Original Paper

Uremic Restless Legs Syndrome (RLS) and Sleep Quality in Patients With End-Stage Renal Disease on Hemodialysis: Potential Role of Homocysteine and Parathyroid Hormone

Katrin Gade[a]   Sabine Blaschke[b]   Andrea Rodenbeck[a,d]   Andreas Becker[c]
Heike Anderson-Schmidt[a]   Stefan Cohrs[d,a]

[a]Department of Psychiatry and Psychotherapy, Georg-August-University Göttingen; [b]Interdisciplinary Emergency Care Unit, Georg-August-University Göttingen; [c]Department of Child and Adolescent Psychiatry, Georg-August-University Göttingen, Göttingen; [d]Clinic of Sleep Medicine, St.Hedwig Krankenhaus, Berlin, Germany

Key Words
Uremic RLS • Homocysteine • Sleep Quality • Dialysis • Parathyroid hormone

Abstract
Background: The aetiology of uremic restless legs syndrome (RLS) remains unclear. Our research investigated whether an elevated plasma concentration of the excitatory amino acid homocysteine might be associated with RLS occurrence in patients with chronic renal insufficiency on hemodialysis. *Methods:* Total plasma homocysteine as well as creatinine, urea, folate, parathyroid hormone, hemoglobin, iron, ferritin, phosphate, calcium, magnesium, and albumin levels were compared between 26 RLS-affected (RLSpos) and 26 non-affected (RLSneg) patients on chronic hemodialysis. We further compared subjective sleep quality between RLSpos and RLSneg patients using the Pittsburgh-Sleep-Quality-Index and investigated possible relationships between laboratory parameters and sleep quality. *Results:* Taking individual albumin concentrations into account, a significant positive correlation between total plasma homocysteine and RLS occurrence was observed (r= 0.246; p=0.045). Sleep quality was significantly more reduced in RLSpos compared to RLSneg patients and RLS severity correlated positively with impairment of sleep quality. Bad sleep quality in all patients was associated with higher concentrations of parathyroid hormone. *Conclusion:* Our results suggest a possible aetiological role of homocysteine in uremic RLS. They confirm that uremic RLS is an important factor causing sleep impairment in patients on hemodialysis. Higher parathyroid hormone levels might also be associated with bad sleep quality in these patients.

Copyright © 2013 S. Karger AG, Basel

Katrin Gade

Department of Psychiatry and Psychotherapy,
Georg-August-University Göttingen, Von-Siebold-Str. 5
D-37075 Göttingen (Germany), Tel. +49-551-39 6610, Fax +49-551-39 9337
E-Mail katrin.gade@med.uni-goettingen.de

Introduction

The uremic form of the Restless Legs Syndrome (RLS) is one of the most important factors contributing to sleep disturbances in patients with chronic renal insufficiency on hemodialysis and affects up to 62% of these patients according to the literature [1]. Its pathophysiology and the role of potential aetiological factors are not fully understood yet. Uremic RLS develops with decreased renal function and often disappears after renal transplantation. Various changes in laboratory parameters, common in renal insufficiency, such as anaemia, decreased serum ferritin, low serum iron and changes in the body's calcium balance (including low and high serum calcium, phosphate, and parathyroid hormone levels) have been cited as potential causes of RLS in patients on dialysis. Several studies have been carried out in the past examining these parameters but unfortunately, the results have been inconsistent [2-4]. However, it has been postulated that metabolic substances accumulating in the blood of uremic patients may cause an imbalance between inhibitory and excitatory effects in the central nervous system (CNS). Our research investigated the hypothesis whether an elevated total plasma concentration of the excitatory amino acid homocysteine might be associated with the occurrence of uremic RLS in patients on hemodialysis. Furthermore, we compared several other laboratory parameters between RLS-affected and non-affected dialysis patients which have been suspected to play an aetiological role in idiopathic and uremic RLS according to previous studies. Subjective sleep quality was assessed in all patients and possible associations of laboratory parameters with sleep quality were also investigated.

Materials and Methods

52 patients with chronic renal insufficiency stage V receiving hemodialysis (3x/week for at least 6 months) were recruited at three different dialysis centres near Göttingen (Germany) between 2005 and 2009. For analytical purposes, patients were divided into two groups. The first group (RLSpos) consisted of 26 patients (8 males, 18 females, aged 34 to 87 years, mean age: 67 ± 13 years) suffering from RLS according to a diagnostic questionnaire which implemented the clinical RLS criteria as defined by the International Restless Legs Study Group (IRLSSG) [5], and a short clinical examination. The other 26 patients in the control group (RLSneg, 12 males, 14 females, aged 25 to 86 years, mean age: 67 ± 15 years) were free of RLS using the same criteria. RLSpos patients whose symptoms had started before the onset of dialysis or who had a family history of RLS were excluded from the study. In RLSpos patients, RLS-severity was measured using the International-RLS-severity-scale (IRLS) [6]. Subjective sleep quality was assessed in all patients using the German version of the Pittsburgh Sleep Quality-Index (PSQI) [7]. Blood samples were taken in the morning after an overnight fast and after two days without hemodialysis (hemodialysis-free interval: 66h). Plasma (P)/serum (S) levels of creatinine, urea, albumin, calcium, magnesium, phosphate, parathyroid hormone, hemoglobin, iron, ferritin and folate were analysed within two hours after blood draw at the Department of Clinical Chemistry, University of Göttingen, according to standard methods. Total plasma homocysteine, defined as the sum of all homocysteine subfractions including free and protein-bound forms in plasma, was determined by fluorescence polarisation immunoassay. The study protocol was approved by the local ethics committee. In accordance with legal requirements, informed written consent was obtained from all patients before the study commenced.

Statistical analyses

For all statistical tests, a significance level of $p \leq .05$ was used, unless stated otherwise. Since the laboratory parameters of the two groups (RLSpos vs. RLSneg) were normally distributed, parametric tests were used for subsequent analyses. The means including standard error of the mean (SEM) were calculated and plotted for each laboratory parameter. Independent one-tailed t-tests were used to analyse group differences. Hypothesising that higher homocysteine levels would be positively correlated with membership in the RLSpos group, a one-directional Pearson's correlation was used to test this assumption. A partial correlation was run to take into account the possible influence of albumin on total plasma homocysteine,

Kidney Blood Press Res 2013;37:458-463

DOI: 10.1159/000355727
Published online: October 25, 2013

© 2013 S. Karger AG, Basel
www.karger.com/kbr

460

Gade/Blaschke/Rodenbeck/Becker/Anderson-Schmidt/Cohrs: Homocysteine in Uremic
RLS on Hemodialysis

since albumin has been reported to be directly correlated to homocysteine levels [8]. Pearson's correlations were carried out to examine the relationships between homocysteine concentration and folate levels, age and gender.

The means of PSQI-sumscores and PSQI-subitem-scores including SEMs were compared between RLSpos and RLSneg patients using ANOVAs. Participants were additionally categorised into two groups based on their PSQI-sumscores, i.e., „good sleepers", versus „bad sleepers". The means and standard errors of lab parameters were also compared between these two groups. Pearson's correlations were used to examine the potential relationships between IRLS-Scores and PSQI-total and subitem-scores, between lab parameters and IRLS-scores in RLSpos patients only, and between lab parameters and PSQI-total-score in all patients.

Results

While all patients showed some degree of hyperhomocysteinaemia, we observed a strong trend towards higher homocysteine levels in the RLSpos group compared to the RLSneg group (p=0.053). A significant positive partial correlation was observed between total plasma homocysteine and RLS occurrence (r= 0.246; p=0.045), taking into account individual albumin concentration levels. No significant correlation was observed between age (r=0.01, p=0.944) or gender (r=0.041, p=0.774) and total plasma homocysteine. However, homocysteine correlated inversely with serum folate concentration in all patients (r=-0.32, p=0.03). Means of folate, albumin, hemoglobin, urea, creatinine, ferritin, iron, parathyroid hormone, magnesium, phosphate and calcium levels did not differ significantly between RLSpos and RLSneg patients (for details see Table1).

Table 1. Laboratory parameters in RLS pos and RLS neg patients

		N*	MEAN incl SEM	Min/Max	p	T
P-Homocysteine	RLSneg	26	32.94± 2.43	16.70/70.90	p=0.053	-1.652
(Ref.: 5-15µmol/l)	RLSpos	26	40.49± 3.87	19.20/101.20		
P-Urea	RLSneg	26	54.42± 3.25	25.00/91.00	p=0.347	-0.397
(Ref.: 8-21mg/dl)	RLSpos	26	56.19± 3.06	30.00/87.00		
P-Creatinine	RLSneg	26	7.96± 0.75	2.10/16.10	p=0.205	0.833
(Ref.: 0.6-1.2mg/dl)	RLSpos	26	7.19± 0.53	2.30/13.20		
Hemoglobin	RLSneg	26	11.59± 0.22	9.70/13.60	p=0.224	-0.767
(Ref.:11.5-17.5g/dl)	RLSpos	26	11.85± 0.25	8.80/14.40		
S-Ferritin	RLSneg	26	519.15± 75.73	23.00/1366.00	p=0.362	0.357
(Ref.: 10-250µg/l)	RLSpos	25	484.88± 58.10	72.00/1008.00		
P-Iron	RLSneg	26	11.49± 1.12	4.40/28.60	p=0.387	-0.290
(Ref.: 50-175µg/l)	RLSpos	26	11.95± 1.16	4.50/36.00		
S-Parathyroid hormone	RLSneg	26	231.04± 32.48	0.00/818.00	p=0.345	-0.401
(Ref.: 11-67pg/ml)	RLSpos	23	253.04± 45.38	0.00/837.00		
P-Magnesium	RLSneg	25	0.93± 0.03	0.51/1.24	p=0.466	0.087
(Ref.: 0.75-1.05mmol/l)	RLSpos	22	0.93± 0.04	0.51/1.25		
P-Phosphate	RLSneg	26	1.64± 0.08	0.91/2.44	p=0.14	-1.095
(Ref.:0.7-1.4mmol/l)	RLSpos	26	1.80± 0.12	0.91/3.75		
P-Calcium	RLSneg	26	2.27± 0.04	1.75/2.58	p=0.45	0.127
(Ref.:2.2-2.6mmol/l)	RLSpos	26	2.26± 0.05	1.60/2.78		
P-Albumin	RLSneg	26	3.93 ± 0.06	3.20/4.50	p=0.678	0.417
(Ref.: 3.4-4.8 g/dl)	RLSpos	26	3.89 ± 0.07	3.30/4.40		
S-Folate	RLSneg	26	8.73 ± 1.00	3.20/4.50	p=0.418	- 0.656
(Ref.: 4-17 µg/l)	RLSpos	20	9.11 ± 1.62	3.30/4.40		

* N= number of patients included in this analysis

Impaired subjective sleep quality was found in 37 (71%) out of all 52 patients (i.e., PSQI-sumscore ≤ 5 points). Mean subjective sleep quality was significantly lower in the RLSpos group than in the RLSneg group, resulting in a higher PSQI-sumscore (for details including PSQI-subitem-scores see Figure 1). There was a significant positive correlation between RLS-severity and PSQI-total-score (r= 0.510; p= 0.004) in RLSpos patients. No significant correlation could be observed between laboratory parameters and RLS-severity in RLSpos patients or between laboratory parameters and PSQI-sumscore in both RLSpos and RLSneg patients. However, when patients were split according to their sleep quality using the PSQI`s cut-off value, i.e. "bad" vs. "good "sleepers, the "bad" sleepers in both RLSpos and RLSneg groups showed significantly higher mean serum levels of parathyroid hormone than the "good" sleepers (280.53± 35,51 vs. 152.60± 26.60, p=0.028).
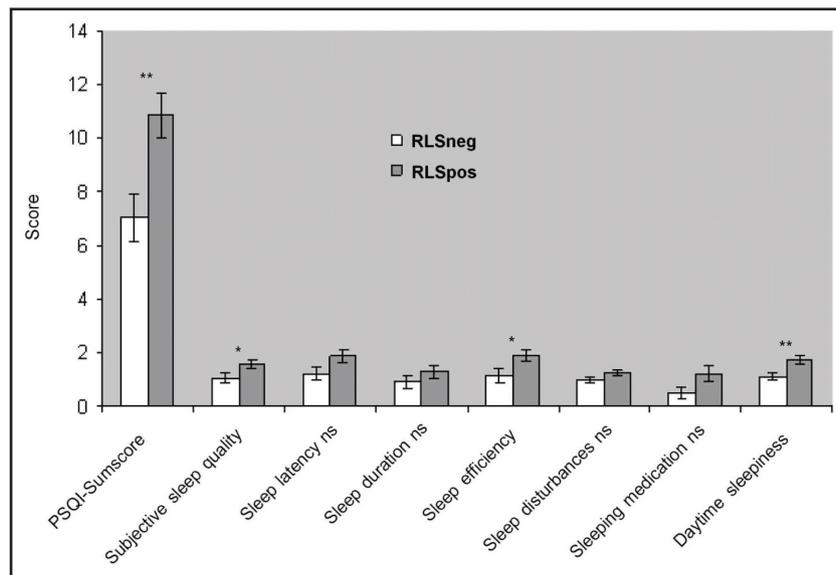
Fig. 1. Sleep quality in RLSpos and RLSneg patients. Means ± SEM of PSQI-Sumscore and Subitem-Scores in RLSpos and RLSneg patients.
**significant at a p-value of .01;
* significant at a p-value of .05;
ns = not significant.

Discussion

As expected, subjective sleep quality was significantly more impaired in patients with RLS compared to patients without RLS, and sleep quality was found to deteriorate with increasing severity of RLS symptoms. Furthermore, bad sleep quality was associated with higher levels of parathyroid hormone in both RLS-affected and non-affected patients. Hyperparathyroidism and sleep problems in patients on dialysis appear to be connected and have previously been reported in other studies [9, 10]; hyper-and hypoparathyroidism have also been associated with RLS in some cases [11]. However, in our study, mean concentrations of parathyroid hormone did not differ significantly between RLS-affected and non-affected patients. There were also no differences between the two groups with regard to other laboratory parameters previously described as risk factors for RLS (hemoglobin, ferritin, iron, calcium, phosphate). Our findings rather suggest an association between hyperhomocysteinaemia and uremic RLS since mean levels of total plasma homocysteine were significantly higher in RLS-affected patients. These results are preliminary and further replication studies are needed to draw any firm conclusions. However, homocysteine may possibly play a causal role in uremic but

Kidney Blood Press Res 2013;37:458-463

DOI: 10.1159/000355727
Published online: October 25, 2013

© 2013 S. Karger AG, Basel
www.karger.com/kbr

462

Gade/Blaschke/Rodenbeck/Becker/Anderson-Schmidt/Cohrs: Homocysteine in Uremic
RLS on Hemodialysis

not in idiopathic RLS. For instance, Bachmann et al. [12] found no significant differences in total plasma homocysteine between idiopathic RLS patients and healthy controls or between RLS groups with and without treatment with levodopa - a drug that can potentially elevate homocysteine levels, especially when given at higher dosages. This is of special importance for our study as we can assume that the low-dose levodopa medication received by most of our uremic RLS patients did not significantly affect their homocysteine levels. In over 80% of patients on dialysis, total plasma homocysteine is at least mildly elevated, most likely due to insufficient degradation. While homocysteine itself gets dialysed only to a small extent, its concentrations decrease up to 30% during a dialysis session due to removal of uremic toxins blocking its metabolic pathways [13]. There are two possible ways how homocysteine could cause or worsen RLS symptoms. It appears to act as an allosteric antagonist at the dopamine D2 receptor subtype [14], and could therefore lead to a functional dominance of D1 receptors within the dopaminergic system. This D2/D1 shift has already been suspected to play a major role in iron deficiency-related RLS and augmentation of RLS-symptoms while undergoing treatment with levodopa [15]. Furthermore, homocysteine is known to be an agonist at the NMDA-receptor and to increase CNS-excitability [16]. Based on previous research, which found an underlying CNS-hyperexcitability in RLS patients [17], hyperhomocysteinaemia could therefore lead to uremic RLS by changing the balance between excitatory and inhibitory influences. There may be no exact threshold, beyond which this effect is expected to occur, as additional metabolic factors most likely also influence this balance in individual patients. Aside from the usual pharmacological treatment, a pragmatic therapeutic approach for RLS patients on chronic hemodialysis should be to optimise all potential RLS risk factors that are present. Most importantly, dialysis dose and quality should be adjusted to optimal levels in RLS patients. However, it could be worthwhile investigating the homocysteine hypothesis, for example, by studying the effects of administering folate in uremic RLS patients with severe hyperhomocysteinaemia. Supraphysiological doses of folate can enhance the activity of the homocysteine remethylation pathway, thereby lowering excess homocysteine in patients with chronic renal insufficiency [18]. As promising therapeutic effects have already been demonstrated in pregnant women with RLS [19], symptoms of uremic RLS are expected to be ameliorated by folate substitution.

Conclusion

The results of our study suggest that homocysteine may play an aetiological role in uremic RLS. In order to test and verify these assumptions further investigations including larger sample sizes need to be carried out. Furthermore, our study confirms that uremic RLS is an important cause of sleep impairment in patients on hemodialysis. However, higher levels of parathyroid hormone might also be associated with bad sleep quality in these patients.

Conflict of Interests

The authors declare that they have no relevant material or financial interests that relate to the research described in this paper.

Acknowledgements

References

1   Kavanagh D, Siddiqui S, Geddes CC: Restless legs syndrome in patients on dialysis. Am J Kidney Dis 2004;43:763-771.

2   Collado-Seidel V, Kohnen R, Samtleben W, Hillebrand GF, Oertel WH, Trenkwalder C: Clinical and biochemical findings in uremic patients with and without restless legs syndrome. Am J Kidney Dis 1998;31:324-328.

3   Siddiqui S, Kavanagh D, Traynor J, Mak M, Deighan C, Geddes C: Risk factors for restless legs syndrome in dialysis patients. Nephron Clin Pract 2005;101:155-160.

4   Kawauchi A, Inoue Y, Hashimoto T, Tachibana N, Shirakawa S, Mizutani Y, Ono T, Miki T: Restless legs syndrome in hemodialysis patients: health-related quality of life and laboratory data analysis. Clin Nephrol 2006;66:440-446.

5   Walters AS, The International Restless Legs Syndrome Study Group: Toward a better definition of the restless legs syndrome. Mov Disord 1995;10:634-642.

6   Walters AS, Le Brocq C, Dhar A, Hening W, Rosen R, Allen RP, Trenkwalder C: Validation of the International Restless Legs Syndrome Study Group rating scale for restless legs syndrome. Sleep Med 2003;4:121-132.

7   Riemann D, Backhaus J: Behandlung von Schlafstörungen. Psychologie Verlags Union; Weinheim 1996, pp. 33-56.

8   Walker MC, Smith GN, Perkins SL, Keely EJ, Garner PR: Changes in homocysteine levels during normal pregnancy. Am J Obstet Gynecol 1999;180:660-664.

9   Sabbatini M, Minale B, Crispo A, Pisani A, Ragosta A, Esposito R, Cesaro A, Cianciaruso B, Andreucci VE: Insomnia in maintenance haemodialysis patients. Nephrol Dial Transplant 2002;17:852-856.

10 Esposito MG, Cesare CM, De Santo RM, Cice G, Perna AF, Violetti E, Conzo G, Bilancio G, Celsi S, Annunziata F, Ianelli S, De Santo NG, Cirillo M, Livrea A: Parathyroidectomy improves the quality of sleep in maintenance hemodialysis patients with severe hyperparathyroidism. J Nephrol 2008;21:S92-S96.

11 Lim LL, Dinner D, Tham KW, Siraj E, Shields R: Restless legs syndrome associated with primary hyperparathyroidism. Sleep Med 2005;6:283-285.

12 Bachmann CG, Guth N, Helmschmied K, Armstrong VW, Paulus W, Happe S: Homocysteine in restless legs syndrome. Sleep Med 2008;9:388-392.

13 Arnadottir M, Berg AL, Hegbrant J, Hultberg B: Influence of haemodialysis on plasma total homocysteine concentration. Nephrol Dial Transplant 1999;14:142-146.

14 Agnati LF, Ferre S, Genedani S, Leo G, Guidolin D, Filaferro M, Carriba P, Casado V, Lluis C, Franco R, Woods AS, Fuxe K: Allosteric modulation of dopamine D2 receptors by homocysteine. J Proteome Res 2006;5:3077-3083.

15 Paulus W, Trenkwalder C: Less is more: pathophysiology of dopaminergic-therapy-related augmentation in restless legs syndrome. Lancet Neurol 2006;5:878-886.

16 Wuerthele SE, Yasuda RP, Freed Freed WJ, Hoffer BJ: The effect of local application of homocysteine on neuronal activity in the central nervous system of the rat. Life Sci 1982;31:2683-2691.

17 Tergau F, Wischer S, Paulus W: Motor system excitability in patients with restless legs syndrome. Neurology 1999;52:1060-1063.

18 De Vriese AS, Verbeke F, Schrijvers BF, Lameire NH: Is folate a promising agent in the prevention and treatment of cardiovascular disease in patients with renal failure? Kidney Int 2002;61:1199-1209.

19 Botez MI, Lambert B: Folate deficiency and restless-legs syndrome in pregnancy. N Engl J Med 1977;297:670.