JMB 2007; 26 (3) DOI: 10.2478/v10011-007-0025-5

UDK 577.1 : 61 **ISSN 1452-8258**

JMB 26: 215–219, 2007

Original paper
Originalni naučni rad

COMPARISON OF BIOCHEMICAL MARKERS OF BONE TURNOVER IN PATIENTS ON CONTINUOUS AMBULATORY PERITONEAL DIALYSIS AND MAINTENANCE HEMODIALYSIS

POREĐENJE BIOHEMIJSKIH MARKERA KOŠTANOG PROMETA KOD PACIJENATA NA KONTINUALNOJ AMBULATORNOJ PERITONEALNOJ DIJALIZI I HEMODIJALIZI

Neda Novaković[1], Svetlana Pejanović[2], Dijana Jovanović[2], Nada Majkić-Singh[1]

[1]Institute of Medical Biochemistry, Clinical Center of Serbia, Belgrade
[2]Institute of Nephrology and Urology, Emergency Center of Serbia, Belgrade

Summary: The aim of this study was to compare the biochemical markers of bone formation – bone isoenzyme alkaline phosphatase, /BALP/ bone resorption – tartarat resistant acid phosphatase /TRAP/ and parameters of bone turnover (Ca, P and iPTH) between 108 patients on continuous ambulatory peritoneal dialysis (CAPD) and 112 patients on hemodialysis (HD). Parameters were determined by spectrophotometry (TRAP, Ca, P), electrochemiluminiscent method (iPTH) and electrophoresis (BALP). There was significant correlation between BALP levels of CAPD patients and Ca values of HD patients ($r = 0.216$, $p < 0.05$), but comparison of P, iPTH and BALP showed no significant difference between the groups. We found significant correlation between BALP and TRAP ($r = 0.570$, $p < 0.01$) and BALP and iPTH values ($r = 0.551$, $p < 0.01$) in HD patients. Also, significant correlations in CAPD patients between BALP and TRAP ($r = 0.194$, $p < 0.05$) and BALP and iPTH values ($r = 0.283$, $p < 0.01$) were found. Our results suggest that the effect of CAPD and HD on bone turnover may be monitored most conclusively via BALP levels. This biochemical marker is a very good index of bone turnover in patients undergoing dialysis and it could indicate an early stage of decreased bone turnover.

Keywords: biochemical markers of bone turnover, renal osteodystrophy, dialysis

Kratak sadržaj: Cilj rada bio je da se uporede biohemijski markeri formiranja kostiju – koštani izoenzim alkalne fosfataze /BALP/, marker resorpcije kostiju – tartarat rezistentna kisela fosfataza /TRAP/, i parametri koštanog prometa (Ca, P i PTH) između 108 pacijenata na kontinuiranoj ambulatornoj peritonealnoj dijalizi (CAPD) i 112 pacijenata na hemodijalizi (HD). Parametri su određivani spektrofotometrijski (Ca, P i TRAP), elektrohemiluminiscentnom metodom (iPTH) i elektroforetski (BALP). Međusobnim poređenjem parametara HD i CAPD uočena je statistički značajna korelacija između BALP vrednosti kod pacijenata na CAPD i Ca vrednosti kod pacijenata na HD ($r = 0,216$, $p < 0,05$), ali poređenjem P, iPTH i BALP pokazano je da ne postoji statistički značajna razlika između grupa. Kod HD pacijenata su dobijene statistički značajne korelacije između nivoa BALP i TRAP ($r = 0,570$, $p < 0,01$), kao i BALP i iPTH ($r = 0,551$, $p < 0,01$). Takođe je dobijena statistički značajna korelacija kod CAPD pacijenata između nivoa BALP i TRAP ($r = 0,194$, $p < 0,05$), i BALP i iPTH ($r = 0,283$, $p < 0,01$). Ovi rezultati sugerišu da efekat obe vrste dijalize na koštani promet može precizno da se prati preko nivoa BALP. Ovaj biohemijski marker predstavlja veoma dobar indeks koštanog prometa kod pacijenata na dijalizi i može ukazati na rane stadijume usporenog koštanog prometa.

Ključne reči: biohemijski markeri koštanog prometa, renalna osteodistrofija, dijaliza

Introduction

Patients undergoing dialysis have disarrangement of bone remodelling as one of the number of complications. This disorder is defined as renal osteodystrophy (ROD) which includes high and low turnover disease lesions. For a complete diagnostic definition of different bone patterns found in dialysed

Address for correspondence:

Neda Novaković
Institute of Medical Biochemistry, Clinical Center of Serbia
Višegradska 26, 11129 Belgrade, Serbia
e-mail: nedanvkvc@yahoo.com

patients, histologic examinations are required. However, this can only be performed in specialized centres. To avoid this invasive procedure, clinical and laboratory techniques could be used.

For more than a decade, biochemical parameters of metabolic bone disease have been analysed (1). The most analysed serum markers are: total calcium (Ca), inorganic phosphate (P), bone alkaline phosphatase (BALP), tartarat resistant acid phosphatase (TRAP) and intact parathyroid hormone (iPTH) (2).

Many authors found good correlations between histomorphometric and biochemical parameters of bone turnover in dialysed patients (1–3).

Dialysed patients have increased risk of adynamic bone disease (ABD) which is one of the low turnover disease lesions. There are different causes of this disorder but almost every patient has relatively low levels of iPTH and hypercalcemia. Also, for early detection and proper treatment of ABD it is important to consider different effects of dialysis types on biochemical markers of bone turnover. Many studies also explained these effects on histomorphometric parameters of bone mineral density (4–6).

Serum iPTH levels have good correlations with bone turnover, but their values reflect bone metabolism indirectly (7). Contrary, BALP and TRAP are produced only from osteoblast and osteoclast and directly present bone formation and resorption. Also, these enzymes activities are independent of glomerular filtration rate, and BALP has better correlation than iPTH with most histomorphometric parameters of bone turnover (8, 9). TRAP can be a good indicator of bone resorption in the early stages of renal osteodystrophy, which is very important for early detection of this disorder (10).

In dialysed patients there are different prevalences of ABD depending on the dialysis type and duration of dialysis, and many studies have explained this relation (11, 12).

In this study we examined correlations of Ca, P, BALP, TRAP and iPTH between patients undergoing continuous ambulatory peritoneal dialysis (CAPD) and hemodialysis (HD). With results of these correlations it could be possible to explain if there is a significant difference between different dialysis types in biochemical markers of bone turnover in dialysed patients.

Material and Methods

This study was performed on 220 dialysed patients who were divided into two groups. There were 108 patients undergoing CAPD, while 112 patients were on maintenance hemodialysis (HD). Patients were treated at the Institute of Urology and Nephrology, Emergency Center of Serbia. Blood samples for the determination of biochemical markers were obtained immediately before dialysis session. Biochemical analyses were performed at the Institute of Medical Biochemistry, Polyclinic Laboratory, Clinical Center of Serbia.

Values of Ca, P and TRAP activities were analysed with standard biochemical methods and commercial reagents of Olympus company, on a biochemical analyser Olympus AU2700 ISE (Olympus Diagnostica GmbH, Hamburg, Germany).

TRAP values were accounted from differences between directly measured total acid phosphatase and total prostatic phosphatase.

iPTH levels were measured with electrochemiluminiscent immunoassay using commercial reagents of Roche company, on a Roche Elecsys 1010/2010 immunoassay analyser (Germany).

BALP activities were determined using electrophoretic technique on an automatic electrophoretic system HYDRASYS (SEBIA, France). Using this technique, it is possible to exclude the potential influence of other alkaline phosphatase isoenzymes activities. Different methods for alkaline phosphatase isoenzymes show good reciprocal correlation, but the electrophoretic method is more sensitive than the other methods when measuring low concentrations of BALP (13).

For statistical analysis we used Kolmogorow-Smirnow test to check the normal distribution of analysed biochemical markers. Also, we used the Mann-Whitney U test and Spearmen correlation coefficient by SPSS 11.5 for Windows for statistical analysis. With linear regression analysis we presented the correlation of the same markers of bone turnover between two different groups of dialysed patients.

Results

In *Table I* we present the obtained results of the determined biochemical markers of bone turnover in HD and CAPD.

Using statistical analysis on two groups of patients we obtained significant correlation of BALP and TRAP ($r = 0.570$, $p < 0.01$), and BALP and iPTH ($r = 0.551$, $p < 0.01$) in HD patients, as well as significant correlation of BALP and TRAP ($r = 0.194$, $p < 0.05$) and BALP and iPTH ($r = 0.283$, $p < 0.01$) in CAPD patients. There was significant correlation of BALP and Ca ($r = -0.213$, $p < 0.05$) in CAPD patients. Reciprocal comparison of HD and CAPD parameters revealed significant correlation only between BALP of CAPD patients and Ca of HD patients ($r = 0.216$, $p < 0.05$). Comparison of P, iPTH and BALP showed no significant difference between groups, in distinction from Ca and TRAP ($p < 0.001$).

Table I Results of the determined biochemical markers of bone turnover in studied hemodialysis and continuous ambulatory peritoneal dialysis patients. Results are expressed as median and range.

Parameters	HD	CAPD	Reference values
Ca, mmol/L	2.39 (1.63–2.71)[b]	2.25 (1.74–2.96)[a]	2.10–2.70
P, mmol/L	1.63 (0.61–6.92)	1.52 (0.79–2.94)	0.80–1.60
TRAP, IU/L	8 (5–19)[b]	6 (2–14)[a]	< 7
BALP, IU/L	20 (3–145)	20 (2–262)	f: ≤ 44 m: ≤ 73
iPTH, pg/mL	142.4 (1.00–1767.0)	128.0 (2.3–835.5)	11–62

[a] $p < 0.001$ as compared to HD
[b] $p < 0.001$ as compared to CAPD

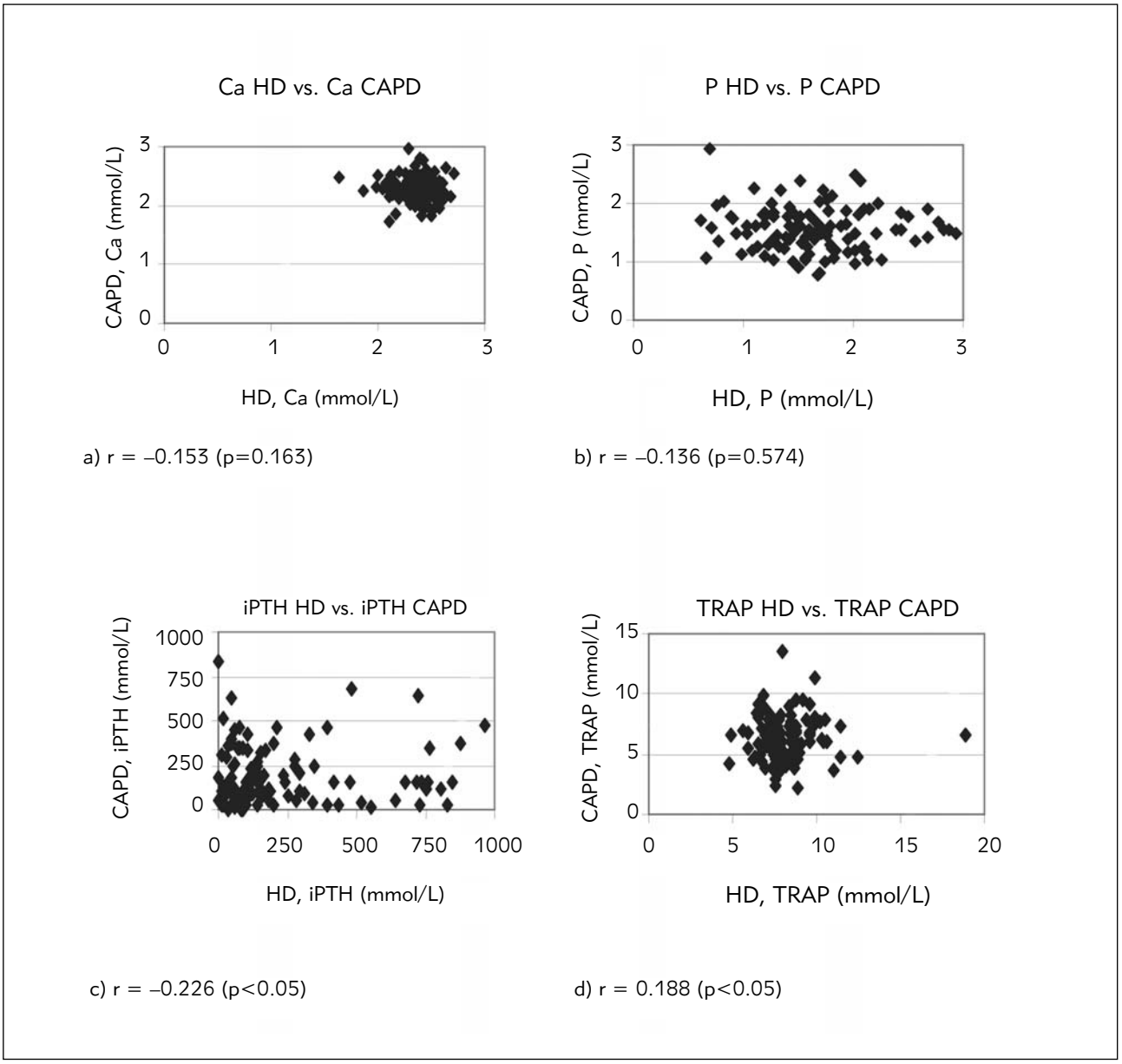

Figure 1 Correlations of measured biochemical markers of bone turnover between dialysed patients divided according to dialysis type: a) Ca HD vs. Ca CAPD, b) P HD vs. P CAPD, c) iPTH HD vs. iPTH CAPD and d) TRAP HD vs. TRAP CAPD.

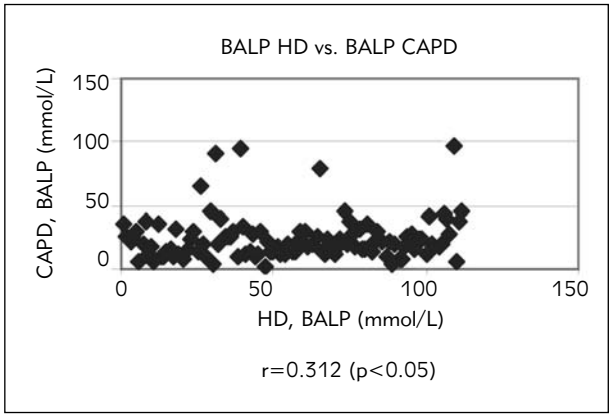

Figure 2 Correlation of BALP between dialysed patients divided according to dialysis type: BALP HD vs. BALP CAPD.

Levels of P and TRAP activities of HD patients were slightly above the reference ranges that we used in our laboratory. Determined TRAP activities can point to decreased bone turnover. Also, BALP and iPTH levels were below the cut-off values recommended for decreased bone turnover according to Kidney Disease Outcomes Quality Initiative (K/DOQI) guidelines: for BALP (≤ 27 IU/L) and for iPTH (≤ 150 pg/mL) (6).

In *Figure 1* we present correlations of measured biochemical markers of bone turnover between dialysed patients divided according to dialysis type: a) Ca HD vs. Ca CAPD, b) P HD vs. P CAPD, c) iPTH HD vs. iPTH CAPD and d) TRAP HD vs. TRAP CAPD.

In *Figure 2* we present the correlation of BALP between dialysed patients divided according to dialysis type: BALP HD vs. BALP CAPD.

Obtained linear regression equations for Ca, P, BALP, iPTH and TRAP in HD (x) and CAPD (y) were: $y = 2.564974 - 0.124x$, $y = 1.602582 - 0.0295x$, $y = 25.75178 + 0.043506x$, $y = 178.046 + 0.03886x$ and $y = 5.944932 + 0.05083x$, respectively.

Discussion

There are two different types of dialysis which are commonly used as treatment in patients with end stage renal disease. It depends on patients which type of dialysis could be used. There are different ways of maintenance of these dialysis types and perhaps this has influence on their effects on kidneys. And, as one of the complications, some of the different levels of renal osteodystrophy could develop.

Our results show very good correlations between biochemical markers of bone turnover in groups of patients undergoing two different dialysis types. Obtained findings support the studies of Hampson et al. (11). They found no significant differences in serum calcium, phosphorus and parathyroid hormone between patients on HD and CAPD.

Ueda et al. (14) explained the advantage of serum BALP values as a clinically useful marker for predicting bone mineral density reduction in hemodialysis patients with low iPTH. Also, Coutteneye et al. (15) presented the high prevalence of ABD diagnosed by biochemical markers in a wide sample of the European CAPD population. They show good sensitivities and specificities of low BALP and low iPTH levels in CAPD patients, as good markers of ABD. Our results of low BALP and low iPTH levels between HD and CAPD show good correlations and indicate decreased bone turnover in these two groups of patients. But BALP levels are independent of any influence which induces bone response to iPTH synthesis. This points to BALP as a better indicator for bone remodeling in dialysed patients.

Torres et al. (12) explained the different bone response to iPTH in CAPD and HD patients. It seems to be related to shorter duration of dialysis rather than to any effect of dialysis modality per se. Our results show evident effect of dialysis independent of type on the iPTH levels which are significantly reduced by therapy. But, when we consider duration of dialysis, there is much effect on iPTH lowering in CAPD, although these patients were on maintenance dialysis for a shorter period.

Many authors (11, 16) point at the relationship between serum levels of Ca and P with iPTH in patients on hemodialysis and continuous ambulatory peritoneal dialysis. Our results show similar correlation in the sense of a desirable increase of analysed serum markers levels after dialysis session. Beside higher P levels in HD patients, Ca values were within the reference range. CAPD patients have lower TRAP and iPTH values, but almost unchanged BALP levels. As we considered that CAPD were dialysed shorter than HD, previous results can induce the effect of dialysis, but also the effect of dialysis duration on the level of bone tissue resorption. Future studies have to investigate how the effect of dialysis type contributes to bone resorption.

No statistically significant differences were recorded between BALP values of either of the examined groups and reference ranges. This is in accordance with previous clinical studies showing BALP as an independent biochemical marker of bone turnover in dialysed patients, but also in very good correlation with histomorphometric findings. Our results show significant correlation in levels of BALP between two dialysed groups. Also, in both analysed groups these values were below the recommended levels according to K/DOQI for decreased bone turnover. This suggests possible decreased bone turnover in both CAPD and HD patients.

On the basis of the results obtained throughout the present study, it may be concluded that there is significant correlation between both examined types of dialysis. Biochemical markers of bone turnover

show a similar trend in both dialysis types, and the effect of continuous ambulatory peritoneal dialysis and hemodialysis on bone turnover may be monitored most conclusively via BALP levels. Although bone biopsy is considered the golden standard of bone turnover definition, determination of catalytic BALP activity in blood sera, as a non-invasive method, is a very good index of bone turnover in patients undergoing dialysis.

Acknowledgment. This study was conducted as a part of the Project No. 145010 financially supported by the Ministry of Science, Technology and Development of the Republic of Serbia.

References

1. Urena P, De Vernejoul MC. Circulating biochemical markers of bone remodeling in uremic patients. Kidney Int 1999; 55: 2141–56.
2. Ferreira MA. Diagnosis of renal osteodystrophy: when and how to use biochemical markers and non-invasive methods; when bone biopsy is needed. Nephrol Dial Transplant 2000; 15: 8–14.
3. Stepan JJ. Clinical value of the biochemical markers of bone remodelling in the assessment of bone metabolic disease. Jugoslov Med Biohem 2006; 25: 241–8.
4. Couttenye MM, D'Haese PC, Van Hoof VO, Lemoniatou E, Goodman W, Varpooten GA, De Broe ME. Low serum levels of alkaline phosphatase of bone origin: a good marker of adynamic bone disease in haemodialysis patients. Nephrol Dial Transplant 1996; 11: 1065–72.
5. Alvarez L, Torregrosa JV, Peris P, Monegal A, Bedini JL, Matrinez De Osaba MJ, Filella X, Martin G, Ricos C, Oppenheimer F, Ballesta AM. Effect of hemodialysis and renal failure on serum biochemical markers of bone turnover. J Bone Miner Metab 2004; 22: 254–9.
6. Noordzij M, Korevaar JC, Boeschoten EW, Dekker FW, Bos WJ, Krediet RT. Netherlands Cooperative Study on the Adequacy of Dialysis (NECOSAD) Study Group, The Kidney Disease Outcomes Quality Initiative (K/DOQI) Guideline for Bone Metabolism and Disease in CKD: association with mortality in dialysis patients. Am J Kidney Dis 2005; 46: 925–32.
7. Sankarasubbaiyan S, Abraham G, Soundararajan P, Chandrasekaran V, Padma C. Parathyroid hormone and biochemical profile in chronic kidney disease patients in South India. Hemodial Int 2005; 9: 63–7.
8. Coen G, Ballanti P, Bonucci E, Calabria S, Centorrino M, Fassino V, Manni M, Mantella D, Mazzaferro S, Napoletano I, Sardella D, Taggi F. Bone markers in diagnosis of low turnover osteodystrophy in haemodialysis patients. Nephrol Dial Transplant 1998; 13: 2294–302.
9. Jorge C, Gil C, Possante M, Silva E, Andrade R, Santos N, Cruz A, Teixeira R, Ferreira A. Bone Alkaline Phosphatase besides Intact Parathyroid Hormone in Hemodialysis Patients–Any Advantage? Nephron Clin Practice 2005; 101: 122–7.
10. Halleen JM, Tiitinen SL, Ylipahkala H, Fagerlund KM, Vaananen HK. Tartrate-resistant Acid Phosphatase 5b (TRAP 5b) as a Marker of Bone Resorption. Clin Lab 2006; 52: 499–509.
11. Hampson G, Vaja S, Evans C, Chesters CA, Pettit R, Evans W, Thomas D, Seed PT, Fraser WD. Comparison of the humoral markers of bone turnover and bone mineral density in patients in haemodialysis and continuous ambulatory peritoneal dialysis. Nephron 2002; 91: 94–102.
12. Torres A, Lorenzo V, Hernandez D, Rodriguez JC, Concepcion MT, Rodriguez AP, Hernandez A, De Bonis E, Darias E, Gonzalez-Posada JM, Losada M, Rufino M, Felsenfeld AJ, Rodriguez M. Bone disease in predialysis, hemodialysis and CAPD patients: Evidence of a better bone response to PTH. Kidney Int 1995; 47: 1434–42.
13. Pricea CP, Milligan TP, Darte C. Direct comparison of performance characteristics of two immunoassays for bone isoform of alkaline phosphatase in serum. Clin Chem 1997; 43: 2052–7.
14. Ueda M, Inaba M, Okuno S, Maeno Y, Ishimura E, Yamakawa T, Nishizawa Y. Serum BAP as clinically useful marker for predicting BMD reduction in diabetic hemodialysis patients with low PTH. Life Sci 2005; 77: 1130–9.
15. Couttenye M, D'Haese P, Deng J, Van Hoof V, Verpooten. High prevalence of adynamic bone disease diagnosed by biochemical markers in a wide sample of the European CAPD population. Nephrol Dial Transplant 1997; 12: 2144–50.
16. Spiechowicz U, Kokot F, Wiecek A. Markers of calcium-phosphate metabolism and bones alterations in long-term kidney transplant patients. Przegl Lek 2003; 60: 690–4.

Received: January 15, 2007
Accepted: May 15, 2007