

Journal of Nephrothology



The relationship between serum ghrelin levels and bone metabolism markers and severity anemia in non-diabetic hemodialysis patients; a pilot study

Leila Mahmoodnia¹, Sara Beigrezaei², Mohammad Reza Tamadon³, Tina Jafari⁴, Marzieh Kafeshani⁵, Fatemeh Mahmoodi¹, Ali Hasanpour Dehkordi⁶, Masoud Amiri^{7*}

¹Department of Internal Medicine, Shahrekord University of Medical Sciences, Shahrekord, Iran

²School of Nutrition and Food Science, Isfahan University of Medical Sciences, Isfahan, Iran

³Department of Internal Medicine, Semnan University of Medical Sciences, Semnan, Iran

⁴Department of Biochemistry and Nutrition, Faculty of Medicine, Shahrekord University of Medical Sciences, Shahrekord

⁵Food Security Research Center and Department of Clinical Nutrition/Community Nutrition/Food Science and Technology, School of Nutrition and Food Science, Isfahan University of Medical Sciences, Isfahan, Iran

⁶Social determinants of Health Research Center/Department of Medical-Surgical, Faculty of Nursing and Midwifery, Shahrekord University of Medical Sciences, Shahrekord, Iran

⁷Department of Epidemiology and Biostatistics, School of Health, Shahrekord University of Medical Sciences, Shahrekord, Iran

ARTICLE INFO

Article type:
Original Article

Article history:
Received: 2 June 2017
Accepted: 19 December 2017
Published online: 7 January 2018
DOI: 10.15171/jnp.2018.10

Keywords:
Ghrelin
Bone metabolism
Inflammation
Anemia
Hemodialysis

ABSTRACT

Background: Chronic kidney disease (CKD) is a consequence of progressive and irreversible destruction of nephrons, mainly due to uncontrolled diabetes mellitus and high blood pressure. Ghrelin is a peptide hormone which could play a substantial role in hunger sensation, may increase body fat percentage and might adjust the long-term body weight and is mostly secreted in the stomach.

Objectives: This study aimed to evaluate the relationship between ghrelin levels and various biochemical and demographic indices in a group of non-diabetic hemodialysis patients.

Patients and Methods: Around 39 non-diabetic patients undergoing hemodialysis in Hajar hospital at Shahrekord city were enrolled.

Results: The mean age of patients was 57.10 ± 20.20 years. Their mean weight was 56.65 ± 16.25 kg. Their hemoglobin level mean was 10.43 ± 1.84 g/dL. The serum ghrelin level had a positive but not significant correlation with the age, dialysis quality, dosage, calcium level, alkaline phosphatase (ALP), parathormone, vitamin D and hemoglobin in these patients.

Conclusions: More studies in this subject of hemodialysis patients to define the exact role of ghrelin in hemodialysis patients are recommended.

Implication for health policy/practice/research/medical education:

In a study on 39 non-diabetic patients undergoing hemodialysis, we found that serum ghrelin level had a positive but not significant correlation with the age, dialysis quality, dosage, calcium level, alkaline phosphatase (ALP), parathormone, vitamin D and hemoglobin in these patients.

Please cite this paper as: Mahmoodnia L, Beigrezaei S, Tamadon MR, Jafari T, Kafeshani M, Mahmoodi F, et al. The relationship between serum ghrelin levels and bone metabolism markers and severity anemia in non-diabetic hemodialysis patients; a pilot study. J Nephrothol. 2018;7(1):32-36. DOI: 10.15171/jnp.2017.10.

1. Background

Chronic kidney disease (CKD) is a consequence of progressive and irreversible destruction of nephrons, mainly due to uncontrolled diabetes mellitus and high blood pressure (1). The prevalence rate of this disease is

increasing worldwide about 8% annually (2,3). In addition, end-stage renal disease (ESRD) would be the final stage of CKD, when the patient needs an alternative method such as dialysis. Moreover, the annual mortality rate of dialysis patients is around 15%-20% and their 5-year survival rate

*Corresponding author: Masoud Amiri Ph.D, Email: masoud.amiri@yahoo.com

is 30%-35% (1). However, cardiovascular and infectious diseases could be the chief causes of death among these patients (1,4). Therefore, the identification and prevention of these diseases and potential increase of life expectancy of dialysis patients are very important.

Ghrelin is a peptide hormone which could be playing a substantial role in hunger sensation, increasing body fat percentage and adjusting the long-term body weight and is mostly secreted in the stomach (5-7). Additionally, other parts of gastrointestinal tract, especially the small intestine, pancreas epsilon cells, kidney glomerular cells, hypothalamus neurons and hypophysis are also able to secrete this hormone (8). The importance of ghrelin in CKD is notable from many aspects. For example, the role of ghrelin in energy balance and reduction of insulin resistance has been widely recognized (9-11). Thus, measuring blood ghrelin level and determining its relationship with other important factors provide some valuable information about the role of ghrelin in the pathophysiology of various diseases such as type 2 diabetes, which is the main cause of chronic kidney failure (9). Moreover, with the decrease of kidney function, the ghrelin serum level may increase, up to 2-3 times higher in patients suffering from stage five of chronic kidney failure compared to healthy individuals emphasizing the important role of kidneys in the destruction of ghrelin (12-14). The high blood levels of ghrelin could entail the increase in the micro- and macro-vascular complications which may lead to ESRD (14). Chang et al demonstrated that high ghrelin levels could be an important risk factor for cardiovascular disease (15). Therefore, the increase in ghrelin levels in dialysis patients may contribute to various complications and increasing mortality rates in this group. Ayala et al reported a significant relationship between the baseline serum levels of ghrelin and serum insulin levels, body mass index (BMI), abdominal fat and serum leptin levels (16). Furthermore, Isguven et al studied ghrelin levels in children suffering from anemia and reported that ghrelin level was significantly lower than normal individuals (17). In another study on menopausal women, the serum ghrelin levels were found to be lower than non-menopausal healthy individuals with a significant relationship between serum ghrelin and bone density (18).

2. Objectives

Despite numerous studies supporting the notion of increased serum levels of ghrelin in hemodialysis patients, there is not enough information regarding the relationship between ghrelin and various metabolic factors such as vitamin D or inflammatory factors like C-reactive protein (CRP) and anemia. The aim of this study was

the evaluation of the relationship between serum ghrelin levels with various biochemical and demographic indices in a group of non-diabetic hemodialysis patients.

3. Patients and Methods

3.1. Study population

In this cross-sectional study, 40 hemodialysis patients of Hajar hospital at Shahrekord were studied in 2015. As one of the patients withdrew from the study, 39 patients were studied. The study consisted of non-diabetic patients undergoing hemodialysis.

3.2. Laboratory assessments

A fasting time of about 8 hours, venous blood samples were obtained for biochemical measurements including ghrelin, calcium, phosphorus, vitamin D, alkaline phosphatase (ALP), and intact parathyroid hormone (PTH), C-reactive protein (CRP) and also hemoglobin levels. Ghrelin was measured by Demeditec Diagnostics kit (Germany) with ELISA method (7).

3.3. Demographic information

To obtain these data, we used a questionnaire including demographic information such as age, gender, the duration of dialysis and BMI. BMI can be calculated by dividing the weight in kilograms to the square root of the height in meters. To calculate the dialysis adequacy we applied urea reduction ratio (URR).

3.4. Ethical issues

1) The research followed the tenets of the Declaration of Helsinki; 2) informed consent was obtained; and 3) This study was approved by the Ethics Committee of Shahrekord University of Medical Sciences (ethical code# IRskums.Rec.1394.272).

3.5. Statistical analysis

For analysis, we applied SPSS software version 20. For correlation, Spearman's test was used and *P* values of less than 0.05 were regarded as statistically significant.

4. Results

The mean age of the patients was 57.10 ± 20.20 years. Their mean weight was 56.65 ± 16.25 kg. Their hemoglobin level mean was 10.43 ± 1.84 g/dL.

Table 1 shows the results of biochemical, clinical and anthropometrical measurements of patients. The mean ghrelin level of non-diabetic hemodialysis patients was 5.62 ± 4.61 ng/mL (median; 3.25 ng/mL).

In addition, we found that the serum ghrelin level had a positive correlation but not significant with the age,

Table 1. Biochemical, clinical and anthropometrical data of non-diabetic hemodialysis patients (n = 39)

Factor	Median	Mean± SD
Age (y)	62	57.10 ± 20.20
Weight (kg)	53.25	56.65 ± 16.25
Calcium, mg/dL	9.1	9.15± 1.18
Phosphorus, mg/dL	6.0	5.96± 0.85
Hemoglobin, g/dL	10.55	10.43± 1.84
URR, %	71	72.30± 6.53
ALP, IU/mL	502.5	528.53± 283.81
Intact PTH, pg/mL	637.95	807.03± 686.86
Vitamin D, ng/mL	20.25	22.64± 11.32
Ghrelin, ng/mL	3.25	5.62± 4.61
Dialysis dosage	960	2000.00± 2249.77
Dialysis duration, months	24	44.7± 50.92

Abbreviations: URR, urea reduction ratio; ALP, alkaline phosphatase; PTH, Parathyroid hormone.

dialysis quality, dosage, calcium level, ALP, parathormone, vitamin D and hemoglobin in these patients. It should be noted that there was no significant association between serum ghrelin level and other factors ($P > 0.05$). Ghrelin serum level was negatively correlated with phosphorus level, but the correlation was not significant too ($P > 0.05$) (Table 2).

Ghrelin level was not significantly correlated with gender (27 men and 13 women), CRP and vitamin D in patients ($P > 0.05$) (Table 3).

5. Discussion

Kidneys play an important role in the biodegradation and removing the ghrelin in blood circulation (19). In fact, when the function of kidneys is decreasing, the serum ghrelin level will increase, which reflects the critical role of kidneys in biodegradation of ghrelin (12-14). In addition, due to the raised ghrelin levels in patients suffering from kidney failure, and the negative impacts of high ghrelin levels on the dialysis prognosis and their quality of life, investigating the factors impacting on the association of ghrelin levels and these problems seems to be necessary in order to find a way to reduce ghrelin levels and consequently reducing the dialysis complications and costs. This study was conducted to investigate the relationship between serum ghrelin level and bone

metabolism factors, inflammation, and anemia severity in non-diabetic hemodialysis patients.

Our study showed that serum ghrelin levels had a positive but not significant correlation with the age, dialysis quality, dosage, calcium level, ALP, parathormone, vitamin D and hemoglobin in non-diabetic hemodialysis patients. The correlation between ghrelin level and dialysis duration was positive but not significant. The serum ghrelin level was shown to be negatively correlated with phosphate level, however it was not significant as well as CRP and gender. Moreover, studies have shown that ghrelin levels in hemodialysis patients are significantly higher than healthy individuals. For example, Pérez-Fontán et al reported higher ghrelin plasma levels in patients receiving hemodialysis and peritoneal dialysis compared to the control group (13). Chang et al have also demonstrated that ghrelin levels are higher in ESRD patients compared to the control group (15). In addition, Firczyk et al found that the average ghrelin plasma level in healthy individuals, the hemodialysis group with diabetic nephropathy and hemodialysis patients without diabetic nephropathy were 4.33, 6.47, and 5.78 ng/mL, respectively, which is significantly higher than that of the healthy control group (19). In our study, the mean ghrelin level of non-diabetic hemodialysis patients was 5.62 ± 4.61 ng/mL which is similar to their study.

Izadi et al also reported that there was a direct linear relationship between serum ghrelin levels and the BMI of patients, but this correlation was not statistically significant (9). Our study showed the positive but not significant correlation between ghrelin level and age of non-diabetic hemodialysis as well. On the contrary, Katsuki et al observed lower levels of plasma ghrelin in obese patients suffering from diabetes mellitus type 2 compared to non-obese patients and argued for a negative significant correlation between ghrelin serum levels and BMI (20). In addition, in the study of Farajallah et al, lower ghrelin levels were observed in diabetic insulin resistant patients (21) which might be accounted for the negative correlation between the ghrelin levels and insulin levels of these patients.

In this study, the duration of dialysis was 44.7 ± 50.92 months. There was a positive but not significant association

Table 2. Correlation between the ghrelin level and the studied factors in non-diabetic hemodialysis factors

Ghrelin	Age (y)	URR (%)	Duration of dialysis* (mon)	Dialysis dosage	Calcium (mg/dL)	Phosphorus (mg/dL)	Hemoglobin (g/dL)	ALP (IU/mL)	PTH (pg/mL)	Vitamin D (ng/mL)	
Ghrelin	1	0.19	0.25	0.14*	0.15	-0.67	-0.009	0.02	-0.2	-0.019	0.09
P value	-	0.23	0.11	0.38	0.34	0.68	0.96	0.90	0.9	0.91	0.58

*The dialysis duration from the beginning.

Table 3. The correlations between ghrelin level and gender, serum CRP and vitamin D levels in non-diabetic hemodialysis patients

	Gender	CRP	Vitamin D level
Ghrelin	Male (n = 27)	1	1
	Female (n = 13)	2	2
P value	>0.05	>0.05	>0.05

between the serum ghrelin levels and duration of dialysis in non-diabetic patients. In the study of Firczyk et al, the average duration of hemodialysis in the hemodialysis with diabetic nephropathy group was 28 months and in the hemodialysis patients without nephropathy was 25 months (19).

Furthermore, in the study by Isguven et al in 2007, among the children suffering from iron deficiency anemia, the ghrelin levels were significantly lower than the control group, but no clear association between the ghrelin levels and their blood parameters was found (17). In our study, the ghrelin levels of non-diabetic hemodialysis patients showed a positive and not significant correlation with their hemoglobin levels.

Similar to our study, Izadi et al also observed a positive but not significant correlation between ghrelin and hemoglobin level (9).

In the recent decades, the probability of vitamin D deficiency in type 2 diabetic patients and the serum levels of 25-hydroxy vitamin D was considered as the predictive factors of long-term complications of diabetes, such as cardiovascular diseases. For instance, Rahimi et al found, among diabetic patients, 82.2% of them suffered from vitamin D deficiency compared to 61.1% in healthy individuals. They have also reported lower levels of vitamin D in overweight individuals compared to those who were normal weight and healthy (22). In our study, the vitamin D level in non-diabetic hemodialysis patients was 22.64 ± 11.32 ng/mL and their mean phosphorus level was 5.96 ± 0.85 mg/dl. We found a negative not significant correlation between serum ghrelin and phosphorus levels in these patients.

Moreover, Rahimi et al demonstrated the mean ALP in diabetic patients to be 195.2 ± 54.5 IU/ml and in non-diabetics to be 181.9 ± 55.7 IU/ml (22), but in our study the mean level of ALP for non-diabetic hemodialysis patients was 528.53 ± 283.81 IU/mL. We may account these differences between various studies into the difference in the sample sizes. Therefore, the comparison of the findings of studies with a bigger population and more dispersion among the population might result in better and more reliable results.

In the studies carried out on chronic obstructive pulmonary diseases (COPD) patients, a negative correlation between

the ghrelin levels and CRP levels was observed (23). However, Barros et al reported that the serum ghrelin levels were significantly higher in hemodialysis patients, but no significant correlation was found between the inflammatory and ghrelin factors (24). In addition, in a study on menopause women, no significant association between the ghrelin levels and ALP was observed (18,25).

6. Conclusions

In this study, serum ghrelin level had a positive but not significant correlation with the age, dialysis quality, dosage, calcium level, ALP, parathormone, vitamin D and hemoglobin levels in non-diabetic hemodialysis patients. More studies in this area are recommended.

Limitations of the study

One of the limitations of this study was its small sample size. Thus, in order to increase the replicability of this study, we suggest further studies with larger sample sizes. We also recommend that other studies be done with the aim of determining serum ghrelin level and its association with different blood and biochemical factors in various populations, based on their gender, age groups as well as investigating the association between ghrelin serum level with blood fat levels in non-diabetic hemodialysis patients and comparing them with those of the healthy population.

Acknowledgements

This study was extracted from Medical Doctorate (MD) thesis of Fatemeh Mahmoodi in Shahrekord University of Medical Sciences.

Authors' contribution

FM, AHD and LM conceived the study and contributed reagents and tools. FM and LM performed the experiments. SBR, MA, TJ, MRT, and MK analyzed the data and drafted the final manuscript. All authors read, revised, and approved the final manuscript.

Conflicts of interest

There were no points of conflicts.

Funding/Support

This project funded by research deputy of Shahrekord University of Medical Sciences.

References

1. Malani PN. Harrison's Principles of Internal Medicine. JAMA. 2012;308(17):1813-4. doi: 10.1001/jama.308.17.1813-b.
2. Jafari F, Hashemi N, Reisi M. The effect of diet training

- on variations in blood pressure, weight, and some biochemical factors in hemodialysis patients: a clinical trial. *J Clin Nurs Midwif*. 2015;3(4):13-9.
3. Din-Mohammadi M, Por-Meamari M. The evaluation of some nutritional parameters of patients undergoing hemodialysis. *J Zanjan Univ Med Sci Health Serv*. 2002;10:41-5.
 4. Wen Y, Guo Q, Yang X, Wu X, Feng S, Tan J, et al. High glucose concentrations in peritoneal dialysate are associated with all-cause and cardiovascular disease mortality in continuous ambulatory peritoneal dialysis patients. *Perit Dial Int*. 2015;35(1):70-7. doi: 10.3747/pdi.2013.00083
 5. Yada T, Dezaki K, Sone H, Koizumi M, Damdindorj B, Nakata M, et al. Ghrelin regulates insulin release and glycemia: physiological role and therapeutic potential. *Curr Diabetes Rev*. 2008;4(1):18-23.
 6. Cummings DE, Purnell JQ, Frayo RS, Schmidova K, Wisse BE, Weigle DS. A preprandial rise in plasma ghrelin levels suggests a role in meal initiation in humans. *Diabetes*. 2001;50(8):1714-9.
 7. Tschöp M, Wawarta R, Riepl R, Friedrich S, Bidlingmaier M, Landgraf R, et al. Post-prandial decrease of circulating human ghrelin levels. *J Endocrinol Invest*. 2001;24(6):RC19-21.
 8. Izadi M, Soheli S, Khorshidi D, Doali H. The Relationship between Serum Ghrelin with Beta-Cell Function Index and Insulin Resistance in People with Type 2 Diabetes. *J Mazandaran Univ Med Sci*. 2011;21(82):18-25.
 9. Eizadi M, Zarifyan A, Sohaily S, Khorshidi D, Doaly H. Relation between serum ghrelin concentration and blood glucose levels in type-2 diabetic obese males. *J Gorgan Uni Med Sci*. 2012;14(1):83-9.
 10. Rodriguez A, Gomez-Ambrosi J, Catalán V, Gil M, Becerril S, Sainz N, et al. Acylated and desacyl ghrelin stimulate lipid accumulation in human visceral adipocytes. *Int J Obesity*. 2009;33(5):541-52.
 11. Kola B, Grossman A, Korbonits M. The role of AMP-activated protein kinase in obesity. *Front Horm Res*. 2008;36:198-211. doi: 10.1159/000115366.
 12. Yoshimoto A, Mori K, Sugawara A, Mukoyama M, Yahata K, Suganami T, et al. Plasma ghrelin and desacyl ghrelin concentrations in renal failure. *J Am Soc Nephrol*. 2002;13(11):2748-52.
 13. Pérez-Fontán M, Cordido F, Rodríguez-Carmona A, Peteiro J, García-Naveiro R, García-Buela J. Plasma ghrelin levels in patients undergoing hemodialysis and peritoneal dialysis. *Nephrol Dial Transplant*. 2004; 19 (8): 2095-2100. doi: 10.1093/ndt/gfh313
 14. Iglesias P, Díez JJ, Fernández-Reyes MJ, Codoceo R, Alvarez-Fidalgo P, Bajo MA, et al. Serum ghrelin concentrations in patients with chronic renal failure undergoing dialysis. *Clin Endocrinol (Oxf)*. 2006;64(1):68-73.
 15. Chang C-C, Hung C-H, Yen C-S, Hwang K-L, Lin C-Y. The relationship of plasma Ghrelin level to energy regulation, feeding and left ventricular function in non-diabetic haemodialysis patients. *Nephrol Dial Transplant*. 2005;20(10):2172-7. doi: 10.1093/ndt/gfi011.
 16. Ayala ER, Pecoits-Filho R, Heimbürger O, Lindholm B, Nordfors L, Stenvinkel P. Associations between plasma ghrelin levels and body composition in end-stage renal disease: a longitudinal study. *Nephrol Dial Transplant*. 2004;19(2):421-426. doi: 10.1093/ndt/gfg559.
 17. Isguven P, Arslanoglu I, Erol M, Yildiz M, Adal E, Erguven M. Serum levels of ghrelin, leptin, IGF-I, IGFBP-3, insulin, thyroid hormones and cortisol in prepubertal children with iron deficiency. *Endocr J*. 2007;54(6):985-90.
 18. Barmaki H, Kazerouni F, Rahimpour A, Amirrasouli H, Asgharzadeh AA, Talebi A. An investigation of serum ghrelin concentration and its relationship with bone mineral density in healthy postmenopausal women over 55 Years in Tabriz in 2013. *Environ Biol*. 2014;4(6):172-7.
 19. Firczyk P, Ignacy W, Adamczak M, Chudek J, Wiecek A. Postprandial secretion of ghrelin in hemodialysis patients with and without diabetic nephropathy. *Pol Arch Med Wewn*. 2009;119(7-8):447-52.
 20. Katsuki A, Urakawa H, Gabazza EC, Murashima S, Nakatani K, Togashi K, et al. Circulating levels of active ghrelin is associated with abdominal adiposity, hyperinsulinemia and insulin resistance in patients with type 2 diabetes mellitus. *Eur J Endocrinol*. 2004;151(5):573-7.
 21. Farajallah A, Ahmed A, Abdullah N, Abusnana S, Andrew P. The Relationship between Type 2 Diabetes and Total Ghrelin Level in a Population Sample in the United Arab Emirates. *IOSR J Pharm*. 2014;4(6):01-08.
 22. Rahimi MA, Izadi N, Niromand E, Rezvanmadani F, Najafi F, Asarezadegan M, et al. Comparison of serum level of 25-hydroxy vitamin d in diabetic patients and healthy subjects. *J Med Sci Islamic Azad Univ Mashhad*. 2016;59(2):97-105.
 23. Luo F-M, Liu X-J, Li S-Q, Wang Z-L, Liu C-T, Yuan Y-M. Circulating ghrelin in patients with chronic obstructive pulmonary disease. *Nutrition*. 2005;21(7-8):793-8.
 24. Barros AdF, Moraes C, Pinto MBS, Lobo JC, Mafra D. Is there association between acyl-ghrelin and inflammation in hemodialysis patients? *J Bras Nefrol*. 2013;35(2):120-6. doi: 10.5935/0101-2800.20130020.
 25. Jürimäe J, Jürimäe T, Leppik A, Kums T. The influence of ghrelin, adiponectin, and leptin on bone mineral density in healthy postmenopausal women. *J Bone Miner Metab*. 2008;26(6):618-23. doi: 10.1007/s00774-008-0861-5.