Impact of Haemodialysis on Certian Trace Elements Among Patients Suffering from End Stage Renal Disease.

EL-Habibi, E.M.[1]; Bakr, M.A.[2]; and Kamal, N.[1]

[1] Zoology Department, Faculty of Science, Mansoura University, Mansoura, Egypt.
[2] Urology and Nephrology, Center, Mansoura University, Mansoura, Egypt.
eelhabibi555@yahoo.com

Abstract: Hemodialysis (HD) patients are at risk of developing trace elements imbalance and thus the establishment of normal value of trace elements in HD patients is of great importance. The objective of this study was to compare plasma trace elements [copper (Cu), zinc (Zn), chromium (Cr), cobalt (Co), manganese (Mn) and selenium (Se)] levels between hemodialysis Egyptian patients and healthy controls. Forty four hemodialysis patients with End Stage Renal disease (ESRD) and 44 control subjects were enrolled. The patients were 21 males and 23 females, their ages ranged from 20 to 77years. The control subjects were 21 males and 23 females, their ages ranged from 20 to 65years. Blood samples were collected before and after dialysis sessions and from control. Atomic absorption spectrophotometer was applied to measure the plasma levels of the studied trace elements. Results showed that Mn, Co, Se and Cr levels in plasma of hemodialysis patients were significantly decreased ($p<0.01$) in comparison to healthy controls. There weren't any significant differences in Cu and Zn concentrations between patients and their controls. On the other hand, the data also revealed that hemodialysis significantly increased ($p <0.05$) Cr level only in male patients, however in female patients a significant increase ($p <0.05$) was observed in Se. In conclusion, this study revealed that plasma trace element concentrations in HD patients are distinctly different compared to that of healthy controls. Elements such as Mn, Co, Se and Cr are reduced in HD patients, while Cu and Zn are not affected. Regular monitoring of trace elements in hemodialytic patients is advisable.

[EL-Habibi, E.M.; Bakr, M.A.; and Kamal, N. **Impact of Haemodialysis on Certian Trace Elements Among Patients Suffering from End Stage Renal Disease**. *J Am Sci* 2012;8(12):827-833]. (ISSN: 1545-1003). http://www.americanscience.org. 115

Keywords: Hemodialysis, Trace elements, chronic renal failure, Atomic absorption spectrophotometer.

1. Introduction

Trace element is defined as one that makes up less than 0.01% of body's mass. Those present at ug/dl in body fluids and at mg/kg in tissues are referred to as trace elements and those found at ng/dl in body fluids or ug/kg in tissues are referred to as ultra trace elements. Although these elements constitute a relatively small amount of total body tissues they are essential for many vital processes **(Anees *et al*., 2011)**.

Trace element plays an important role in chemical, biological, biochemical, metabolic, catabolic and enzymatic reactions in the living cells of plants, animals and human beings. Trace element have great significance due to their tendency to accumulate in the vital human organs over prolong period of time **(Hashmi *et al*., 2007)**.

Chronic renal failure (CRF) is a clinical syndrome that results in the permanent loss of renal function due to the progressive loss of nephrons. Whatever it is, that causes the renal damage in CRF, it is ultimately the progressive loss of nephrons that leads to End Stage Renal disease (ESRD). Hemodialysis (HD) remains the most common technique for the treatment of patients with ESRD **(Yilmaz *et al*., 2000)**.

Altered blood and tissue concentrations of trace elements have been described in patients with chronic renal failure treated by hemodialysis **(El-Sherbeny *et al*., 2006)**. During dialysis some trace elements can accumulate in the body because of dialysis fluid impurities and others may remove from blood to dialysate leading to deficiency of some trace elements in the body **(Esfahani *et al*., 2007).**

In ESRD patients different factors affect serum concentrations of trace elements like increased oral intake, failure of renal excretion, degree of renal failure, use of medications, contamination of dialysate, quality of water used for dialysis and metabolic alterations associated with renal failure **(Miura *et al*., 2002)**.

Excessive accumulation or depletion of trace elements may have significant clinical implication, including increased risk for cancer, cardiovascular disease, immune deficiency, anaemia, renal function impairment and bone disease **(Vanholder *et al*., 2002)**.

Accordingly, the present study was designed to compare plasma trace elements [copper (Cu), zinc

(Zn), chromium (Cr), cobalt (Co), manganese (Mn) and selenium (Se)] levels between hemodialysis patients (before and after HD) and healthy controls.

2-Subjects and Methods

2.1. Patients

This study involved 44 hemodialysis patients with end stage renal failure ("hemodialysis group", 23 females and 21males) who were treated in the Urology and Nephrology Center, Mansoura University, Mansoura, Egypt. The mean age of the patients was 41.71±14.41 years for males and 48.00±14.03 years for females. The control group was composed of 44 healthy volunteers (23females, 21 males) with a mean age 36.52±12.04 years for males and 40.17±11.58 years for females. The study subjects were all in the same socioeconomic class and had similar nutritional habits. Each patient was medically stable and was dialyzed through arteriovenous fistula on AK 200 machine (Gambro) for 4 hours session using a blood flow rate 350 ml/min and dialysate flow of 500 ml/min. Each patient was dialyzed 3 times per week using polysulfone membrane. The dialysate fluids were prepared from concentrated salt solutions and from bicarbonate powder in sealed containers. The final dialysate contained the following components: Na^+ 140 mmol/l, K^+ 2.0 mmol/l, Ca^{++} 1.50 mmol/l, Mg^{++} 0.50 mmol/l, HCO_3^- 33.00 mmol/l and $CH3COO^-$ 2.0 mmol/l. The peroid of dialysis ranged from 1-3 years.

2.2. Samples Collection

Blood samples were taken from all the hemodialysis patients before (pre-hemodialysis) and after (post-hemodialysis) the dialysis sessions and from control subjects by collecting 4 ml venous blood using a disposable plastic syringe and transferred to heparinized plastic tubes to obtain plasma for measuring trace elements (Cu, Zn, Cr, Co, Mn and Se) and non heparinized tubes to obtain sera for estimation of Creatinine, Alkaline Phosphatase, total protein, albumin, total bilirubin and SGPT.

Sera and plasma from all subjects were obtained by centrifuging blood samples at 2000 rpm for 10 min at 4°C and separated into sterile tubes. All samples were stored at 4°C in a refrigerator until they were analyzed.

2.3. Chemicals and Reagents

Bidistilled water was used throughout. All glass ware were cleaned by soaking for 24 hrs in concentrated nitric acid followed by rinsing 3 times with high-purity water. Digestions of plasma were carried out with Analar concentrated nitric acid **(Krachler and Wirnsberger, 1997)** and subsequently brought up to 5 mL with double distilled water.

Creatinine level was estimated by the method of **(Vasiliades, 1976)**, alkaline phosphates were estimated by the method of **(Babson *et al.,* 1966)**, total protien was estimated by the method of **(Ryan and Chopra, 1976)**, albumin was estimated by the method of **(Pinnell and Northam, 1978)**, total bilirubin was estimated by the method of **(Malloy and Evelyn, 1937)** and SGPT was estimated by the method of **(Henry *et al.,* 1960)** using kits supplied by Bickman coulter. U.S.A.

2.4. Mineralization of Plasma and Determination of Trace Elements

Six trace elements were measured: Cu, Zn, Co, Mn, Cr and Se by Atomic Absorption Spectrophotometer. Atomic absorption spectrometer {Perkin- Elmer model 2380} equipped with multi-elemental hallow cathode lamp and air-acetylene burner was used for the determination of trace elements in plasma. The concentration of trace elements was determined at part per million (ppm) or part per billion (ppb) concentrations.

2.5. Statistical Analysis

Results were expressed as mean ± standard deviation (SD). Statistical analysis was performed using the Statistical Package for Social Sciences (SPSS) 19.0 software. One-way analysis of variance (ANOVA) was performed followed by a post hoc Tukey test for between-group comparison. Correlation coefficients were calculated by Pearson's correlation coefficient. A p-value of <0.05 was considered significant for ANOVA.

3-Result

Forty four patients of ESRD on HD were included in this study. There were 23(52.3%) females and 21 (47.7%) males in the patient group. The mean age of the patients was (45±14.40) years. Lab parameters of dialysis patients and controls are shown in table (1). The obtained results showed a significant difference ($p<0.05$) in serum creatinine, total protein, albumin, total bilirubin and SGPT in HD male and female patients compared to matched controls. However, non-significant difference was observed in alkaline phosphatase activity.

The comparison of creatinine and trace elements before and after dialysis among male patients with control subjects are shown in table (2). In pre-hemodialysis patients, the level of creatinine was higher and levels of Mn, Co, Se and Cr were lower than in controls. There were no significant differences in Zn and Cu levels between controls and the pre-hemodialysis patients.

While creatinine level was significantly higher and levels of Mn, Co, Se and Cr were significantly lower in patients after hemodialysis than in the control group, non- significant differences in levels of Zn and Cu between the control and post-hemodialysis groups were observed.

Creatinine level of post-hemodialysis patients was significantly lower while the level of Cr was significantly higher than levels before the dialysis session. In those groups, levels of Zn, Cu, Mn, Co and Se were not significantly different.

The comparison of creatinine and trace elements before and after dialysis of female patients with control subjects are shown in Table (3). In pre-hemodialysis patients, the level of creatinine was significantly higher and levels of Mn, Co, Se and Cr were significantly lower than in controls. There were no significant differences in Zn and Cu levels between controls and the pre-hemodialysis values.

While creatinine level was significantly higher and levels of Mn, Co, Se and Cr were significantly lower in patients after hemodialysis than in the control group, we did not determine any significant differences in levels of Zn and Cu between the control and post-hemodialysis groups.

Creatinine level of post-hemodialysis patients was significantly lower while the level of Se was significantly higher than levels before the dialysis session. In those subjects, levels of Zn, Cu, Mn, Co, and Cr were not significantly different.

Correlation test in the (male) post-hemodialysis patients showed significant positive correlations between Se and Cr (r=0.75, p <0.001), Cu and Cr (r=0.60, $p < 0.01$), Cu and Se (r=0.63, $p < 0.01$) (Figure 1), and a significant Negative correlations between Cr and Mn (r= -0.073, $p < 0.01$) (Figure 2).

In the (female) post-hemodialysis patients a significant positive correlations were observed between Se and Cr (r=0.87, $p < 0.01$) and Cu and Cr (r=0.57, $p < 0.05$) (Figure 3).

Table1: Descriptive data of patients and control subjects.

	Hemodialysis patient(males)	Hemodialysis patient(females)	Control(males)	Control(females)
Age(years)	41.71±14.41	48.00±14.03*	36.52±12.04	40.17±11.58
Sex(M/F)	21	23	21	23
Kt/V	1.50±0.25	1.54±0.20	----	----
Creatinine(mg/dl)	12.87±4.87*	9.87±.2.04*	0.94±0.17	0.91±0.20
Alkaline Phosphatase (IU/L)	162.2±114.1	145.7±99.81	198.38±62.78	189.52±71.55
Total protein(g/dl)	7.18±0.68	6.95±0.86*	7.44±0.79	7.45±0.71
Albumin(g/dl)	3.27±0.27*	3.21±0.55*	3.96±0.34	3.93±0.32
Total bilirubin(mg/dl)	0.52±0.13*	0.50±0.12*	0.82±0.14	0.80±0.14
SGPT(IU/L)	21.81±10.67*	21.83±20.31*	31.81±7.30	32.30±8.11

Data are given as (mean ± standard deviation).
* Hemodialysis patients significant to control at P< 0.05 level.

Table 2: Parameter levels of control and hemodialysis male groups (mean ± standard deviation).

Parameters	Groups			*P*-value		
	Control (A)	Pre-hemodialysis (B)	Post-hemodialysis (C)	A vs B	A vs C	B vs C
Creatinine(mg/dl)	0.94±0.17	12.87±4.87	5.13±2.41	0.000	0.000	0.000
Zn(ppm)	65.58±16.1	66.50±15.6	64.73±17.18	0.98	0.98	0.93
Cu(ppm)	1.48±0.41	1.60±0.58	1.61±0.60	0.74	0.71	0.998
Mn(ppm)	4.99±1.36	1.45±0.59	1.37±0.66	0.000	0.000	0.957
Co(ppm)	7.97±2.47	4.44±2.47	5.03±2.35	0.000	0.003	0.763
Se(ppm)	9.65±1.55	4.09±1.14	5.27±2.33	0.000	0.000	0.92
Cr(ppm)	15.38±3.02	7.27±2.24	9.83±3.30	0.000	0.000	0.015

- A significant value was $p < 0.05$

Table 3: Parameter levels of control and hemodialysis female groups (mean ± standard deviation).

Parameters	Groups			*P*- value		
	Control (A)	Pre-hemodialysis (B)	Post-hemodialysis (C)	A vs B	A vs C	B vs C
Creatinine(mg/dl)	0.91±0.20	9.87±2.04	3.88±0.88	0.000	0.000	0.000
Zn (ppm)	65.47±17.8	61.27±16.2	66.63±19.31	0.705	0.973	0.567
Cu (ppm)	1.47±0.56	1.48±0.54	1.74±0.59	0.999	0.260	0.282
Mn (ppm)	4.87±1.67	1.26±0.44	1.46±0.51	0.000	0.000	0.834
Co (ppm)	7.84±2.52	4.30±1.79	4.59±2.43	0.000	0.000	0.930
Se (ppm)	9.07±1.85	4.04±0.98	5.64±2.60	0.000	0.000	0.021
Cr (ppm)	13.11±3.93	7.60±2.05	8.08±4.29	0.000	0.000	0.892

-A significant value was $p < 0.05$

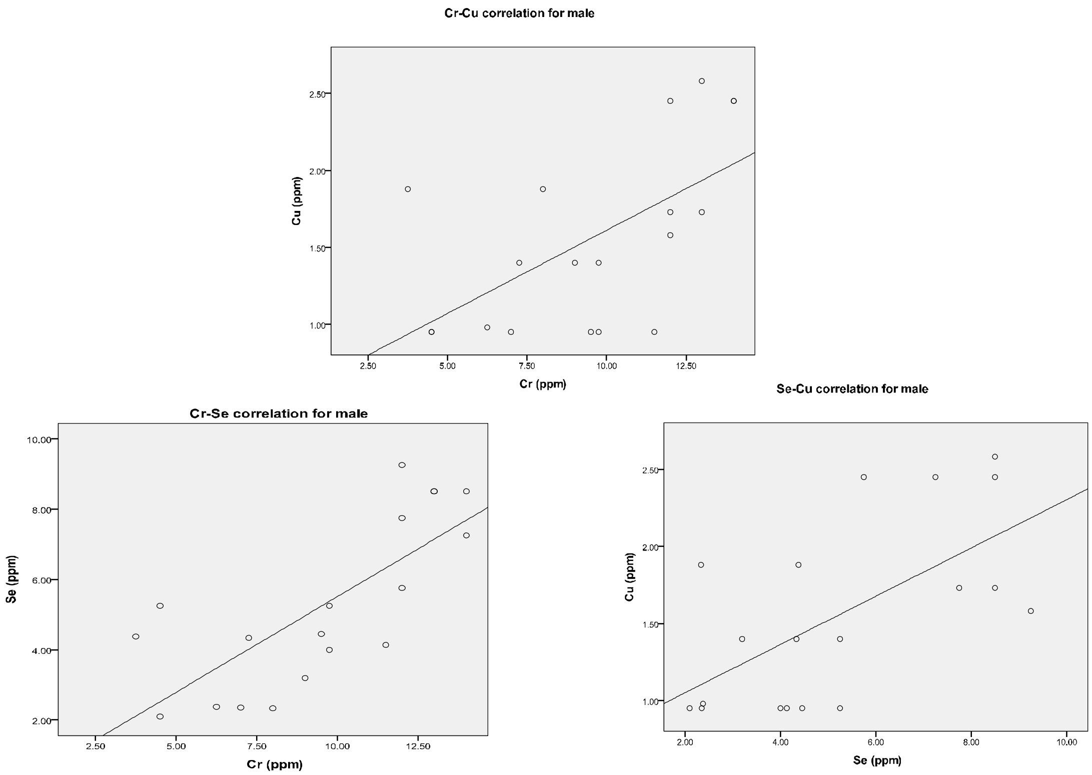

Figure 1: positive correlations between trace elements in male subjects.

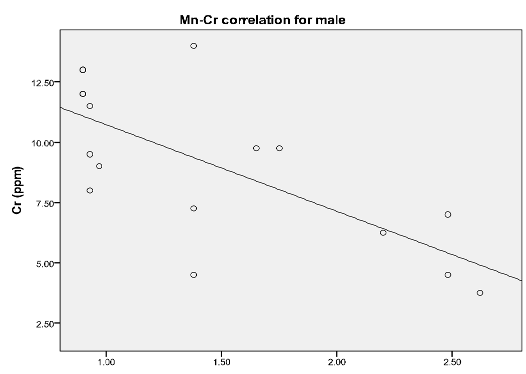

Figure 2: negative correlation between Cr and Mn in male subjects.

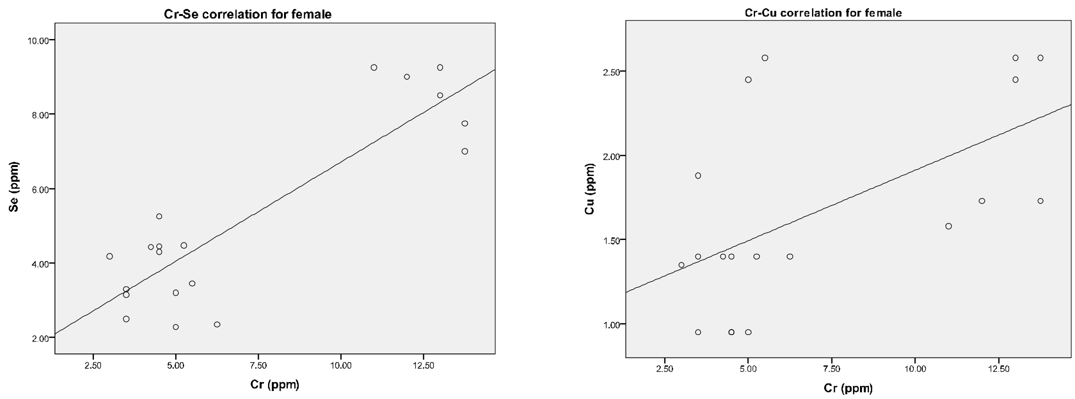

Figure 3: positive correlation between trace elements in female subjects.

4-Discussion

Chronic renal failure results in impaired renal excretion and accumulation of some trace elements in the body. During dialysis some trace elements can accumulate in the body because of dialysis fluid impurities and others may move from blood to dialysate leading to deficiency of these trace elements in the body **(Esfahani *et al.,* 2007)**. Also, the distribution of trace elements can be affected by insufficient dietary intake, degrees of gastrointestinal absorption, and drug treatment, and it can lead to some clinical abnormalities in uremic patients **(D'Haese and De Broe, 1996)**.

The trace elements status in chronic renal failure patients may also be influenced by residual renal function, the dialyzer membrane and its size, the nature of the water used for preparation of dialysate, the method of purification and the composition of dialysis concentrate **(Gault *et al.,* 1991)**.

The present study revealed that there was no significant difference in Zn level between controls and the hemodialysis male and female patients. In addition, there was no significant difference in Zn level between pre- and post-hemodialysis Patients. These results were in accordance with the data reported by **Sen *et al.* (1991), Kaminska-Galwa *et al.* (1994) and Koca *et al.* (2010)** who found no significant differences between HD groups or between HD groups and the control group. This can be attributed to the fact that the studied subjects in the present work had similar regional changes and dietary habits. Also, soil and water contamination affects trace element levels. These results can be attributed also to the fact that Zn in the blood is complexed with albumin forming a large particle unable to pass through the pores of dialysis membranes.

Previous reports have indicated a significant decrease in plasma zinc concentration in hemodialysis patients as compared with healthy controls **(Cabral *et al.,* 2005 and Hsieh *et al.,* 2006)**. Other old reports revealed that HD increased serum Zn level **(Atlihan *et al.,* 1991 and Lin *et al.,* 1996)**.

The results illustrated that there was no significant difference in Cu level between controls and the hemodialysis patients. These results are in agreement with the results obtained by **Hsieh *et al.* (2006)** in adult patients and **Shouman *et al.* (2009)** in pediatric HD patients.

Our results are contradicted with previous studies that reported that Cu was significantly deficient in hemodialysis patients **(Weissgarten *et al.,* 2001 and Anees *et al.,* 2011)**, while **Lin *et al.* (1996) and Alarcon *et al.* (2005)** observed a significant increase in Cu level in HD patients.

There was no significant difference in Cu level between pre and post-hemodialysis Patients in the present study. These results are in agreement with the result of **Tetiker *et al.* (1993)**.

On the other hand the data showed that Mn level was significantly decreased ($p<0.01$) in male and female patients either pre- or post-hemodialysis compared to control. Also, there was non-significant change in Mn level between pre- and post-hemodialysis. These results are in agreement with **da Silva *et al.* (2007)** who suggested the reason is thought to be removal of manganese from blood during hemodialysis. It, thus, theoretically is reasonable that manganese intoxication would be unlikely in patients undergoing hemodialysis if the dialysate is not severely contaminated by manganese.

The results also revealed that there was a significant decrease in the level of Co in male and female HD group compared to control group. Non significant increase was obtained between pre and post- hemodialysis Patients. On the other hand

Salvadeo *et al.* (1979) reported a statistically significant increase in blood Co concentrations comparing pre- and post-dialysis values in six HD patients. **Lins and Pehrsson (1984)** reported that uremic patients have elevated serum Co values and that HD treatment does not reduce but rather increases the serum Co concentrations.

Selenium level was significantly lower in male and female HD patients than control group. Similar results have been reported in previous studies **(Yavuz *et al.*, 2004; Hsieh *et al.*, 2006).** These findings seemed to be justifiable because in human among other organs, the kidney contains the highest level of selenium **(Zachara *et al.*, 2001)** and plays an important regulatory role in homeostasis of the element **(Robinson *et al.*, 1985)**. Renal failure itself may have some influence on selenium concentration probably through decreased absorption in the small intestine **(Bonomini *et al.*, 1996)**. Moreover, a low dialysate concentration of selenium may result in its removale during HD with resulting in deficiency **(D'Haese *et al.*, 1996)**.

Data obtained concerning selenium levels in pre- and post-hemodialysis patients are consistent with those of **(Bogye *et al.*, 2000)** who suggested that there was no significant difference between serum selenium concentration in male and female subjects before and after HD session. In order to clarify the selenium lost, it may be useful to analyze the levels of selenium in dialysis fluid in addition to that in blood samples.

Data also revealed that there was a significant decrease in the level of Cr in HD group compared to control group. On the other hand **Tonelli *et al.* (2009)** reported that level of Cr was high in HD patients compared to control. The study concluded that plasma trace element concentrations in HD patients are distinctly different compared to that of healthy controls. Elements such as Mn, Co, Se and Cr were reduced in HD patients, while Cu and Zn were not affected. Regular monitoring of trace elements in hemodialytic patients is advisable.

Refrences

1. **Alarcon, N. M.; Pérez, R. A.; Garcia, L. H.; Bayo,P. M.; Herrera, O. M. and Martinez, L. M. C. (2005):** Longitudinal study of serum zinc and copper levels in hemodialysis patients and their relation to biochemical markers. Biological Trace Element Research, 113 (3):209-222.
2. **Anees, M.; Mumtaz, A.; Frooqi, S.; Ibrahim, M. and Hameed, F. (2011):** Serum trace elements (Aluminum, Copper, Zinc) in haemodialysis patients. Biomedica., 27:106-110.
3. **Atlihan, F.; Soylemezoglu, T.and Devecioglu, C. (1991):** Serum zinc and copper in chronic renal failure. In: Trace Elements in Health and Disease (Trace '89), Yuregir, G.T.; Donma, O.; Kayrin, L. (eds.). Adana: Cukurova University Medical Faculty Press, pp 517-522.
4. **Babson, L.A.; Greeley, S.J.;Coleman, C.M.and Philips, G.D. (1966):** Serum alkaline phosphatase determination. Clin. Chem., 12: 482-490.
5. **Bogye, G.; Tompos, G. and Alftan, G. (2000):** Selenium depletion in hemodialysis patients treated with polysulfone membranes. Nephron., 84: 119-123.
6. **Bonomini, M.;Foster, S.; Manfrini, V.; De Risio, F.; Stiner, M.;Vidovich, M.J.; Klinkmann ,H., Ivanovich, P. and Albertazzi, A. (1996):** Geographic factors and plasma selenium in uremia and dialysis. Nephron., 72: 197-204.
7. **Cabral, P. C.; Diniz, A. D. S. and De Arruda, I .K. J. (2005):** Vitamin A and zinc status in patients on maintenance haemodialysis. Nephrology, 10: 459-463.
8. **da Silva, C.J.; da Rocha, A.J.; Jeronymo, S.; Mendes, M.F.; Milani, F.T.; Maia, Jr, A.C.M.; Braga, F.T.; Sens, Y.A.S. and Miorin, L.A. (2007):** A Preliminary Study Revealing a New Association in Patients Undergoing Maintenance Hemodialysis: Manganism Symptoms and T1 Hyperintense Changes in the Basal Ganglia. AJNR Am J Neuroradiol, 28:1474-1479.
9. **D'Haese, P. C. and De Broe, M. E. (1996):** Adequacy of dialysis: Trace elements dialysis fluids. Nephrol. Dial. Transplant 11(suppl2): 92-97.
10. **El-Sherbeny, S.S.A.; Abd Allah, A.M. and Massoud,W. (2006):** Copper, Zinc, Selenium, Cadmium and Lead Levels in plasma of Hemodialysis Patients. The Egyptian Journal of Hospital Medicine, 24: 556 – 563.
11. **Esfahani, S.T.; Hamidian, M.R.; Madani, A.; Ataei, N.; Mohseni, P.; Roudbari, M.; Hadjizaeh, N. and Haddadi, M. (2007):** Serum trace elements in children on maintenance hemodialysis. Acta Medica Iranica., 45(5): 351-354.
12. **Gault, M. H., Longerich, L. and Longerich, H. (1991):** Trace elements and hemodialysis. Nephron., 58: 498-499.
13. **Hashmi, D.R.; Ismail, S. and Shaikh, G.H. (2007):** Assessment of the level of trace metals in commonly edible vegetables locally available in the markets of karachi city. Pak. J. Bot., 39(3):747-751.
14. **Henry, R.J.; Chiamori, N.; Golub, O.J. and Berkman, S. (1960):** Revised spectrophotometric methods for determination of glutamic-oxalacetic transaminase, glutamic-pyruvic transaminase, and

lactic dehydrogenase. Am. J. Clin. Path., 34:381–398.

15. **Hsieh, y.; Shin, W.; Lee, L.; Wu, T.; Ning, H. and Sun, T. (2006):** Long-term changes in trace elements in patients undergoing chronic hemodialysis. Biol. Trav. Elem. Res., 109: 115-121.
16. **Kaminska-Galwa, B.; Grzeszczak, W.; Jedryczko, A. and Pachelski, J. (1994):** Influence of long-term hemodialysis on serum trace elements concentration in patients with chronic renal failure. Przegl Lek., 51:9-14.
17. **Koca, T.; Berber, A.; Koca, H.B.; Demir, T.A. and Koken, T. (2010):** Effects of hemodialysis period on levels of blood trace elements and oxidative stress. Clin Exp Nephrol., 14:463–468.
18. **Krachler, M.; Wirnsberger, G. and Irgolic, K.J. (1997:** Trace element status of hemodialyzed patients. Biol Trace Elem Res., 58:209–221.
19. **Lin, T.H.; Chen, J.G.; Liaw, J.M. and Juang, J.G. (1996):** Trace elements and lipid peroxidation in uremic patients on hemodialysis. Biol Trace Elem Res., 51(3):277-283.
20. **Lins, L.E. and Pehrsson, S.K. (1984):** Cobalt in serum and urine related to renal function. Trace Elem Med., 1:172–174.
21. **Malloy, H. T. and Evelyn, K. A. (1937):** the determination of bilirubin with the photoelectric colorimeter. J. Biol. Chem., 119: 481-490.
22. **Miura, Y.; Nakai, K.; Suwabe, A. and Sera, K. (2002):** Trace elements in renal disease and hemodialysis. Nuclear instruments and methods in Physics Research, 189: 443-449.
23. **Pinnell, A. E. and Northam, B. E. (1978):** New automated dye- binding method for serum albumin determination with bromcresol purple. Clin. Chem., 24:80-86.
24. **Robinson, J. R.; Robinson, M. F.; Lavander, O.A. and Thomson, C.D. (1985):** Urinary excretion of selenium by New Zealand and North American human subjects on different intakes. Am. J. Clin. Nutr., 41:1023-1031.
25. **Ryan, M.T. and Chopra, R.K. (1976):** The paradoxical effect of fatty acid on steroid-albumin interaction. Biochim Biophys Acta., 427:337-349.
26. **Salvadeo, A.; Minoia, C.; Segagni, S. and Villa, G. (1979):** Trace metal changes in dialysis fluid and blood of patients on hemodialysis. Int J Artif Organs, 2:17–21.
27. **Sen, S.; Bor, N.; Colakoglu, M. and Gultekin, A. (1991):** Clearance of zinc and copper during hemodialysis. Journal of Islamic Academy of Sciences, 4: 3, 265-267.
28. **Shouman, M.G.; Ibrahim, H.Y.; Salama, E.E.E.; El-Khayat, Z. and Ashour M. (2009):** Trace Elements in Pediatric Hemodialysis Patients. Res. J. Medicine & Med. Sci., 4(2): 435- 440.
29. **Tetiker, T.; Paydas, S.; Yuregir, G. and Sagliker, Y. (1993):** Trace elements alterations in chronic hemodialysis patients with chronic renal failure and proteinuria. Journal of Islamic Academy of Sciences, 6: 1, 33-35.
30. **Tonelli, M.; Wiebe, N.; Hemmelgarn, B.; Klarenbach S.; Field, C.; Manns, M.; Thadhani, R. and Gill, J. (2009):** for the Alberta Kidney Disease Network: Trace elements in hemodialysis patients: a systematic review and meta-analysis. BMC Medicine, 7: 1-25.
31. **Vanholder, R.; Cornelis, R.; Dhondt, A. and Lameire, N. (2002):** The role of trace elements in uraemic toxicity. Nephrol DialbTransplant., 17(2):2-8.
32. **Vasiliades, J. (1976):** Reaction of alkaline picrate with creatinine. Kinetics and mechanism of formation of the mono-creatinine picric acid complex. Clin Chem., 22:1664-1671.
33. **Weissgarten, J.; Berman, S.; Bilchinsky, R.; Modai, D. and Averbukh, Z. (2001):** Total Cell-Associated Zn and Cu and Proliferative responsiveness of Peripheral Blood Mononuclear Cells from Patients on Chronic Hemodialysis. Metabolism, 50 (3): 270-276.
34. **Yavuz, O.; Bicik, Z.; Cinar, Y.; Guney, Y. and Guler, S. (2004):** The effect of different dialysis memberanes on oxidative stress and selenium status. Clin. Chem. Acta., 364 (2): 153-160.
35. **Yilmaz, M.E.; Kiraz, M. and Kara, I.H. (2000):** The Evaluation of Serum Zinc and Copper Levels in Hemodialysis Patients in Southeast Turkey. Dialysis and Transplantation, 29(11):718-720.
36. **Zachara, B.A.; Pawluk, H.; Bloch-Bogustawska, E.; Sliwka, K.M.; Korenkiewicz, J.; Skok, Z. and Ryć, K. (2001):** Tisuue level distribution and total body selenium content in healthy humans and in some diseases in Poland. Arch. Environ. Health, 56: 461-465.

08/12/2012