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Hepcidin and ferritin in hemodialyzed patients

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SUMMARY
Objective: Peptide hormone hepcidin is a key systemic regulator of the iron metabolism. Hepcidin binds to the iron cell exporter ferroportin so iron is kept in the cells unavailable for erythropoiesis. The synthesis of hepcidin is up-regulated by high iron stores and inflammation. Dialyzed patients have very often impaired iron management – they suffer from anemia, which is caused by many factors including the state of micro inflammation and hepcidin retention due to decreased glomerular filtration rate. Our aim was to describe the relationship of hepcidin and other parameters of iron metabolism, erythropoiesis and inflammation.
Design: Observation
Settings: Institute of Clinical Biochemistry and Hematology, Charles University Hospital in Pilsen, Alej Svobody 80, 304 60 Plzeň
Material and Methods: Complete blood cell count, iron, ferritin, transferrin, CRP, albumin, creatinine, hepcidin, soluble transferrin receptors (sTfR) and IL-6 were measured in samples from 164 patients included in chronic hemodialysis program (age 66 ± 13, 25 - 92 years), 63 women and 101 men and 37 control healthy volunteers (age 55 ± 20, 21 - 92 years), 21 women and 16 men.
Results: Iron, transferrin and hemoglobin were significantly lower in the patients group ($p<0.0001$) while ferritin ($p<0.0001$), sTfR ($p<0.05$), hepcidin ($p=0.0003$), CRP and IL-6 ($p<0.0001$) were significantly higher in the patients group. Hepcidin weakly positively correlated with CRP and ferritin and weakly negatively correlated with transferrin in hemodialyzed patients. No correlation of hepcidin with other biochemical parameters in controls was found.
Conclusion: It seems that the influence of inflammation on hepcidin levels in hemodialyzed patients is not crucial and other factors including its retention in end-stage renal disease participate.
Key words: hepcidin, hemodialysis, iron metabolism

SOUHRN
T. Sedláčková, J. Racek, J. Eiselt, L. Kielberger, L. Malánová: Hepcidin a ferritin u dialyzovaných pacientů
Cíl studie: Metabolismus železa musí být velmi pečlivě regulován. Klíčovým systémovým regulátorem je peptidový hormon hepcidin, který způsobuje "uzamčení" železa uvnitř buňky; to pak nemůže být utilizováno. Dialyzovaní pacienti často trpí anémií, která je způsobena mnoha faktory, mj. stavem mikrozánětu a také zřejmě zvýšenou retencí hepcidinu v důsledku snížené glomerulární filtrace. Naším cílem bylo popsat vztahy mezi hepcidinem a dalšími parametry metabolismu železa, zánětu a erytropoezy u těchto pacientů.
Typ studie: Observační
Název a sídlo pracoviště: Ústav klinické biochemie a hematologie LF UK a FN v Plzni, Alej Svobody 80, 304 60 Plzeň
Materiál a metody: Do studie bylo zařazeno 164 chronicky dialyzovaných pacientů (věk 66 ± 13, 25 - 92 let; 101 mužů a 63 žen) a 37 zdravých kontrol (věk 55 ± 20, 21 - 92 let; 16 mužů a 21 žen), u kterých byl stanoven jejich kompletní krevní obraz, parametry metabolismu železa, zánětu a výživy.
Výsledky: Koncentrace železa, transferrinu a hemoglobinu byla významně nižší u dialyzovaných pacientů ($p<0,0001$), naopak ferritin ($p<0,0001$), solubilní transferrinové receptory($p<0,05$), hepcidin ($p=0,0003$), CRP a IL-6 ($p<0,0001$) byly u těchto pacientů významně vyšší než u zdravých kontrol. Hepcidin slabě koreloval pozitivně s CRP a ferritinem a negativně s transferrinem u dialyzovaných pacientů, u zdravých kontrol nebyla nalezena žádná korelace hepcidinu s ostatními parametry.
Závěr: Zdá se, že vliv zánětu na hladiny hepcidinu není u hemodialyzovaných pacientů zcela zásadní a na jeho koncentraci se podílí i jiné faktory, např. jeho retence.
Klíčová slova: hepcidin, hemodialýza, metabolismus železa

Introduction

Iron is a very important biogenous trace element. It participates e.g. in the processes of hematopoiesis, oxygen transfer or cell differentiation [1]. Iron is also a transition element which means that it can take part in Fenton reaction, where the toxic hydroxyl radical is produced [2]. The daily uptake of iron compensates its daily loss caused e.g. by cell desquamation or blood loss. Any other mechanism of iron loss is not known. So iron metabolism must be very strictly regulated. Systemic regulation is mediated by hepcidin. Hepcidin is a relatively recently discovered peptide hormone [3, 4] which plays a key role in the regulation of iron metabolism [5]. It binds to the only one known iron cell exporter ferroportin and causes its internalization and degradation, so the iron is trapped in the cells and cannot be utilized [6]. The expression of hepcidin is up-regulated by iron overload and inflammation [7].

Dialyzed patients often have impaired iron management – they suffer from anemia, which is caused by many factors including the state of micro inflammation and possible hepcidin retention due to decreased glomerular filtration rate.

Our aim was to describe the relationship of hepcidin and other parameters of iron metabolism (ferritin, transferrin, and soluble transferrin receptors), erythropoiesis and inflammation.

Methods

Complete blood cell count, iron, ferritin, transferrin, CRP, albumin, creatinine (routine laboratory methods), hepcidin (ELISA, Bachem, Merseyside, UK), soluble transferrin receptors (sTfR; Roche, Mannheim, Germany) and IL-6 (ELISA, R&D Systems, Minneapolis, USA) were measured in samples from 164 patients included in chronic hemodialysis program (age 66 ± 13, 25-92 years), 63 women and 101 men and 37 control healthy volunteers (age 55 ± 20, 21-92 years), 21 women and 16 men. 79 patients started their dialysis session in the morning (6 am), 64 patients round midday and 21 patients in the evening (6 pm).

Results and discussion

Table 1 shows the results of biochemical parameters in the control group and in hemodialyzed patients (the whole group and groups according to the time of hemodialysis). The levels of all parameters were between the whole group of dialyzed patients and the control group significantly different (p-values: hemoglobin, IL-6, albumin, CRP, iron, ferritin and transferrin $p<0.0001$, hepcidin $p=0.0003$ and soluble transferrin receptors $p=0.05$). These differences between hemodialyzed patients and healthy controls indicate that the hemodialyzed patients are in the micro inflammation state, which can be one of the factors that induce the hepcidin expression.

Morning hepcidin level was significantly different from the midday and evening levels. Levels of ferritin and iron did not significantly differ in time (Fig. 1). The levels of iron and hepcidin have a daily rhythm in healthy persons, where the levels of hepcidin follow the levels of iron – minimal levels are in the morning and the peaks of maximum are during the day [8, 9]. The levels of iron in three subgroups of hemodialyzed patients (according to the time of hemodialysis) were not significantly different, maybe because of the loss of the diurnal variability of iron levels in the hemodialyzed patients caused by impaired sleep-wake rhythm [10], lifestyle, the hemodialysis procedures and the iron therapy. On the other hand, the levels of hepcidin were different in the time, but show high intra-individual and inter-individual variability [9].

Non-parametric correlation showed these weak, but statistically significant correlations in the whole groups of patients and controls: iron positively correlated with ferritin ($p=0.0001$), negatively with soluble transferrin receptors ($p<0.0001$), CRP ($p<0.0001$) and

Table 1 Results of biochemical analysis in the groups of hemodialyzed patients (all and divided into the groups according to the time of their dialysis) and healthy controls. Data expressed as *median **interquartile range

	DIALYZED PATIENTS				CONTROLS (n = 37)
	ALL (n = 164)	TIME OF DIALYSIS			
		morning (n = 79)	midday (n = 64)	evening (n = 21)	
Hemoglobin (g/l)	112*	112	113	109	139
	**104 - 121	104 - 121	107 - 122	103 - 116	133 - 148
Hepcidin (µg/l)	21.2	32.1	18.8	9.6	10.4
	9.8 - 36.9	14.5 - 62.2	10.1 - 25.7	6.0 - 23.9	7.5 - 17.5
IL-6 (ng/l)	4.7	5.2	4.0	4.6	1.0
	3.0 - 8.0	4.2 - 8.5	2.3 - 6.7	2.6 - 8.0	0.7 - 1.8
Albumin (g/l)	40.9	39.8	42.4	40.3	47.9
	38.8 - 43.0	37.8 - 42.4	40.3 - 43.6	39.3 - 42.0	45.9 - 49.9
CRP (mg/l)	3.0	4.0	2.0	3.0	1.0
	1.0 - 8.0	2.0 - 7.0	1.0 - 8.0	1.0 - 8.0	1.0 - 2.0
Iron (µmol/l)	11.1	10.5	12.0	9.3	18.4
	8.5 - 13.6	9.0 - 13.1	8.5 - 14.3	7.4 - 15.2	13.2 - 21.9
Ferritin (µg/l)	551	557	557	495	107
	426 - 823	429 - 826	415 - 875	428 - 573	64 - 172
Transferrin (g/l)	1.78	1.62	1.87	1.81	2.59
	1.52 - 1.98	1.43 - 1.88	1.65 - 2.10	1.53 - 1.92	2.43 - 2.83
Soluble transferrin receptors (mg/l)	2.99	2.94	3.03	2.99	2.51
	2.25 - 3.87	2.34 - 3.78	2.18 - 3.83	2.34 - 4.24	2.32 - 3.19

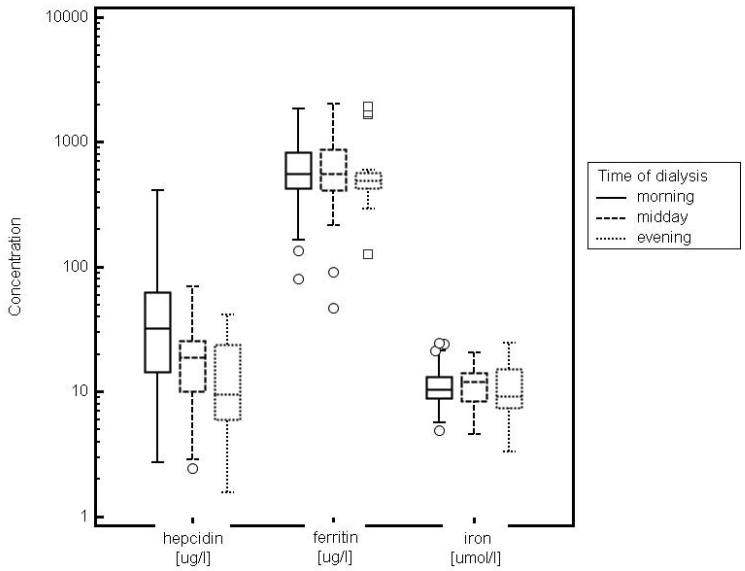

Fig. 1 Concentration of hepcidin [µg/l], ferritin [µmol/l] and iron [µg/l] levels in the group of hemodialyzed patients according to the time of hemodialysis.

IL-6 (p=0.001); ferritin positively correlated with IL-6 (p=0.02) and with hepcidin (p=0.02), negatively with soluble transferrin receptors (p=0.001); transferrin negatively correlated with IL-6 (p=0.0006). No correlations of hepcidin with other biochemical parameters in controls and in group of all patients were found.

The absence of a significant correlation of hepcidin with any parameter of inflammation can indicate that the influence of the inflammation on the hepcidin production is not crucial and other factors like hepcidin retention due to decreased glomerular filtration rate can participate. The relations in the groups of hemodialyzed patients after dividing according to the time of their hemodialysis will be further studied.

Conclusion

Hemodialyzed patients have often impaired iron homeostasis. It is caused by peptide hormon hepcidin, which is up-regulated due to inflammatory state and possible retention in organism. We studied the hemodialyzed patients and the healthy controls to describe the relationship of hepcidin and parameters of iron metabolism, erythropoiesis and inflammation. Parameters of iron metabolism, erythropoiesis and inflammation were significantly different between the patients group and control group. There was a weak correlation between hepcidin and ferritin and CRP in the hemodialyzed patients; it seems that the influence of inflammation on hepcidin levels in hemodialyzed patients is not crucial and other factors including its retention in end-stage renal disease participate.

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