ORIGINAL ARTICLE

Inflammation and anaemia as predictors of cardiovascular mortality in hemodialysis patients

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Abstract

Back ground: Cardiovascular diseases are the most common causes of death among hemodialysis (HD) patients, yet the risk factors for these events have not been well established. Our study objective was to determine predictors of cardiovascular mortality, considering the non-traditional/disease-related and treatment-related/ cardiovascular risk factor in HD patients.

Material and Methods: Disease-related cardiovascular risk factors, such as anaemia, calcium-phosphate disorders, nutrition-inflammation and treatment/dialysis-related cardiovascular risk factors such as HD dose, using the index Kt/V were analyzed in 214 patients on HD. Mortality was monitored prospectively over a two year period.

Results: Fifty-three of the 214 HD patients died during the follow-up period and the main cause of death was cardiovascular events (56.6%), followed by infection/sepsis (26.4%). The patients who died were significantly older than those alive, had significantly lower serum levels of hemoglobin (Hb), albumin and Kt/V. Serum levels of calcium, C-reactive protein (CRP) and fibrinogen were significantly higher in patients who died during the follow-up period. Kaplan-Meier analysis showed that the all cause and cardiovascular mortality was considerably higher in patients with Hb<110g/l, albumin <40g/l, CRP>8mg/l and spKt/V<1.2 (log rank, p=0.000/p=0.000, p=0.000/p=0.001, p=0.000/p=0.000, p=0.000/p=0.000), respectively. No difference in cardiovascular mortality was observed between the fibrinogen <4g/l> levels. High CRP, low Hb levels and low spKt/V were significant predictors of all-cause mortality, but low albumin and high fibrinogen levels were not in the Cox proportional hazards analysis. When only cardiovascular mortality was entered into the Cox model, high CRP and low Hb levels were the only significant predictors for mortality.

Conclusions: It can be concluded that, inflammation (elevated CRP) and anaemia (decreased Hb), were identified as significant independent non-traditional, disease-related cardiovascular risk factors that predict cardiovascular mortality in HD patients. *Hippokratia 2007; 11 (1):39-43*

Key words: *cardiovascular mortality, non-traditional cardiovascular risk factor, hemodyalisis, CRP*

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Cardiovascular disease (CVD) is the leading cause of morbidity and mortality in HD patients, accounting for almost 40% of hospitalizations and 50% of deaths, and after stratification for age, race and gender, the cardiovascular mortality rate in these patients is ´10-20 times that in general population[1,2].

Identification of cardiovascular risk factors in HD patients is important in order to devise preventive and interventional strategies. However, although the prevalence of traditional cardiovascular risk factors (age, male gender, hypertension, diabetes, dyslipidemia, and physical inactivity) is high in dialysis patients, the extent and severity of CVD is clearly disproportionate to the underlying risk factor profile in this population[3]. Besides the traditional risk factor, HD patients face the additional non-traditional /disease-related and treatment-related/ cardiovascular risk factors, as anaemia, hyperhomocysteinemia, hyperparathyroidism, hypoalbuminemia, chronic inflammation. The weight of these risk factors in comparison with traditional risk factors is still undefined, although many are convinced that non-traditional, especially related to the uremic state risk factors, play a major role in CVD in hemodyalisis patients[4].

Our study objective was to determine predictors of cardiovascular mortality, considering the disease-related and dialysis-related cardiovascular risk factors, in a cohort of hemodialysis patients treated in a single hemodialysis center.

Material – methods

Two hundred fourteen patients on HD (129 men and 85 women) were followed up for a period of 24 months. An inclusion criterion was patients undergoing

HD for at least 6 months. In 209 patients, dialysis was performed three times per week, in 5 patients two times per week. End-stage renal failure was due to interstitial nephritis in 39 (18%), diabetes mellitus in 37 (17%), nephroangiosclerosis in 38 (17.7 %), glomerulonephritis in 38 (17.7%), polycystic kidney disease in 16 (7.5%), plasmocytoma in 3 (1.4%) and other or unknown cause in 43 (20%) patients. Hemodialysis treatment was performed using conventional bicarbonate-buffered dialysate in all patients.

We analyzed two types of non-traditional cardiovascular risk factors: *disease-related cardiovascular risk factors* such as anaemia, calcium-phosphate disorders, nutrition-inflammation and *treatment/dialysis- related cardiovascular risk factor* as HD dose, using single-pool spKt/V and equilibrated eKt/V. Serum levels of anaemia parameters: hemoglobin (Hb), hematocrit (Hct), red blood cells (RBC), white blood cells (WBC), serum iron, total iron-binding capacity and ferritin; calcium-phosphate disorders parameters: calcium, phosphate and PTH; and nutrition/inflammation parameters:albumin and CRP were measured monthly, but only serum fibrinogen every third month. The serum concentration of CRP was measured by a nephelometric immunoassay and fibrinogen was measured by the thrombin time method in a blood sample anticoagulated with sodium citrate.

Single-pool spKt/V was calculated monthly, using a second – generation formula Daugirdas 2 and eKt/V was calculated also monthly, using Daugirdas – Schneditz formula,

spKt/V = -Ln (R-0.008 * t) + (4-3.5 R) * UF/W

where R=postdialysis/predialysis blood urea nitrogen, t=dialysis hours, UF=predialysis-postdialysis weight change, and W=postdialysis weight.

Statistical analysis: Comparison between hemodialysis patients who died and those alive after 24 months of follow-up period was performed using t-test for normally distributed and Mann-Whitney rank-sum test for non-normally distributed variables. The risk of death among patients with hemoglobin (< 110g/l >), albumin (< 40 g/l >), fibrinogen (< 4g/l >), CRP (< 8mg/l >) levels and spKt/V (< 1.2 >) were compared using Kaplan-Meier survival function analysis. Differences in survival were assessed with the log rank test. Multivariate analysis was performed using the Cox proportional hazards model to determine which factors were most closely associated with the all-cause and cardiovascular mortality.

Results

Comparison between patients who died vs. those alive at 24 months

The mean age of the patients was 55.90±12.87 years (men 55.33±12.61, women 56.78±13.19) and duration of hemodialysis treatment in months was 89.43±71.98, range from 6 to 308 months (men 84.73±71.18, women 96.58±72.58). The patients who died were significantly older than those alive at 24 months, 61.30±10.01 vs. 54.12±13.27 (p=0.0003), however duration on HD in months did not differ between the two groups, 80.57±75.82 vs. 92.35±70.89 (p=0.3032).

Among levels of anaemia parameters, patients who died had significantly lower serum levels of Hb, Hct, RBC and serum iron, whereas total iron-binding capacity and ferritin levels did not differ between the two groups. The serum level of calcium was significantly higher in patients who died vs. those alive, but there was no difference in other parameters of calcium-phosphate disorders: serum phosphate (1.48±0.40 vs. 1.44±0.34, p=n.s.), PTH (169.34±207.30 vs. 190.64 ±215.56, p=n.s.) and CaxP product (3.31±0.95 vs. 3.30±0.80, p=n.s.). During the follow-up period patients who died had significantly lower serum levels of albumin, and they had significantly higher serum levels of CRP and fibrinogen (Table 1).

Table1. *Parameters of patients who died vs. those alive during follow-up period*

Parameters	Dead (No = 53)	Survived (No=161)	p
Hb g/l	93.13 ± 15.43	109.16 ± 12.08	0.0000
Hct	0.29 ± 0.05	0.33 ± 0.04	0.0000
RBC ($x10^{12}$/L)	3.19 ± 0.58	3.59 ± 0.46	0.0000
WBC ($x10^{9}$/L)	7.230 ± 3.01	6.35 ± 1.40	0.0044
Ca (mmol/L)	2.30 ± 0.16	2.22 ± 0.15	0.0067
Alb g/l	36.06 ± 4.17	39.74 ± 3.31	0.0000
Fibrinogen g/l	5.28 ± 1.28	4.41 ± 0.95	0.0001
CRP mg/l	40.05 ± 35.44	8.71 ± 7.69	0.0000

Mean spKt/V and eKt/V were significantly lower in patients who died compared with those alive at 24 months (Table 2).

Table2. *Parameters of patients who died vs. those alive during follow-up period*

Parameters	Dead (No = 53)	Survived (No=161)	p
UF (l)	2.75 ± 0.73	3.31 ± 0.88	0.00004
HD treatment (hours)	3.81 ± 0.33	4.03 ± 0.19	0.00000
spKt/V	1.15 ± 0.25	1.21 ± 0.19	0.04140
eKt/V	0.99 ± 0.21	1.06 ± 0.17	0.01992

Analysis of two-year all-cause and cardiovascular mortality

During the follow-up period of two years, 53 out of 214 patients (24.7%) had died (58.5% men N=31; 41.5% women N=22) most from cardiovascular events (30 out of 53; 56.6%), for example, myocardial infarction (36.6%, N=11), congestive heart failure (40%, N=12) or stroke (23.3%, N=7). Noncardiac causes of death were septicemia (26.4%, N=14), neoplasms (7.5%, N=4) or other unknown causes (9.4%, N=5).

Kaplan-Meier analysis on all-cause and cardiovascular mortality

The patients were divided according to Hb <110g/l>, albumin <40g/l>, fibrinogen <4g/l>, CRP <8mg/l> levels and spKt/V <1.2>. The all cause mortality rate was considerably higher in patients with Hb <110g/l (log rank, p=0.00000), albumin < 40g/l (log rank, p=0.00082), fi-

brinogen >4g/l (log rank, p=0.01558), CRP >8mg/l (log rank, p=0.00000) and spKt/V <1.2 (log rank, p=0.00003) than in patients with Hb >110 g/l, albumin >40g/l, fibrinogen <4g/l, CRP <8mg/l levels and spKt/V >1.2. (Figure 1,2,3; curves for spKt/V and fibrinogen not shown). The levels of Hb<110g/l (log rank, p=0.00001), albumin<40g/l (log rank, p=0.00125), CRP>8mg/l (log rank, p=0.00000) and spKt/V<1.2 (log rank, p=0.00042) were strong predictors of cardiovascular mortality, also. No difference in cardiovascular mortality was observed between the fibrinogen levels (log rank, p=0.08817).

Cox Proportional Hazard Analysis on all-cause and cardiovascular mortality

The Cox model was used to identify significant predictors of all-cause and cardiovascular mortality in HD patients. High CRP, low Hb levels and low spKt/V were significant predictors of all-cause mortality, but low albumin and high fibrinogen levels were not, although they were associated with a reduced survival rate in the Kaplan-Meier analysis.

When only cardiovascular mortality was entered into the Cox model, high CRP and low Hb levels were still more significant than low albumen, high fibrinogen, and low spKt/V in predicting mortality (Table 3).

Figures 1, 2, 3. *Kaplan Meier survival curves*

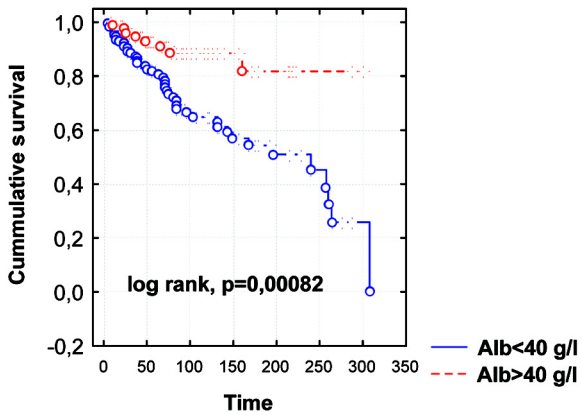

Figure 1. *Patients with Alb levels above and below 40g/L*

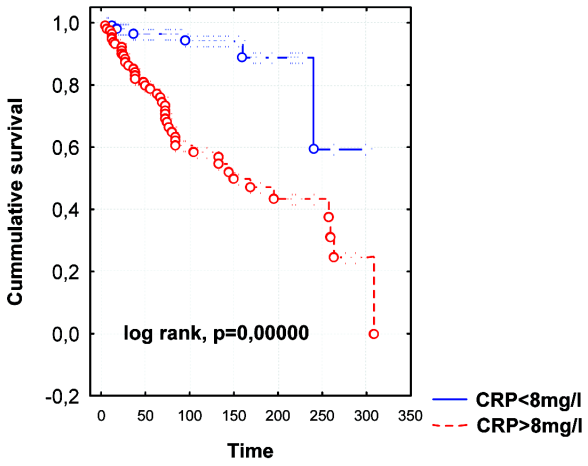

Figure 2. *Patients with CRP levels above and below 8mg/L*

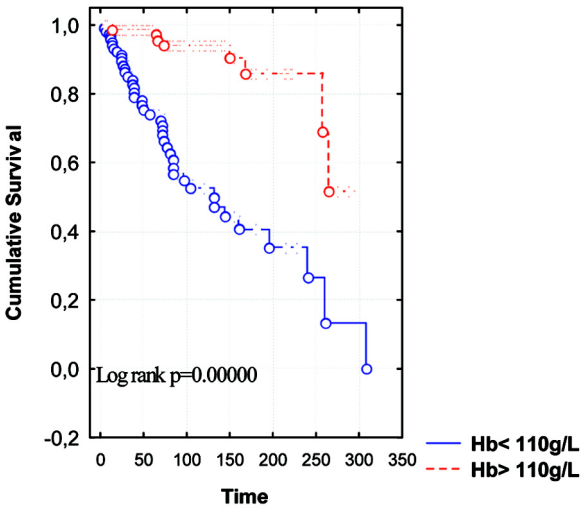

Figure 3. Patients with Hb levels above and below 110g/l

Table 3. *Parameters that predict all-cause and cardiovascular mortality in the Cox Proportional Hazards Model*

	All-cause mortality			*Cardiovascular mortality*		
Parameter	*Beta*	*t-value*	*p*	*Beta*	*t-value*	*p*
Hemoglobin	-0.0423	-4.0806	0.0000	-0.0371	-2.9240	0.0034
CRP	0.0193	4.7210	0.0000	0.0242	1.0245	0.000
spKt/V	-2.3470	-2.8651	0.0041	-	-	-
	Chi-Square = 98.5853, p=0.0000			Chi-Square=65.8077, p=0.0000		

Discussion

Cardiovascular disease remains the main cause of morbidity and mortality in HD patients. The USRDS annual data from prevalent patients in the years 1998-2000, show that 75.47 (42.2%) of the 178.92 deaths per 1,000 patient years at risk have cardiovascular causes[1]. Although traditional risk factors are common in these patients, they may not be sufficient alone to account for the high prevalence of CVD in this condition. In uraemic patients, traditional risk factors are added to specific, disease-related and treatment-related risk factors.

Consequences of renal anaemia in the HD patients are well described. There is a close association between low Hb levels, LV dilatation, cardiac failure and mortality in this population. Given the prevalence of anaemia in the dialysis population, and its association with poor outcomes, anaemia is considered a "uremia-specific" CVD risk factor[5,6]. A correlation between survival in HD patients and Hb concentration has been established in a large number of retrospective and prospective studies, but the most appropriate Hb value for optimum cardiovascular risk reduction is still a matter of debate. Strippoli et al, in a meta-analysis of 15 randomized controlled trials, emphasizes that Hb levels <12g/dl are associated with a lower all-cause mortality than Hb>13 g/dl, whereas Hb levels <10g/dl are associated with increased risk of seizures[7]. The study of Besarab et al showed that, in

dialysis patients the CVD mortality in the group with high Hb was greater than in the group with standard Hb levels, but this between-group difference was not significant.[8] In a large DOPPS study higher Hb levels were seen to be associated with a decreased relative risk for mortality and hospitalization[9]. In the study presented here, patients who died had significantly lower serum levels of Hb ~ 93 g/l and in the Kaplan-Meier analysis both all cause and cardiovascular mortality rate was considerably higher in patients with Hb <110 g/l. The results of the Cox analysis show that low Hb levels was a significant predictor of all cause and cardiovascular mortality, suggesting that anaemia is one of the most important disease – related risk factors of cardiovascular mortality in HD patients.

Current literature suggests that abnormal mineral metabolism, particularly high serum phosphorus levels and CaxP product, are not only implicated in the pathogenesis of bone disease but also significantly contribute to the high rates of cardiovascular mortality among dialysis populations. In a large-scale study of 6047 hamodialysis patients, Block et al demonstrated that hyperphosphataemia is associated with an 18-39% higher risk of death compared with normal reference groups [10]. Recently, the data of the DOPPS (17.236 patients) and the USRDS waves 1,3 and 4 study (14.829 patients) suggest that not only hyperphosphataemia, but also higher calcium levels are associated with fatal and nonfatal cardiovascular events and all-cause mortality in hemodialysis patients [11,12]. Collectively, the data clearly indicate that cardiovascular calcifications of dialysis patients secondary to an increase in phosphate and calcium levels are predictors of excessive cardiovascular morbidity and mortality. In this study we found that only serum levels of calcium were significantly higher in patients who died, but there was no difference in the other parameters of calcium-phosphate disorders, as in serum phosphate, PTH and CaxP product. The shorter duration of follow-up period in this study and the small number of patients who died during this period, may explain why calcium-phosphate disorders parameters did not predicted either all cause or cardiovascular mortality.

Dialysis efficacy is one of the predominant factors determining survival in HD patients. Among treatment/dialysis-related cardiovascular risk factor we analyzed HD dose, using the index Kt/V, which is function of dialyser urea clearance, treatment time and urea distribution volume and is the most commonly used marker for dialysis adequacy. It once was believed that factors related to dialysis treatment were the main causes of poor clinical outcome, however, in a recent multicenter, randomized clinical trial known as the HEMO Study, death from any cause was not significantly influenced by the dose or flux assignment[13]. In our study, mean spKt/V and eKt/V were significantly lower in patients who died and in the Kaplan-Meier analysis both all cause and cardiovascular mortality rate was considerably higher in patients with spKtV <1.2, but in the Cox analysis spKt/V was a significant predictor only for all cause mortality.

Numerous studies have shown that hypoalbuminemia is strongly associated with mortality and CVD in HD patients[14,15]. Currently, although several approaches have been used to assess nutrition, albumin is probably still the most commonly used nutritional marker in HD patients. However, its value has been questioned because a low albumin level may reflect not only poor nutrition, but also reflect the presence of an inflammatory reaction. In our study, serum albumin was taken as nutritional marker and, all-cause and cardiovascular mortality was higher in patients with albumin levels lower than 40 g/L, but, in the multivariate Cox proportional hazards model albumin lost its significance as a risk factor for all-cause and cardiovascular mortality. Recent evidence demonstrates that inflammation, a non-traditional risk factor is the cause of both, malnutrition and atherosclerosis in HD patients. The prevalence of chronic inflammation is high in dialysis patients and it is associated with an increased mortality risk, yet the origin of chronic inflammation in dialysis patients remains unclear. CRP, a nonspecific marker of inflammation, is regarded as a fundamental biomarker for cardiovascular risk stratification in HD patients. Most studies with high applicability did show that inflammation, as reflected by elevated levels of CRP predicted all-cause and cardiovascular mortality in HD patients[16-19]. The results of the Cox analysis in this study show that even among the subgroup of patients who died of cardiovascular causes, high CRP and low Hb level remained a more powerful predictors of death than low albumin level and other investigated parameters. Unlike other studies[16], where fibrinogen is one of the positive acute-phase proteins, in this study, in the multivariate Cox analysis it failed to be a predictor of mortality in HD patients. In addition, there appears to be a complex association between inflammation and anaemia with cardiovascular mortality, because when only cardiovascular mortality was entered into the Cox model, high CRP and low Hb levels were the only significant predictors for cardiovascular mortality. Apart from uraemia, chronic inflammation is associated with anaemia as a result of the suppression of bone marrow erythropoiesis by some cytokines[20]. These finding suggest that the key to the cardiovascular mortality in dialysis patients is either the cause of the inflammation or the consequences of the inflammatory process. However, the possible interactions between inflammation and anaemia, as potentially modifiable risk factors of dialysis-associated cardiovascular mortality, are still unclear.

In the study presented here, high CRP and low Hb levels were identified as significant independent non-traditional, disease-related cardiovascular risk factors that predict cardiovascular mortality in HD patients. It can be concluded that among non-traditional risk factors, inflammation and anaemia, as uremia-related risk factors per se have an important effect on cardiovascular mortality in HD patients. Identification of these risk

factors may lead to new diagnostic and therapeutic approaches in order to improve long-term survival in HD patients.

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