# NT-proBNP plasma levels and echocardiographic assessment of cardiac function in patients after renal transplantation

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**Abstract**

**Background:** Cardiovascular diseases are the most important causes of death in patients with chronic renal disease (CRD). Successful renal transplantation (RTx) corrects water and electrolyte disturbances and decreases or eliminates anaemia. It favourably influences cardiac haemodynamics and reduces risk of cardiovascular events. NT-proBNP plasma concentration is one of the prognostic and risk factors in such cases, whereas echocardiography that enables evaluation of the left atrium and ventricle allows detailed analysis of haemodynamic condition of the heart.

**Aim:** To analyse NT-proBNP plasma concentration and selected echocardiographic parameters in patients after RTx at various time intervals after the procedure.

**Methods:** Seventeen patients after RTx were included in the study (age 46.5±16 years, 7 men and 10 women). NT-proBNP plasma level measurements and echocardiography were performed immediately before and at 3 and 6 months after RTx. Additionally, these parameters were assessed in patients receiving cyclosporine A (CsA) and tacrolimus (TAC).

**Results:** NT-proBNP plasma level decreases significantly after RTx (initially 4369±2420, at 3 months 2056±576, at 6 months 1580±572 pg/ml). In the TAC group, a significant reduction was observed at 3 months (from 13291±3563 to 1845±1022 pg/ml). In patients treated with CsA reduction occurred at 6 months after RTx (from 9447±3369 to 1246±436 pg/ml). At six-month follow-up significant changes in ejection fraction were not found. However, a significant increase in LV mass in CsA patients was observed.

**Conclusions:** Reduction of NT-proBNP levels seems to be more the result of transplanted kidney function than of an improvement in circulation. Significant LV mass increase in CsA patients may be a result of higher blood pressure levels observed before and after RTx.

**Key words:** renal transplantation, immunosupression, NT-proBNP, echocardiography

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## Introduction

Cardiovascular diseases are the major causes of death in patients with chronic renal disease (CRD). The risk considerably increases with the severity of renal failure and peaks in patients demanding renal replacement therapy. Successful renal transplantation (RTx) reduces the number of cardiovascular events; however, their incidence remains higher than in healthy subjects [1]. Frequent cardiovascular complications in CRD patients currently represent real diagnostic and therapeutic challenges. The reasons for high incidence of such complications are complex. Apart from classic cardiovascular risk factors, such as arterial hypertension, lipid abnormalities, sympathetic activation and fluid overload, CRD patients present with additional risk factors, including: anaemia, secondary hyperparathyroidism with calcium and phosphate metabolism abnormalities, high homocysteine levels and "microinflammations" [2].

Arterial hypertension and rapid atherosclerosis progression in CRD patients lead to left ventricular hypertrophy (LVH). Left ventricular hypertrophy extent correlates with CRD stage, more severe chronic renal disease, and more pronounced LVH. Left ventricular hypertrophy leads directly to heart failure with secondary neurohormonal and haemodynamic abnormalities involved in this process. Anaemia, systemic water and sodium retention and also arteriovenous fistulas in haemodialysed patients additionally promote LVH. For this reason, LVH assessment is one of the most important cardiovascular risk factors [3].

Apart from LVH, B-type natriuretic peptide (BNP) or its precursor N-terminal pro-B-type natriuretic peptide (NT-proBNP) plasma concentrations are additional factors investigated in mortality risk stratification in patients with heart failure. Secretion of these compounds is thought to compensate for neurohumoral changes present in heart failure, and may appear before the symptoms occur. These oligopeptide neurohormones play a role in water-sodium balance and cardiovascular homeostasis regulation. They increase glomerular filtration rate, decrease renal sodium reabsorbtion and inhibit renin and aldosterone secretion. These peptides induce a withdrawal of sympathetic activity and directly dilate blood vessels [4]. Their plasma concentrations have been reported to have a diagnostic and prognostic value in heart failure. Normal values of BNP and NT-proBNP blood concentrations vary depending on the method of assessment used, gender and age. The reference normal values for NT-proBNP for people <55 years are <155 pg/ml for women and <64 pg/ml for men, and for people aged 55-56 years <222 and <194 pg/ml, respectively [5].

Increased BNP levels are observed also in arterial and pulmonary hypertension, aortic stenosis, myocardial infarction, pulmonary diseases complicated by right ventricular heart failure, pulmonary embolism, thyreotoxicosis, Cushing's syndrome, primary hyperaldosteronism, hepatic cirrhosis with ascites and in renal failure in elderly people, whereas decreased concentrations are found in obese subject [6].

The following drugs affect NT-proBNP levels: corticosteroids, thyroid hormones, angiotensin-converting enzyme inhibitors, agonists and antagonists of adrenergic receptors [5]. Studies on relative risk of death or cardiovascular events in patients with heart failure as well as asymptomatic systolic dysfunction revealed that increased BNP or NT-proBNP levels are strong and independent predictors of poor outcome in all heart failure stages [7]. It was demonstrated that high BNP or NT-proBNP levels are present in patients with end-stage renal failure treated with chronic haemodialysis, irrespective of LVH and heart failure, whereas these concentrations in patients with cardiovascular disease are even higher [8]. Higher BNP levels are observed in haemodialysed patients in comparison with patients on peritoneal dialysis [9].

Renal transplantation reduces the risk of cardiovascular disease in comparison with dialysed patients [9], but it still remains higher than in healthy people. The risk of CVD results from severity of atherosclerosis before RTx, or insufficient modification of remaining risk factors, such as arterial hypertension, diabetes, hyperlipidaemia and cigarette smoking.

Our aim was to assess NT-proBNP plasma levels and selected LV and left atrial (LA) function echocardiographic parameters in RTx patients at various time intervals after the procedure. Additionally, these parameters were analysed with respect to immunosuppression with cyclosporine A (CsA) or tacrolimus (TAC).

## Methods

### Study population

Tests were performed in 17 patients aged 46.5±16 years (7 men and 10 women) in stage V of CRD. All subjects were on renal replacement therapy and qualified for RTx. Mean time from initiation of the dialysis programme was 28.1±5.9 months. Cold ischaemia time was 19.3±1.7 hours. Mean antigen compatibility score was 10.65±0.8 points. For 3 patients it was the second RTx procedure.

### Study protocol

NT-proBNP level was measured using electrochemiluminescence assay, by means of Elecsys proBNP (Roche) diagnostic test. Measurements were performed directly before RTx and at 3 and 6 months after the procedure. Furthermore, haemoglobin, creatinine and urea concentrations as well as echocardiography using Vivid 4 GE device were performed at the same time intervals.

The CsA and TAC administration effect on NT-proBNP levels and echocardiographic parameters was assessed in two groups of patients. CsA was the basic immunosuppressive agent in the first group (n=9), TAC in the second group (n=8).

### Immunosuppression

Prednisone and azathioprine were administered in all patients. Mean daily doses of these agents in analysed time intervals were: 6.3 – 5.4 – 4.8 mg/kg body weight for CsA, 0.76 – 0.12 – 0.08 mg/kg body weight for TAC, 166.2 – 76.9 – 77.3 mg of azathioprine and 26.8 – 13.2 – 9.6 mg of prednisone. The choice of immunosuppressive drug was based only on the clinical indications. No calcium or vitamin $D_3$ or its derivatives were given to patients after RTx.

### Statistical analysis

Results are presented in tables as x±SEM. Statistical analysis was performed using nonparametric Wilcoxon rank test for comparison of dependent variables. To compare the variables between two groups F Snedecor test was applied (ANOVA analysis of variance). A p value <0.05 was considered significant

## Results

During the analysed period of time after RTx, in the majority of patients renal graft function was stable with plasma creatinine level of about 1.5 mg/dl; only in 2 patients were these concentrations >2 mg/dl (2.1 and 4 mg/dl, respectively). In 2 patients acute tubular necrosis occurred after the procedure and 4 patients suffered from recurrent urinary tract infections.

At 3 and 6 months after the procedure, a significant reduction of NT-proBNP level in relation to the baseline values was observed in the whole study population, at a rate of 81 and 85%, respectively. A similar tendency and statistical relations, although at a different significance level, were seen in patients treated with TAC. Insignificant reduction of NT-proBNP level was observed also in the CsA group at 3 months, whereas at 6 months of follow-up NT-proBNP level was reduced by 90% and achieved statistical significance. The results are shown in Table I.

In the whole study population and in defined subgroups NT-proBNP levels did not correlate with creatinine, urea or haemoglobin concentrations.

In all patients systolic blood pressure decreased significantly ($p < 0.05$) by 12.6% as early as at 3 months after RTx. In the TAC group systolic and diastolic pressure was decreased at 3 months of follow-up by 13.8% and 12.1% respectively ($p < 0.05$). However, in the CsA group, despite the fact of a downward tendency of systolic blood pressure at 3 months after RTx, its decrease was insignificant. Blood pressure values in the CsA group were higher than in the TAC group (Table I).

At 3 months of follow-up, there was a positive correlation between systolic blood pressure and

**Table I.** Results od biochemical parameters and blood pressure values before and after transplantation

| Group | Months | NT-proBNP [pg/ml] | Systolic blood pressure [mmHg] | Diastolic blood pressure [mmHg] | Hb [g/dl] | Creatinine [mg/dl] | Urea [mg/dl] |
|---|---|---|---|---|---|---|---|
| CsA + TAC | 0 | 11369.2±2419.9 | 150.9±6.6 | 85.0±3.5 | 11.2±0.4 | 8.4±0.6 | 108.3±7.0 |
| (n=17) | 3 | 2056.3±576.4 [a] ** | 131.9±3.3 [a] * | 80.6±2.5 | 12.4±0.4 [a] * | 1.5 0.2 [a] *** | 57.2±5.7 [a] *** |
| | 6 | 1580.2±572.4 [b] *** | 135.6±4.0 | 79.1±2.9 | 13.0±0.3 [b] *** | 1.5±0.2 [b] *** | 62.5±7.4 [b] *** |
| CsA | 0 | 9447.4±3369.3 | 156.1±8.9 | 83.3±3.7 | 11.4±0.6 | 7.6±0.5 | 109.6±8.4 |
| (n=9) | 3 | 2244.4±662.1 | 137.2±3.7 | 83.9±3.8 | 12.7±0.4 [a] * | 1.4±0.2 [a] * | 58.0±6.3 [a] * |
| | 6 | 1245.7±436.1 [b] * | 145.0±4.9 | 82.8±4.5 | 13.3±0.4 [b, c] * | 1.4 ±0.1 [b] * | 66.8±7.8 [b] * |
| TAC | 0 | 13291.1±3562.5 | 145.0±10.0 | 86.9±6.5 | 10.9±0.7 | 9.4±1.0 | 106.8±12.1 |
| (n=8) | 3 | 1844.7±1021.6 [a] * | 125.0±4.8 [a] * | 76.4±2.4 [a] * | 12.1±0.7 | 1.6±0.3 [a] * | 56.3±10.3 [a] * |
| | 6 | 1956.5±1146.5 [b] * | 125.0±4.2 | 76.3±3.2 | 12.7±0.4 [b] * | 1.5±0.4 [b] * | 57.7±13.6 [b] * |

*[a] 0-3 months, [b] 0-6 months, [c] 3-6 months; *p <0.05; **p <0.005; ***p <0.001.*
*Abbreviations: CsA – cyclosporine A, TAC – tacrolimus, RTx – renal transplantation procedure*

**Table II.** Results of echocardiographic parameters before and after transplantation

| Group | Months | LVDd [cm] | LVSd [cm] | LA [cm] | LV mass | LV mass/m² | EF [%] |
|---|---|---|---|---|---|---|---|
| CsA + TAC | 0 | 5.35±1.8 | 3.58±1.8 | 4.38±1.3 | 366.59±27.8 | 206.15±17.9 | 61.6±2.7 |
| (n=17) | 3 | 5.32±1.7 | 3.55±2.0 | 4.29±1.6 | 374.24±26.6 [a] | 200.52±15.7 | 62.1±2.2 |
| | 6 | 5.43±1.6 | 3.66±1.7 | 4.42±1.4 [c] * | 383.47±26.6 [b, c] * | 211.15±16.2 [b, c] * | 61.8±2.3 |
| CsA | 0 | 5.01±2.2 | 3.38±2.6 | 4.23±2.2 | 288.56±23.7 | 163.55±11.9 | 61.7±4.5 |
| (n=9) | 3 | 5.22±2.0 | 3.39±2.3 | 4.01±2.6 | 302.33±25.8 a* | 170.62±12.1 a* | 63.8±3.6 |
| | 6 | 5.21±1.5 | 3.58±2.1 [c] * | 4.26±2.3 [c] * | 314.89±27.8 [b, c] * | 175.00±12.4 [b] * | 62.1±3.8 |
| TAC | 0 | 5.74±2.1 | 3.80±2.3 | 4.55±1.2 | 454.37±31.0 | 260.93±26.8 | 61.5±2.8 |
| (n=8) | 3 | 5.43±2.8 | 3.73±3.4 | 4.54±1.3 | 455.13±28.7 | 245.36±26.6 | 60.3±2.6 |
| | 6 | 5.68±2.9 | 3.75±2.8 | 4.61±1.2 | 460.63±29.4 | 257.62±24.4 [c] * | 61.4±0.7 |

*[a] 0-3 months, [b] 0-6 months, [c] 3-6 months; *p <0.05.*
*Abbreviations: CsA – cyclosporine A, TAC – tacrolimus, LVDd – left ventricular diastolic diameter, LVSd – left ventricular systolic diameter, LA – left atrium, LVmass – left ventricular mass, EF – left ventricular ejection fraction, RTx – renal transplantation procedure*

NT-proBNP levels in the whole study population as well as in the TAC group at the significance level p <0.05 and p <0.01, whereas at 6 months of follow-up such a correlation was observed only in the study population (p <0.05).

Echocardiography at 6 months after RTx revealed no significant changes of ejection fraction (EF) in all analysed groups. Furthermore, no significant differences in left ventricular diastolic diameter (LVDD), left ventricular systolic diameter (LVSD) or LA dimension between the analysed groups were seen. However, an increase in left ventricular mass (LVmass) was observed, also when indexed for body surface area (LVmass/m$^2$) in the whole study population at 6 months (p <0.05). In CsA patients systematic increase in LVmass and LVmass/m$^2$ was noted at 3, as well as at 6 months, and the differences were statistically significant (p <0.05). Also in TAC patients in consecutive months there was an increase in LVmass and LVmass/m$^2$, but this trend did not reach statistical significance.

Baseline LVmass and LVmass/m$^2$ values were higher in the TAC group than in the CsA group; these differences were statistically significant, with p <0.001 and p <0.005, respectively, and were maintained at 3 as well as at 6 months, with p <0.001 and p <0.05 respectively for LVmass, and with p <0.05 and p <0.005 for LVmass/m$^2$.

In the whole study population there was a positive correlation between NT-proBNP and LVmass at 3 months at the significance level of 0.05, and at 6 months after the procedure, at significance level of 0.01. In both CsA and TAC groups there was only a negative correlation between NT-proBNP and LVmass immediately after RTx (p <0.05) noted. Echocardiography parameters are displayed in Table II.

## Discussion

Cardiovascular diseases are the main causes of death in CRD patients. The risk increases with the severity of the disease and it is highest in patients requiring renal replacement therapy. Mortality rate in such patients due to cardiovascular events is assessed to be 20 times higher than in the general population [10]. This could be explained by the considerably greater number of classical or novel cardiovascular risk factors. Left ventricular hypertrophy is one of the most extensively investigated and recognised risk factors, and is present as early as in the third stage of CRD, whereas in the fifth stage it can be observed in more than 70% of patients qualified for renal replacement therapy [11]. The consequences include haemodynamic changes due to LV diastolic dysfunction and neurohormonal changes leading to heart failure [12, 13].

Renal transplantation gives a chance of reversing these unfavourable processes. With renal graft function restoration, some risk factors are partly or completely eliminated. Anaemia, calcium-phosphate abnormalities and, particularly, water-electrolyte balance become corrected. However, together with the introduction of calcineurine inhibitors to immunosuppressive therapy, which significantly extends graft survival, in 70-90% of recipients arterial hypertension ensues. Also corticosteroids used in this group of patients can contribute to arterial hypertension. Apart from the

above-mentioned factors, other causes of arterial hypertension in patients after RTx include: graft arterial stenosis, obstruction of urine outflow from the graft (lymphocoele, ureteral stenosis), long time of kidney ischaemia, and kidney harvested from an older donor, especially one with arterial hypertension [14].

High NT-proBNP concentrations in all analysed patients immediately after RTx are consistent with data from published reports. These papers demonstrated increased natriuretic peptides levels in CRD patients treated with haemodialysis [15]. The levels of the peptide also depend on the type of dialyzer membrane used. Increased NT-proBNP levels were observed after dialyses performed with low-flux dialyzer membranes, in contrast to procedures with high-flux membranes, in which a significant reduction in NT-proBNP levels was noted. Irrespective of the type of dialyzer membrane applied, haemodialysis reduces concentrations of BNP itself, although the concentrations of this peptide increase again between the procedures [16, 17].

Reduced NT-proBNP levels observed in all patients after RTx seem to confirm favourable renal graft influence on some risk factors. Despite its significant reduction during half-year follow-up, NT-proBNP levels did not reach reference values for a healthy population, which is also corroborated in the literature [15]. The NT-proBNP level reduction as early as at first 3 months after RTx in the TAC group may be the result of lower corticosteroid doses administered in these patients and of lower blood pressure levels, rarely requiring blood pressure lowering therapy with angiotensin-converting enzyme inhibitors, beta-blockers and diuretics. After 6 months the NT-proBNP levels were still lower than at baseline, although further reduction was not observed. They were higher than in the CsA group, and this seems to be attributable to higher values of LVmass and LVmass/$m^2$ on echocardiography. The absence of a statistically significant correlation between NT-proBNP and renal graft efficiency as well as anaemia should be emphasised, though increased glomerular filtration rate after organ transplant seems to contribute to the reduction of NT-proBNP levels by itself.

Renal function restoration after RTx and associated cardiovascular risk factor correction should be reflected in improvement of heart function assessed based on measurable echocardiographic parameters. During 6 months of follow-up no significant changes in EF, LVDd, LVSd or LA dimensions, suggesting myocardial remodelling, were observed, despite favourable biochemical changes (i.e. reduction of NT-proBNP concentrations). Perhaps 6 months of follow-up is too short for significant normalisation of echocardiographic parameters.

Special attention should be paid to the increase in LVmass and LVmass/$m^2$ observed in the whole study population at 3 and 6 months after RTx, seen also in specified subgroups. The present study revealed a significant increase in LVmass and LVmass/$m^2$ in patients treated with CsA at 3 and 6 months of follow-up. Coincidentally, initial values of blood pressure as well as values at 3 and 6 months of follow-up were higher than in the TAC group, which may be a cause of significant increase in LVmass and LVmass/$m^2$. Although patients receiving TAC had initially higher values of LVmass and LVmass/$m^2$, the increase of these parameters in the following 6 months was statistically insignificant.

Renal transplantation remains the optimal treatment of end-stage renal failure. It removes or considerably reduces unfavourable changes caused by loss of renal function, and also eliminates some cardiovascular risk factors. However, general cardiovascular risk remains higher than in the healthy population. This is a result of permanent changes in circulation in the course of long-term CRD, and also insufficient care to eliminate risk factors, among which arterial hypertension seems to be of the greatest importance.

## Conclusions

1. The NT-proBNP levels decrease significantly after renal transplantation. In patients receiving TAC a significant reduction was observed at 3 months after RTx, and in CsA patients at 6 months after the procedure.
2. Rapid decrease of NT-proBNP levels results from renal graft function restitution, rather than from an improvement in heart function.
3. A six-month follow-up revealed no significant changes in ejection fraction.
4. There was a significant increase in LVmass, especially in patients treated with CsA.
5. Significant LVmass in receiving CsA patients may be a result of higher values of blood pressure, observed before and after RTx.

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# Zachowanie się stężenia NT-proBNP w surowicy krwi oraz ocena echokardiograficzna funkcji serca u chorych po przeszczepieniu nerki

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## Streszczenie

**Wstęp:** Choroby układu sercowo-naczyniowego są najważniejszą przyczyną zgonów u chorych z przewlekłą chorobą nerek (PChN). Pomyślny zabieg przeszczepienia nerki (RTx) koryguje zaburzenia wodno-elektrolitowe oraz zmniejsza lub eliminuje niedokrwistość. Wpływa to korzystnie na hemodynamikę serca oraz zmniejsza ryzyko incydentów sercowo-naczyniowych. Jednym z czynników prognostycznych i oceny tego ryzyka jest stężenie NT-proBNP w surowicy, natomiast badanie echokradiograficzne z oceną lewego przedsionka i lewej komory pozwala dokładnie określić stan hemodynamiczny serca.

**Cel:** Ocena stężenia NT-proBNP w surowicy krwi oraz wybranych parametrów echokardiograficznych u pacjentów po RTx w różnym czasie po zabiegu.

**Metodyka:** W badaniu uczestniczyło 17 chorych po RTx (wiek 46,5±16 lat, 7 mężczyzn i 10 kobiet). Stężenie NT-proBNP i badanie echokardiograficzne wykonywano bezpośrednio przed oraz w 3. i 6. mies. od RTx. Dodatkowo oceniono ww. parametry u pacjentów leczonych cyklosporyną A (CsA) oraz takrolimusem (TAC).

**Wyniki:** Stężenie NT-proBNP zmniejsza się istotnie po RTx (wyjściowo: 4369±2420, po 3 mies.: 2056±576, po 6 mies.: 1580±572 pg/ml). W grupie leczonych TAC znamienne obniżenie zaobserwowaliśmy w 3. mies. (z 13291±3563 do 1845±1022 pg/ml). U przyjmujących CsA obniżenie wystąpiło po 6 mies. od RTx (z 9447±3369 do 1246±436 pg/ml). Półroczna obserwacja nie wykazała istotnych zmian frakcji wyrzutowej. Stwierdzono natomiast znamienne zwiększenie LVmass u chorych leczonych CsA.

**Wnioski:** Zmniejszenie stężenia NT-proBNP wydaje się bardziej wynikać z zadowalającej czynności przeszczepionej nerki niż z poprawy wydolności krążenia. Znamienny wzrost LVmass w grupie leczonych CsA może wynikać z wyższych wartości ciśnienia tętniczego przed i po RTx.

**Słowa kluczowe:** przeszczepienie nerki, immunosupresja, NT-proBNP, echokardiografia

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