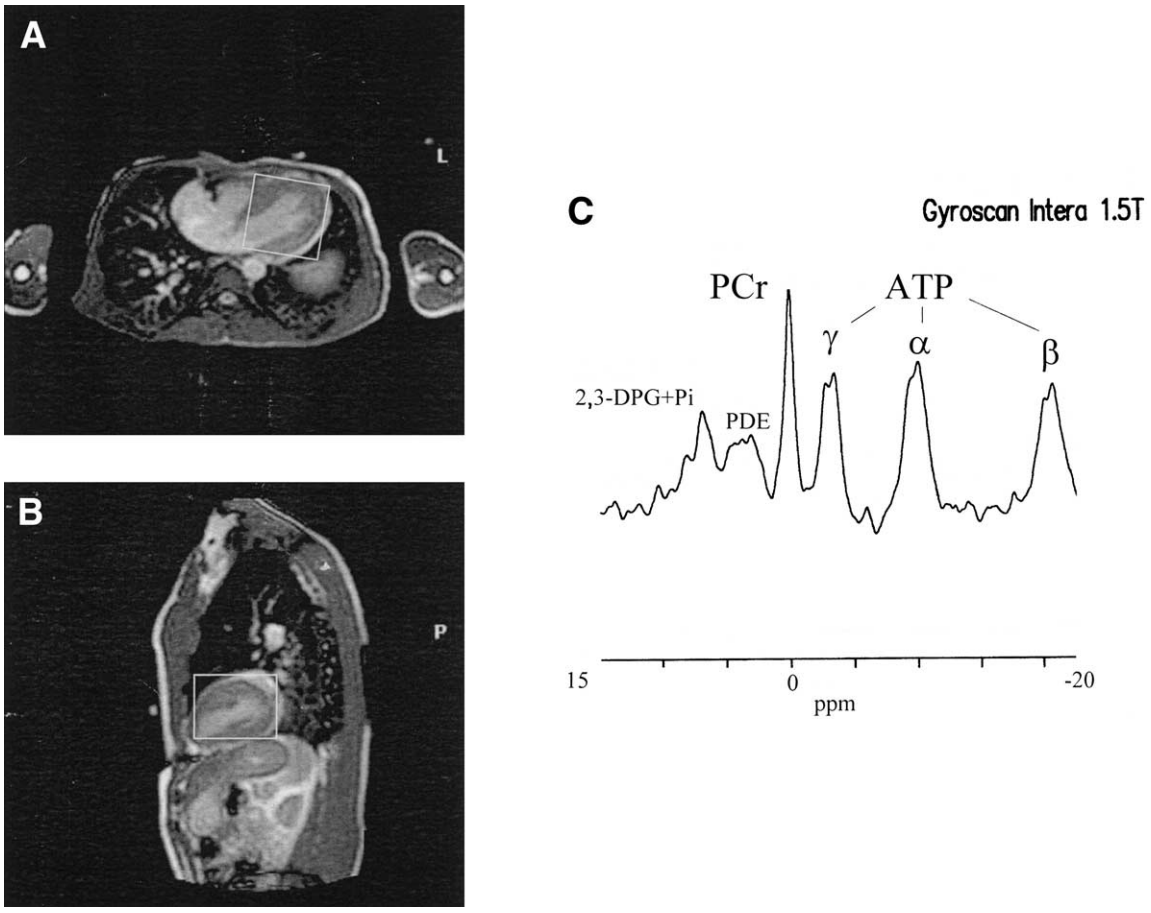

Figure 1. Scout images in the transverse **(A)** and sagittal **(B)** image planes and cardiac ^{31}P-spectrum **(panel C)**. In **panels A and B**, the squares indicate the three-dimensional ISIS volume. In **panel C**, the ^{31}P-spectrum shows resonance peaks of phosphocreatine (PCr), adenosine triphosphate (ATP), phosphodiesters (PDE), and the combined signals of 2,3-diphosphoglycerate (2,3-DPG) originating from blood and inorganic phosphate (Pi).

determined for each cardiac phase and subsequently automatically, curves of flow rate versus time were reconstructed. From this analysis we measured the E and A peaks features and volumes (Table 3).

Statistical analysis. All data are presented as mean ± SEM. Analyses were performed using SSPS software (version 10.0; SPSS, Chicago, Illinois). Comparisons between groups were performed using one-way analysis of variance and Tukey's post hoc tests. Linear regression analysis was used to determine the association between PCr/ATP ratio and uremia, diabetes, and the LV mass index separately. Stepwise multiple regression analysis was performed (using F ratio-to-remove of 4 and F ratio-to-enter of 3.996) to assess which of these variables were more relevant. Whenever it is stated that groups were not different for a specific variable, one should bear in mind that with the present

Table 2. Left Ventricular Morphology and Systolic Function

	Diabetic-Uremic Patients	Nondiabetic Uremic Patients	Diabetic Recipients of Kidney Transplantation	Recipients of Combined Kidney-Pancreas Transplantation	Nondiabetic Recipients of Kidney Transplantation	Normal
Number of patients (F/M)	7 (1/6)	4 (0/4)	7 (1/5)	9 (1/8)	9 (1/8)	13 (2/11)
LV mass (g)	180 ± 25*‡	185 ± 24*†	148 ± 23	121 ± 19	140 ± 7	127 ± 24
LVMI (g/m^2)	91 ± 22	106 ± 28*‡	84 ± 18	68 ± 14	83 ± 11	67 ± 8
End-diastolic volume (ml)	131 ± 14*‡	140 ± 26*†	113 ± 22	100 ± 13‡	109 ± 5‡	135 ± 23
End-systolic volume (ml)	53 ± 6*‡	48 ± 12*	31 ± 13	29 ± 3‡	34 ± 2‡	49 ± 12
Stroke volume (ml)	79 ± 8	92 ± 18	74 ± 11	72 ± 10	73 ± 7	86 ± 15
Ejection fraction (%)	60 ± 1	69 ± 5	67 ± 3	71 ± 2	67 ± 2	63 ± 5
Cardiac output (l/min)	6.1 ± 0.6	6.1 ± 1.3	5.4 ± 0.7	4.5 ± 0.3	5.2 ± 0.5	6.0 ± 1.7
Heart rate (beats/min)	78 ± 3	65 ± 5	75 ± 7	64 ± 5	72 ± 3	69 ± 12

One-way analysis of variance and Tukey's post-hoc test. *$p < 0.01$ vs. recipients of combined kidney-pancreas transplantation. †$p < 0.01$ vs. non-diabetic recipients of kidney transplantation. ‡$p < 0.05$ vs. normals.
LV = left ventricular; LVMI = left ventricular mass index.

Table 3. Left Ventricular Diastolic Function

	Diabetic-Uremic Patients	Nondiabetic Uremic Patients	Diabetic Recipients of Kidney Transplantation	Recipients of Combined Kidney-Pancreas Transplantation	Nondiabetic Recipients of Kidney Transplantation	Normal
Number of patients (F/M)	7 (1/6)	4 (0/4)	7 (1/5)	9 (1/8)	9 (1/8)	13 (2/11)
E/A peak flow	1.4 ± 0.3§	1.4 ± 0.1§	1.2 ± 0.2§	1.6 ± 0.1	1.6 ± 0.3	2.0 ± 0.5
E-peak filling rate (ml/s)	422 ± 22*†‡	456 ± 39*†‡	338 ± 23§	373 ± 25§	367 ± 30§	482 ± 103
E-peak filling rate/EDV (s^{-1})	3.3 ± 0.5	3.5 ± 0.3	3.3 ± 0.4	3.8 ± 0.3	3.4 ± 0.4	3.7 ± 0.6
E acceleration peak ($ml/s^2 \times 10^{-3}$)	6.5 ± 0.5	5.8 ± 1.9	5.9 ± 0.6	5.7 ± 0.6	5.0 ± 0.6	6.3 ± 1.6
E deceleration peak ($ml/s^2 \times 10^{-3}$)	−3.1 ± 0.4	−3.2 ± 0.9	−2.2 ± 0.3§	−2.8 ± 0.2§	−2.7 ± 0.4§	−3.9 ± 1.1
A-peak filling rate (ml/s)	364 ± 80*§	304 ± 29*§	331 ± 56	236 ± 28	264 ± 31	237 ± 78
A-peak filling rate/EDV (s^{-1})	2.7 ± 0.3§	2.4 ± 0.4§	3.0 ± 0.3*§	2.4 ± 0.1§	2.4 ± 0.3§	1.8 ± 0.5
A acceleration peak ($ml/s^2 \times 10^{-3}$)	3.9 ± 0.8	5.1 ± 0.4	4.7 ± 0.7	4.0 ± 0.5	3.9 ± 0.4	4.8 ± 2.4
A deceleration peak ($ml/s^2 \times 10^{-3}$)	−5.0 ± 1.0*§	−4.6 ± 0.9	−4.9 ± 1.1*§	−3.3 ± 0.4	−3.3 ± 0.7	−3.3 ± 1.4
Deceleration time (ms)	95 ± 24*†‡§	163 ± 15	134 ± 19	157 ± 25	163 ± 10	165 ± 26

One-way analysis of variance and Tukey's post-hoc test. *$p < 0.01$ vs. recipients of combined kidney-pancreas transplantation. †$p < 0.01$ vs. non-diabetic recipients of kidney transplantation. ‡$p < 0.05$ vs. diabetic recipients of kidney transplantation. §$p < 0.05$ vs. normals.

sample size, the power to detect clinically meaningful differences might not be sufficient.

RESULTS

Patient characteristics (Table 1). Anthropometric features of study subjects were not different. Dialysis and diabetes durations were not different among the appropriate study groups. Patients with diabetes were in poor control as reflected by the HbA1c level (Table 1), and patients with uremia were characterized by high plasma creatinine. Microalbuminuria and the lipid profile were not different among groups with the exception of plasma triglyceride which was higher in nondiabetic uremic patients and diabetic and nondiabetic recipients of kidney transplantation in comparison with recipients of combined kidney-pancreas transplantations and normal subjects. In contrast with normal subjects, the majority of patients were taking antihypertensive drugs, but blood pressure was not different among groups. The 11 individuals with known heart failure (1 female:10 males; age 66 ± 6 yrs; body mass index 27.1 ± 3.4 kg/m^2; 6 in NYHA functional class I, 3 in class IIA, and 2 in class IIB) had ischemic cardiomyopathy (n = 8) or dilated cardiomyopathy (n = 3) and had an ejection fraction of 35 ± 2%, based on standard echocardiography.

Anatomical and functional assessment of the left ventricle (Tables 2 and 3). The LV mass was higher in uremic patients (Table 2). Larger end-diastolic and end-systolic volumes were also detected in diabetic-uremic patients in comparison with recipients of combined kidney-pancreas transplantation and nondiabetic recipients of kidney-only transplantation. Parameters of systolic function were not different among groups (Table 2). In contrast, parameters of diastolic function were altered in diabetic and nondiabetic uremic patients as reflected by the lower E/A ratio when compared with normal subjects (Table 3). Interestingly, diabetic recipients of kidney transplantation also showed abnormalities of the diastolic function in comparison with normal subjects (Table 3). Dialysis may induce loading-dependent modifications of the indices of the LV function in patients with chronic renal failure. To minimize this issue the uremic patients were studied the day after dialysis. It is important to state that it is reported that after hemodialysis cine-MRI did not show a significant change in the ejection fraction but detected 14% to 27% reduction of the end-diastolic, end-systolic, and stroke volumes; meanwhile LV mass was reduced by 4% (20); in addition, hemodialysis was reported to induce a reduction in the E/A ratio and no change in the deceleration time (21).

PCr/ATP ratio: accuracy and reproducibility. Mean rCRSD was not different among groups (15 ± 2%, 11 ± 2%, 14 ± 2%, 15 ± 1%, 14 ± 1%, and 14 ± 3% in diabetic and nondiabetic uremic patients, diabetic recipients of kidney transplantation, recipients of combined kidney-pancreas transplantation, nondiabetic recipients of kidney transplantation, and normal subjects, respectively). An estimate of intra-examination differences was obtained by studying 12 subjects twice and consecutively in the same session without changing the position of the surface coil, the sensitive volumes, and the acquisition parameters. The coefficient of variation was 4 ± 2%. Interexamination variability was studied by examining 8 subjects on two separate occasions, with a time interval of 7 to 16 days. No efforts were made to minimize variability. The coefficient of variation was 8 ± 3%.

Metabolic assessment of the HEPs of the left ventricle (Fig. 2). The PCr/ATP ratio was not different in diabetic uremic (1.33 ± 0.05) in comparison with nondiabetic uremic (1.42 ± 0.07; $p = 0.34$) patients but was lower in both subgroups in comparison with nondiabetic recipients of kidney transplantation (1.75 ± 0.07; $p < 0.0001$) and

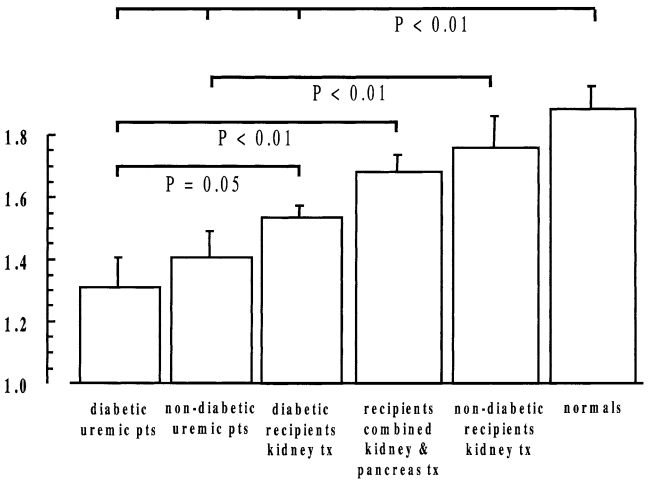

Figure 2. Comparison of the left ventricular (LV) phosphocreatine/ATP (PCr/ATP) ratios of diabetic patients with uremia, nondiabetic patients with uremia, diabetic-uremic recipients of kidney transplantation (Tx) alone or combined kidney-pancreas transplantation, nondiabetic uremic recipients of kidney transplantation alone, and normal subjects. One-way analysis of variance and Tukey's post-hoc tests.

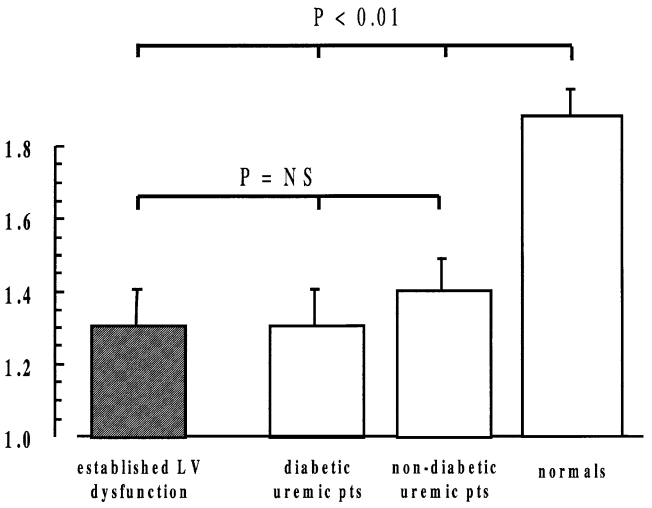

Figure 3. Comparison of the phosphocreatine/ATP (PCr/ATP) ratios of the left ventricle of individuals with established left ventricular (LV) systolic dysfunction (with ejection fraction 35% at echocardiography), diabetic patients with uremia, nondiabetic patients with uremia, and normal subjects. One-way analysis of variance and Tukey's post-hoc tests.

normal subjects (1.91 ± 0.05; $p < 0.01$). In diabetic recipients of kidney transplantation the PCr/ATP ratio was higher (1.55 ± 0.07) in comparison with uremic patients ($p = 0.03$) but was lower in comparison to the controls ($p < 0.05$). In contrast, recipients of combined kidney-pancreas transplantation had a higher ratio (1.68 ± 0.11) in comparison with the uremic patients ($p = 0.02$) and no difference was detected in comparison with control subjects (p = NS).

Linear regression analysis (Table 4). Linear regression analysis showed that when taken separately the variables uremia, diabetes, and LV mass index were significantly associated with the PCr/ATP ratio. The multiple stepwise regression analysis selected uremia as the best predictor of the PCr/ATP ratio; meanwhile, diabetes ($p = 0.057$), and LV mass index ($p = 0.258$) did not add a significant predictive value.

Severity of the HEPs abnormalities (Fig. 3). MRI confirmed that patients with overt systolic dysfunction had reduced ejection fraction (38 ± 5%) and stroke volume (61 ± 13 ml; $p < 0.01$ vs. normal subjects) along with increased LV mass (194 ± 25 g; $p < 0.01$ vs. normal subjects) and diastolic dysfunction (E/A ratio 0.8 ± 0.3; $p < 0.01$ vs. normal subjects). The PCr/ATP ratio was significantly lower when compared with normal subjects (1.36 ± 0.12 vs. 1.91 ± 0.05; $p < 0.01$), and the reduction was proportional to the NYHA functional class of the patients (1.50 ± 0.20 in class I, 1.31 ± 0.12 in class IIA, and 1.0 ± 0.15 in class IIB; $r = -0.51$; $p < 0.05$). The PCr/ATP ratio of diabetic and nondiabetic uremic patients (1.33 ± 0.05 and 1.42 ± 0.07, respectively; p = NS) was not different in comparison with the individuals with impaired systolic dysfunction.

Table 4. Results of the Linear Regression Analysis Between the PCr/ATP Ratio and Uremia, Diabetes, and LVMI

	Variable	r	p Value
Linear regression analysis	Uremia	0.40	0.001
	Diabetes	0.37	0.003
	LVMI	0.36	0.024
Stepwise multiple regression analysis			
Entered variables	Uremia	0.42	0.008
Excluded variables	Diabetes	—	0.057
	LVMI	—	0.258

LVMI = left ventricular mass index.

DISCUSSION

This work demonstrates that the left ventricle functional abnormalities affecting patients with type 1 diabetes and uremia are associated with reduced myocardial PCr/ATP ratio. In diabetic recipients of kidney-only transplantation, abnormal parameters of diastolic function were found, and the PCr/ATP ratio, though higher in comparison with uremic patients, was still lower in comparison with the control groups. In contrast, in the recipients of combined kidney-pancreas transplantation the PCr/ATP ratio was not different in comparison with the control groups.

LV function and HEPs metabolism in type 1 diabetes and uremia. Our type 1 diabetic-uremic patients had left ventricular hypertrophy and diastolic dysfunction (Tables 2 and 3) as previously reported (22,23). Less severe abnormalities also were detected in the nondiabetic uremic patients, suggesting that uremia per se is associated with the derangement of the left ventricular function and that diabetes may worsen the heart function. The key finding of the present work was the discovery of an alteration of the HEPs content in both diabetic and nondiabetic uremic patients as assessed by means of ^{31}P-MRS. Similar results were reported in patients with hypertension and left ventricular hypertrophy; reduced creatine kinase activity and a lower total creatine content may sustain these metabolic alterations (24), and the reduced PCr/ATP ratio was associated

with the progression of heart failure in patients with dilated and hypertrophic cardiomyopathy (25). In this study we included a group of patients with overt impairment of the ejection fraction and ascertained that the severity of the abnormal HEPs observed in the uremic patients was severe despite the fact that the systolic function was unaffected (Fig. 3).

LV function and HEPs metabolism in transplant recipients. Diabetic recipients of kidney-only transplantation, despite the better cardiac morphologic, functional, and whole-body metabolic features, showed reduced PCr/ATP ratio when compared with nondiabetic recipients of kidney-only transplantation and normal subjects, suggesting that diabetes was associated with the presence of abnormal intracardiac HEPs. Combined kidney-pancreas recipients, in which the renal failure and diabetes were simultaneously cured, showed a better left ventricular function in comparison with the diabetic-uremic patients, and, overall, the PCr/ATP ratio was not different in comparison with the control groups.

These results may provide important insights. First, the abnormalities characterizing the uremic state may be due to uremia per se, and chronic hyperglycemia may be an independent aggravating factor regardless of hypertension. In fact, the better functional and metabolic parameters characterizing combined kidney-pancreas recipients were found despite the fact that in these patients hypertension was also present (Table 1) and an antihypertensive drug regimen was required as in the diabetic and nondiabetic uremic patients and despite the fact that blood pressure was not different among groups. Second, despite better morphologic, functional, and metabolic parameters in comparison with uremic patients, diabetic recipients of kidney-only transplantation showed persisting abnormal features, supporting a deleterious effect of diabetes per se (Tables 2 and 3, Fig. 2). Third, in combined kidney-pancreas recipients, the restoration of a normal metabolic milieu was successful in achieving normal resting left ventricular function and metabolism despite the immunosuppressive therapy.

Pathogenic remarks. The difference in prevailing substrates (glucose and fatty acids) in the heart may affect myocardial HEPs metabolism. Excessive intracellular glucose uptake may down-regulate the expression of fatty acid-metabolizing genes through peroxisome proliferator-activated receptor-alpha repression (26) and may increase advanced glycation end-products with a consequent generation of reactive oxygen species affecting mitochondrial function (27). Excessive intracellular long-chain acyl-CoA esters may induce heart dysfunction in vivo in animal models (28) and in humans (29). Combined ^{31}P-MRS and ^{1}H-MRS will give insights to the cardiac HEPs and triglyceride content simultaneously, exploring the reciprocal relationships. Glucolipotoxicity has been indicted as a key factor inducing an initial adaptation and subsequent maladaptation of the heart to the diabetic environment (30,31).

Methodologic considerations. Because we did not perform angiography, the presence of subclinical atherosclerosis could not be entirely excluded. The worse functional and metabolic profile reported in the diabetic recipients of kidney-only transplantation versus the recipients of combined kidney-pancreas transplantation may be due to the persistence of the diabetes-induced microangiopathy; in fact, a different endothelial function (32) and ultrastructural microvascular features were already reported in these patients (33). The large volume of interest used in this work represents a limitation of the study, and contamination of blood could not be avoided as performed by using ^{31}P-spatial localization with optimum pointspread function (SLOOP)-MRS (34). Nevertheless, blood correction was appropriately performed as classically proposed (30,31) and the three-dimensional ISIS approach was appropriately used to study metabolic alterations involving the entire heart and not local abnormalities within a small amount of tissue. Another limitation of the study was the lack of the performance of a stress-test study potentially able to reveal stronger difference.

Conclusions. This work indicates that altered resting myocardial HEPs may contribute to the left ventricular functional alterations observed in type 1 diabetic-uremic patients. The cure of renal failure explored in diabetic recipients of kidney transplantation was associated with better parameters of heart function and metabolism; the combined cure of uremia and diabetes explored in recipients of combined kidney-pancreas transplantation was not associated with a different left ventricular function and HEPs in comparison with the control groups. A longitudinal study is warranted to confirm whether these alterations may be reversed and whether they may be a useful prognostic marker of heart failure in patients with type 1 diabetes and uremia.

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