

EVALUATION OF SOME TRACE ELEMENTS AND PARATHYROID HORMONE LEVELS IN CHILDREN WITH CHRONIC RENAL FAILURE ON REGULAR HEMODIALYSIS

By

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ABSTRACT

Background: Secondary hyperparathyroidism and trace elements metabolism disturbance are common, important and treatable complications of CRF. Both parathyroid hormone (PTH) and trace elements disturbances lead to bone disease and affect the growth of the children on regular hemodialysis (HD).

Objectives: To measure the serum levels of the following trace elements (zinc –copper - manganese) and parathormone in children with end stage renal disease (ESRD) on regular hemodialysis and their relation to the duration of dialysis. In addition to, study the effect of trace elements dysequilibrium on the mental status in these children.

Patients and Methods: This is a prospective case control study was done on thirty cases with ESRD on regular hemodialysis. Also twenty of apparently healthy children age and sex matched with cases were included as a control group. After complete clinical evaluation of cases and controls including weight, height, body mass index, blood pressure and all body systems, serum zinc (Zn), copper (Cu), manganese and parathyroid hormone was measured. Also, Cognitive and behavioural assessment using Stanford-Binet intelligence score was done.

Results: There was a significant decrease in serum zinc and copper in patients when compared to controls. On the other hand, no significant changes in serum manganese in all patients relative to controls. Also, there was a significant increase in PTH in patients relative to controls. There was a significant inverse correlation between serum PTH and both of zinc and copper, with insignificant correlation between PTH and manganese. Patients on HD were significantly associated with more severe decreased intelligence according to Stanford Binet Score when compared to control group.

Conclusion: Majority of patients with ESRD on regular hemodialysis showed decreased Zn & Cu and elevated PTH level with inverse correlation between duration of dialysis and serum levels of Zn and Cu and direct correlation with PTH level.

Key words: *Chronic Kidney Disease, Parathyroid Hormone, Hemodialysis, End Stage Renal Disease, Zinc, Copper.*

INTRODUCTION

End-Stage Renal Disease (ESRD) is the final stage of chronic renal failure (CRF) characterized by progressive, irreversible deterioration in renal function and body fails to maintain fluid and electrolyte balance resulting in uremia. ESRD is characterized by a decrease in GFR and evidence of less than 10% nephron function remaining (**Aravind kumar et al, 2015**).

Trace elements play an important role in the structure of proteins, enzymes and complex carbohydrates to participate in biochemical reactions. It also involved in a number of metabolic activities, including nerve conduction, transport excretory processes and serving as cofactors for enzymes (**Boosalis et al, 2008**).

In renal insufficiency, trace elements metabolism can be affected with the reduced renal function, alteration of metabolic balance, special diet, medications, and dialysis treatment. Various trace elements abnormalities in long term hemodialysis patients have been reported. These disturbances are influenced by the

direction and rate of the elements transfer during dialysis procedure, which depends on their concentrations in the dialysis fluid (**Kiziltas et al, 2008**).

Copper plays an essential role in the function of some enzymes like lysyl oxidase which helps in maintaining the integrity of connective tissue in the heart and blood vessels. It also plays a part in bone formation and teeth. Clinical characteristics of Copper deficiency are leucopenia, microcytic hypochromic anaemia and osteopenia. Serum levels of total protein and especially of albumin in patients with ESRD are usually low (**Food and Nutrition Board, 2001**).

Manganese is an essential trace element for all life forms. The classes of enzymes that require manganese cofactors are broad. Several defects have yet been associated with manganese deficiency such as hair colour and growth alterations, loss of weight, inhibition of vitamin k dependent clotting and altered cholesterol transport and utilization. Manganese toxicity has been linked to impaired motor skills and cognitive disorders (**Culotta et al, 2007**).

Secondary hyperparathyroidism and trace elements metabolism disturbance are common, important and treatable complications of CRF. Both PTH and trace elements disturbances lead to bone disease and affect the growth of the children on regular hemodialysis (El Tayeb et al, 2009).

AIM OF THE WORK

The aim of this study was to measure the serum levels of the following trace elements (zinc – copper - manganese) and parathormone in children with ESRD on regular hemodialysis and their relation to the duration of dialysis. Also, to study the effect of trace elements dysequilibrium on the mental status in these children.

PATIENT AND METHODS

This is a prospective case control study was carried out on thirty cases with ESRD on regular hemodialysis. Those patients were chosen for the study from Nephrology Unit, Pediatric Department, Al-Azhar University Hospitals from March 2017 to March 2019.

Their ages ranged from 6 to 18 years. They were 18 males and 12 females. All patients are under regular hemodialysis (for at least one year), three times per week,

with each dialysis session lasting for three to four hours. Also twenty of apparently healthy children age and sex matched with the cases were included as a control group.

Inclusion criteria:

All children with ESRD and treated by regular hemodialysis (for at least one year) will be included in the study aging from 6 to 18 years old.

Exclusion criteria:

- Patients with malabsorption syndrome.
- Children with parathyroidectomy.
- Children with chronic comorbid conditions.
- Children on hemodialysis with dialysis duration less than one year.

Ethical considerations:

- An informed consent was obtained from all parents of patients and control group before getting them involved in the study.
- This study was approved by the ethical committee for researches of both Pediatric Department and Al-Azhar faculty of medicine.
- The steps of the study, the aim, the potential benefits and

hazards, all were discussed with the parents of the studied groups.

- Confidentiality of all data was ensured.
- The patients and controls had the right to withdraw from the study at any time without giving any reasons.
- The authors declared that there was no conflict of interest regarding the research and publication
- The authors received no financial support regarding the research and publication

All patients and controls were subjected to the following:

1. Detailed history especially for: the cause of ESRD, duration of dialysis, regular drug taking, bone disease (fractures, bone deformity or muscular weakness) and recurrent infections.
2. Thorough clinical examination including: height, weight, body mass index (BMI), blood pressure and all body systems.
3. Laboratory investigations including:
These investigations were done just pre-dialysis and sent directly to the lab:
 - a. Complete blood count.
 - b. Serum creatinine and blood urea nitrogen (BUN).
 - c. Serum albumin and total protein.
 - d. Serum calcium and phosphorus.
 - e. Serum alkaline phosphatase.
 - f. Serum intact parathyroid hormone (iPTH) level.
 - g. Serum zinc, copper and manganese.
4. Cognitive and behavioural assessment: using Stanford-Binet intelligence score.

RESULTS

The results of our study are shown in the following tables and figures:

Table (1): Age & sex of the studied cases & controls

		Patient group N= 30		Control group N=20		test	P-Value
Age (years)	Range	8-18		6-17		T=0.854	0.397 ^T
	mean±SD	13.2±3.2		13.9±2.6			
Sex		N	%	N	%	X ² =1.08 7	0.297 ^C
Male		18	60	9	45		
Female		12	40	11	55		

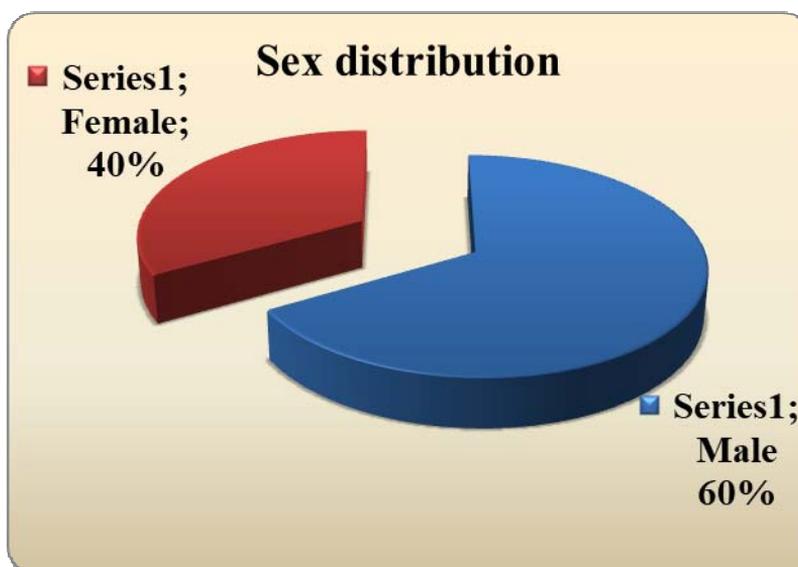


Figure (1): Sex distribution of the studied cases.

Table (1) and Figure (1) revealed that there was no significant difference between the two groups as regard age and

sex and prevalence of CKD in males (60%) more than females (40%).

Table (2): Etiology of CKD in the studied patients

Causes of ESRD		HD cases N= 30	
		N	%
❖ Obstructive Uropathy:		9	30
	a. Neurogenic bladder	4	13.4
	b. Posterior urethral valve	2	6.7
	c. Vesico-ureteric reflux	2	6.7
	d. Bilateral ureteric stenosis	1	3.3
❖ Hypoplastic kidneys		5	16.7
❖ Glomerulonephritis:		4	13.3
	a. Lupus glomerulonephritis	2	6.7
	b. Mesangioproliferative glomerulonephritis	1	3.3
	c. Focal segmental glomerulonephritis	1	3.3
Hereditary Nephropathy (Nephronophthisis)		3	10
Chronic pyelonephritis		2	6.7
Cystinosis		1	3.3
Barter Syndrome		1	3.3
Distal RTA (2ry to nephrocalcinosis)		1	3.3
Bilateral Polycystic Kidneys		1	3.3
Urolithiasis (Familial hyperoxaluria type I)		1	3.3
Unknown		2	6.7

Table (2) and Figure (2) revealed that obstructive

uropathy is the most common cause of CKD in our patients.

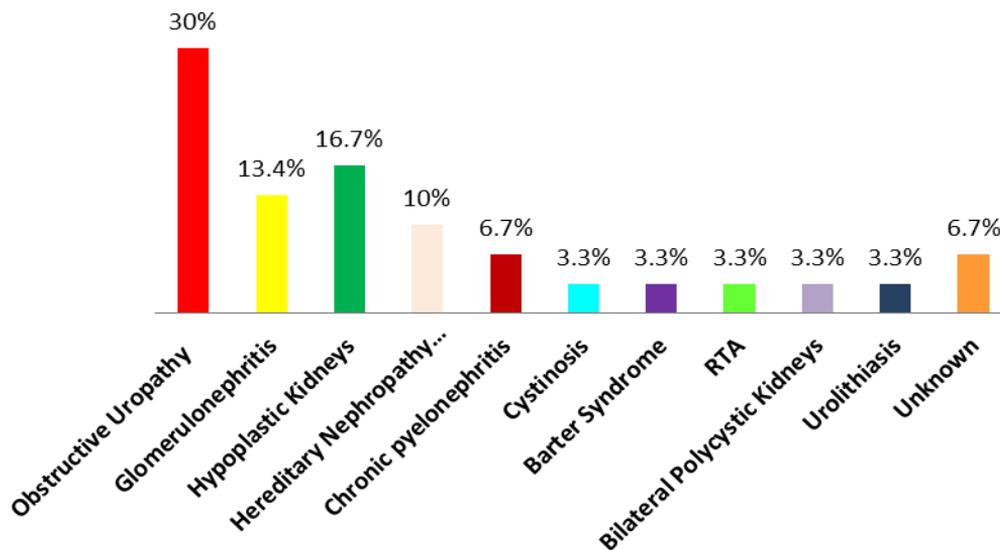


Figure (2): Etiology of CKD in the studied cases

Table (3): Comparison of anthropometric data among studied groups

		Patient group N= 30	Control group N=20	test	P Value
Weight (kg)	Range	14-51	17.2-74.5	T=1.986	<0.001
	mean±SD	26.4 ± 8.7	34.9 ±10.6		
Height (cm)	Range	101-153	113-175	T=1.973	<0.001
	mean±SD	121.5± 14.2	139.2 ± 19.4		
BMI (Kg/m ²)	Range	9.7-21.3	13.4 -24.4	T=2.819	<0.001
	mean±SD	14.6 ± 2.4	17.9 ± 3.1		

Table (3) revealed that HD cases were significantly associated with lower weight,

shorter height, lower BMI when compared to control group.

Table (4): Comparison of laboratory data among studied groups

		Patient group N=30	Control group N=20	test	P- Value
Total Protein (g/dl)	Range	5.2-7.1	6.9-8.2	T=1.288	0.204
	mean±SD	6.3±0.8	7.2±0.7		
Albumin (g/dl)	Range	2.8-4.1	3.5-5.3	T=3.839	<0.001
	mean±SD	3.2±0.6	4.1±0.6		
Calcium (mg/dl)	Range	7-14	9-11	T=1.318	0.194
	mean±SD	10±2	9.4±0.5		
Phosphorus (mg/dl)	Range	2-10	4-6	T=3.338	<0.001
	mean±SD	5.2±1.1	4.6±0.6		
Alkaline Phosphatase (U/L)	Range	29-1158	38-95	T=4.246	<0.001
	mean±SD	370.2±106.7	77.7±16.8		
PTH (pg/ml)	Range	6.3-1761	9.6-56.9	T=3.738	<0.001
	mean±SD	421.8±125.6	31.1±10.1		

Table (4) revealed that patient group was significantly associated with higher phosphorus, ALP, PTH and lower albumin concentration when compared to control group.

Table (5): Comparison of trace elements concentration among studied groups

		Patient group N= 30	Control group N=20	test	P- Value
Zinc (ug/dl)	Range	27 - 121	35 - 127	T=4.431	<0.001
	mean±SD	48.9 ±15.1	87.3 ± 23.9		
Copper (ug/dl)	Range	47 - 142	42 - 152	T=3.271	0.002
	mean±SD	70 ± 23.9	101 ± 29.1		
Manga nese (mg/l)	Range	0.04 - 0.34	0.09 - 0.34	T=1.116	0.270
	mean±SD	0.19 ±- 0.06	0.21 ±- 0.07		

- Reference range for Zinc: (70 – 127) mcg/dl.
- Reference range for Copper: (70 – 155) mcg/dl.
- Reference range for Manganese: (0.03 – 0.34) mg/dl.

Table (5) revealed that patient group was significantly associated with lower zinc and copper concentrations when compared to control group.

Manganese concentration was within normal range in both case and control groups and did not differ significantly between both groups.

Table (6): Correlation between zinc, copper and manganese levels and other parameters in all studied hemodialysis cases

	Zinc		Copper		Manganese	
	r	P-Value	r	P-Value	r	P-Value
Age	0.105	0.579	0.116	0.543	0.164	0.388
Dialysis duration	-0.496	0.005	-0.527	0.003	-.137	.470
Weight percentile	-0.128	0.499	-0.211	0.263	-0.197	0.297
Height percentile	0.536	<0.001	0.609	<0.001	-0.097	0.610
BMI percentile	-0.271	0.147	-0.274	0.143	-0.155	0.414
SBP	0.046	0.808	-0.111	0.559	-0.153	0.419
DBP	0.121	0.526	-0.011	0.955	-0.146	0.440
WBC	0.192	0.309	0.207	0.272	-0.123	0.516
RBC	-0.001	0.996	0.044	0.819	0.329	0.076
Hemoglobin	0.122	<0.001	0.139	<0.001	0.054	0.789
Platelets	0.131	0.489	0.172	0.364	-0.186	0.325
BUN	-0.120	0.529	-0.213	0.258	-0.079	0.680
Creatinine	0.006	0.975	-0.115	0.547	-0.165	0.384
Total Protein	0.200	0.290	0.300	0.108	0.086	0.653
Albumin	0.106	<0.001	0.291	0.118	0.118	0.533
Calcium	-0.067	0.725	0.044	0.818	-0.095	0.619
Phosphorus	0.005	0.981	-0.004	0.983	0.142	0.455
Alkaline Phosphatase	0.352	0.056	0.129	0.498	0.224	0.235
PTH	-0.530	0.003	-0.589	0.001	0.125-	0.511

Table (6) revealed that:

- Zinc level showed significant direct correlation with height, hemoglobin and albumin; and significant inverse correlation with dialysis duration and PTH.
- Copper level showed significant direct correlation with

height and hemoglobin; and significant inverse correlation with dialysis duration and PTH.

- No significant correlations were found between manganese level and other studied parameters.

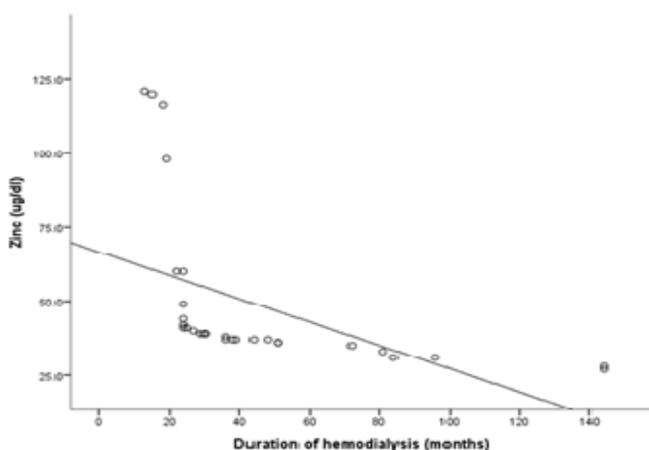


Figure (3): Correlation between zinc and dialysis duration in HD cases

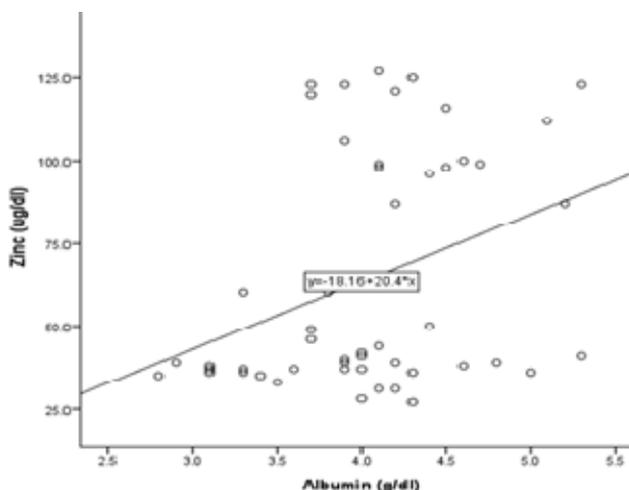


Figure (4): Correlation between zinc and albumin in HD cases

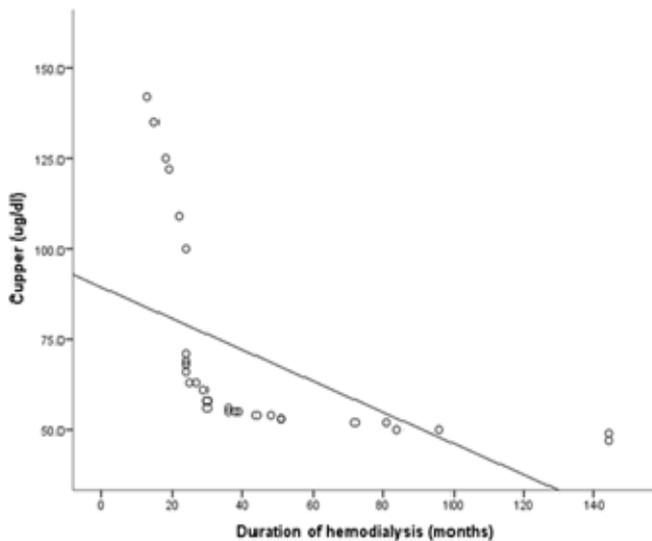


Figure (5): Correlation between cupper and dialysis duration in HD cases

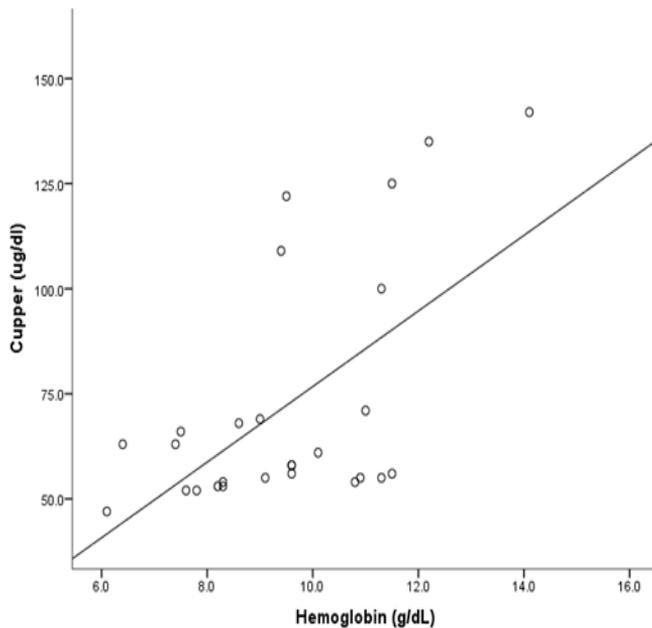


Figure (6): Correlation between cupper and hemoglobin in HD cases

Table (7): Comparison of Stanford Binet intelligence Score among studied groups

		Patient group N= 30		Control group N=20		test	P-Value
		N	%	N	%		
Stanford Binet Score	Average	6	20	10	50	F=11.946	0.020
	Dull	2	6.7	3	15		
	Borderline	5	16.7	5	25		
	Mild M.R	8	26.7	2	10		
	Moderate M.R	7	23.3	0	0		
	Severe M.R	2	6.7	0	0		

- F, Fisher exact test.

Stanford Binet intelligence Score:

- Average : (90-109)
- Dull : (80-89)
- Borderline: (70-79)
- Mild M.R: (55-69)
- Moderate M.R: (40-54)
- Severe M.R: (25-39)

Table (7) revealed that HD cases were significantly associated with more severe decreased

intelligence according to Stanford Binet intelligence Score when compared to control group.

DISCUSSION

Hemodialysis (HD) patients are at risk of developing trace elements imbalances, most of the elements must be kept in a rather narrow physiological range during HD; otherwise life threatening events may occur. Reduced or enhanced blood levels of some

trace elements may be deleterious for the chronic HD children. Low levels of some essential trace elements have been associated with adverse outcome in the general population (**Kalantar, 2003**). Secondary hyperparathyroidism and trace elements metabolism disturbances are common and important

complications of chronic renal failure (**El Tayeb et al, 2009**).

Laboratory assessment of serum some trace elements [zinc (Zn), copper (Cu) and manganese (Mn)] levels as well as PTH in children with ESRD on regular hemodialysis was the main scope of our study and to compare these elements with the healthy control group. Also, to assess cognitive and behavioural patterns in these children using Stanford-Binet intelligence score.

As regard to demographic data, our study revealed that the prevalence of CKD was higher in males (60%) than in females (40%) (**Table 1, Figure 1**). Male preponderance may be explained by higher prevalence of obstructive uropathy in our study which is generally common in boys. This agrees (**Harambat et al., 2012**) who reported that the incidence and prevalence of CKD is greater in males than females because of the higher frequency of congenital abnormalities of the kidney and urinary tract (CAKUT) in males.

Regarding the etiology of CKD, obstructive uropathy was the commonest cause of CKD in our study (**table 2, figure 2**). This agrees with (**Harambat et al., 2012**) who reported that congenital anomalies of the kidney

and urinary tract (CAKUT) mainly obstructive uropathy was the commonest cause of CKD (50%).

Our results also agree with (**Ragab, 2007**) who reported that the most common cause of CKD in dialysis group is congenital kidney diseases mainly obstructive uropathy (55.6%).

Zinc depletion was suggested to play a role in the pathogenesis of some uremic symptoms, e.g_ growth retardation, immunological impairment, impotence (testicular atrophy), abnormalities of taste (dysgeusia, anorexia) and olfaction. Previous reports have indicated a significant decrease in plasma zinc concentration in hemodialysis patients as compared with healthy controls (**Berger, 2004**).

Our study showed that there was a marked significant decrease in the serum zinc between patients and controls (P value < 0.001) (**table 5**). This agrees with the study done by (**Yilmaz et al, 2012**) on 26 patients on regular HD and found a highly significant decrease in Zn level between patients than controls.

Copper plays an essential role in the function of some enzymes like lysyl oxidase which helps in maintaining the integrity of connective tissue in the heart and blood vessels. It also plays a part

in bone formation and teeth (**Food and Nutrition Board, 2001**).

Our study showed that there was a marked significant decrease in the serum copper between patients and controls (P value < 0.001) (**table 5**). This agrees with the study done by (**Omali et al., 2012**) on patients on regular HD and found a highly significant decrease in Zn level between patients than controls.

Manganese has an important role in regulating metabolic processes which mainly include lipid and carbohydrate metabolism, bone and tissue formation, skeletal growth and reproduction (**Ishimura et al, 2007**).

The present study showed that there was no significant change in the serum manganese between patients and controls.

Conflicting results have been reported regarding serum level of the manganese in hemodialysis patients. There are reports of low, normal or high manganese level in chronic dialysis patients. Dialysis fluid may be a source of increased manganese level (**Hosokawa, 2009**).

There was a strong positive correlation between zinc and height, hemoglobin and albumin. Also, there was a strong positive

correlation between copper and both height and hemoglobin.

There was a strong negative correlation between zinc and both dialysis duration and PTH. Also, there was a strong negative correlation between copper and dialysis duration and PTH.

No significant correlations were found between manganese level and other studied parameters.

The present study evaluated intelligence using Stanford Binet Score and showed that patients on HD were significantly associated with more decreased intelligence than control group (**Table 7**).

CONCLUSION

- The children with ESRD on regular hemodialysis showed decreased serum Cu and Zn levels with no significant changes in serum Mn levels.
- The children with ESRD on regular hemodialysis showed elevated PTH levels.
- The serum levels of Cu and Zn had inverse correlation with the hemodialysis duration.
- The serum PTH levels had direct correlation with the hemodialysis duration.

RECOMMENDATIONS

- The need of more studies on more trace elements and their

disturbance's effects on the bone disease, CVD and immune system in the HD children.

- Measurement and follow up of the trace elements and PTH when the child starting dialysis and at regular intervals during dialysis.
- Oral trace element supplementation as (oral zinc) to be given to the patients on hemodialysis.
- Regular nutritional assessment of the children with ESRD on regular hemodialysis.
- The need of program for renal transplantation for children with ESRD to decrease the longtime of dialysis and improvement of their life quality and outcome.

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تقييم مستوى بعض العناصر النادرة وهرمون الغدة الجاردرقية في الأطفال المصابين بالفشل الكلى المزمن والخاضعين للغسيل الكلى الدموي المنتظم

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حماد**

مقدمة البحث:

مرض الكلى المزمن هو مشكلة صحية عامة رئيسية يمكن الوقاية من النتائج الضارة له من خلال الكشف والعلاج المبكر ويمكن اكتشاف المراحل المبكرة من مرض الكلى المزمن من خلال التحاليل المعملية الروتينية. العناصر النادرة حيوية لجسم الإنسان للحفاظ على الوظائف الفسيولوجية المعقدة العادية المرتبطة بنمو الجسم وتنميته. الاحتياجات اليومية من العناصر النادرة الغذائية هي قليل من المليغرام والمعروفة أيضا باسم المغذيات الدقيقة. فرط نشاط هرمون الغدة الجاردرقية الثانوي المصاحب للفشل الكلى واضطرابات التمثيل الغذائي للعناصر النادرة هي المضاعفات الأكثر أهمية وشيوعا في مريض الفشل الكلى المزمن.

الهدف من البحث:

والهدف من هذه الدراسة هو تحديد مستوى بعض العناصر النادرة (النحاس، الزنك، المنجنيز) وهرمون الغدة الجاردرقية بالدم وتقييم مستوى الذكاء بإستخدام مقياس

ستانفورد بينيه في الأطفال الذين يعانون من الفشل الكلوي المزمن والخاضعين للغسيل الكلوي الدموي المنتظم وعلاقتهم بمدّة الغسيل الكلوي وكيف تؤثر على الحالة الغذائية للأطفال.

طريقة البحث:

أجريت هذه الدراسة بوحدة الغسيل الكلوي الدموي بقسم طب الأطفال بمستشفيات جامعة الأزهر وقد أجريت على ثلاثين طفلا من الذين يعانون من المراحل النهائية للفشل الكلوي والذين يخضعون لجلسات الغسيل الكلوي الدموي بصفة دورية ومنتظمة (ثلاث مرات أسبوعيا) وقد تم اتخاذ عشرين طفلا طبيعيا تماما ممن يتناسب سنهم ونوعهم مع الأطفال موضوع البحث كمجموعة ضابطة. تم أخذ التاريخ المرضي للمجموعتين شاملا مدة وسبب المرض والفترة منذ بدء الغسيل الدموي وتم تسجيل البيانات الإكلينيكية شاملة الوزن والطول وسمك طبقة الجلد وضغط الدم الشرياني وتم قياس نسبة الزنك والنحاس والمنجنيز وهرمون الغدة الجاردرقية بالدم وتقييم مستوى الذكاء باستخدام مقياس ستانفورد بينيه.

نتائج البحث:

وجد في مجموعة الأطفال المرضى انخفاض بنسبة الزنك والنحاس بينما وجد ارتفاع بنسبة هرمون الغدة الجاردرقية وكان ذو دلالة إحصائية بالمقارنة بالمجموعة الضابطة مع عدم وجود تغير ملموس في المنجنيز.

كما وجد علاقة سلبية ذات دلالة إحصائية بين هرمون الغدة الجاردرقية و الزنك والنحاس في مجموعة الأطفال المرضى، وعدم وجود ارتباط بين هرمون الغدة الجاردرقية والمنجنيز.

التوصيات:

ولذا فإننا نوصي بقياس مستوى العناصر النادرة وهرمون الغدة الجاردرقية عند بداية الغسيل الكلوي وعلى فترات منتظمة أثناء الغسيل، مع اضافة بعض تلك العناصر الضرورية كمكملات غذائية عن طريق الفم (كالزنك) لمرضى الغسيل الكلوي مع مراقبة جيدة ومتابعة بانتظام إلى امتثال المرضى لنظامهم الغذائي وتناول الأدوية. ونوصي أيضا بتوفير برنامج لزراعة الكلى للأطفال المصابين بالفشل الكلوي المزمن لتقليل فترة الغسيل الكلوي وتحسين أسلوب حياتهم وطول أعمارهم.