Chaotic Modeling and Simulation (CMSIM) **3: 151-160, 2019**

Toxicological evaluation of the Copper in chronic insufficient subjects, in vivo investigations by blood tests

Dr N. Sahraoui[1] & Dr H. Benabid[2] & A. Brahimi[3]

1-Institute of Health and Industrial Safety University Batna 2; LRPI Laboratory
2- Institute of Health and Industrial Safety University Batna 2; LRPI Laboratory
3- Hematology laboratory of University hospital center Batna

Abstract:
Renal insufficiencies are the result of chronic intoxications due to repeated and prolonged exposures to multiple harmful substances, in addition to unbalanced diet in addition to the consumption of tobacco, alcohol etc ...
All these factors lead to the onset of chronic renal insufusance disease which necessitates permanent hemodialysis by artificial ankle which carries out the purging of blood from the body through an artificial membrane based on copper.
With the aim of verifying the effect of copper in chronically deficient subjects, we have tried, to make a toxicological evaluation of these subjects, and to compare them with the results of these studies. They have a great deal of patience because they spend an impressive amount of time (four hours per session).

Keywords: Toxicological evaluation, copper, insuficient subjects, investigations, blood.

Introduction:

The kidney is a vital organ whose function is the purification of blood from urea; it happens that certain risk factors such as diabetes, hypertension, heart disease etc can cause degenerative

Received: 18 July 2019 / Accepted: 31 July 2019

ISSN 2241-0503

effects on the kidney and the kidney reaches the terminal stage: dialysis.
It is in order to evaluate the impact of dialysis on the health of the dialysis patient; that I try to evaluate the risk of cuprophane membrane on the health of the selected patients: this original idea aims to estimate the copper content in the serum and plasma of patients.

I-Kidney function:

The kidney normally plays an important role in the metabolism of AGEs, which
undergo glomerular filtration followed by uptake by tubular cells where they are metabolized. Most of the circulating pentosidine being linked to high molecular weight proteins such as albumin, dialysis has little effect on the concentration of AGEs. AGEs can play
a role in activating mononuclear cells, thereby directly triggering an
inflammatory response [15]. However, the inflammation itself can reciprocally
play a role in the production of AGEs .

II-Presentation of chronic renal failure:

II.1 The stages of the IRC:

Chronic renal failure is manifested in four stages:
Stage 1: This stage is manifested by a decrease of the renal reserve.
Stage 2: Considered as the beginning of renal insufficiency proper because the kidney keeps with difficulty his excretory functions and regulatory functions, there is a moderate hyperazotemia accompanied by a slight anemia.

Stage 3: Stage of major renal failure, elevated azotemia is accompanied by acidosis and hypocalcemia, hyperkalemia is common and anemia is often severe.
Stage 4: This is the stage of uremia where terminal evolution.

II.2 Treatment of chronic renal failure:

Treatment of chronic renal failure can be a medical treatment up to the point where the treatment is ineffective, and treatment with periodic extra-renal cleansing or renal transplantation becomes necessary, in case of contraindication of kidney transplant or the impossibility of finding a donor; hemodialysis and the only chance for chronic kidney failure to survive.

II.2.1 Artificial filtration process;
Hemodialysis by artificial kidney carries out the purification of the blood out of the body, the exchange between this blood is a dialysis bath which is carried out through an artificial membrane.
The artificial kidney or hemodialyzer is punctured on the blood exit path from an artery or vein and one entry into another.

III. Analytical part:

III.1 Sampling procedure:
The first stage of the analysis is the most impressive because it consumes the double samples (serum and plasma)
The samples taken at the hemodialysis resuscitation department CHU Batna.
The blood taken before dialysis undergoes a centrifugation to recover the serum.
During dialysis, an anticoagulant is added to the blood to which it is subjected to centrifugation to obtain plasma.

The same mode of preparation is carried out for the control samples.

III.2 picking samples:

Samples are taken directly from a sample of eight patients before and after the dialysis operation
* before the dialysis operation the eight serum samples are taken
* after the dialysis operation we take the eight samples of Plasma.

III.3 Sampling conditions:
The sampling is done in the morning to fast to avoid the nyctemerales variations, to avoid the emotions, to reassure the patient and to place it in a comfortable place to manage the influences on the rate of the glucose, the phosphate, the fatty acids esterified of the plasma under the influence of adrenaline

III.3.1 Separation
Blood must be centrifuged as quickly as possible.

NOTE: For practical reasons our choice is fixed for eight patients who have undergone regular dialysis for five years.

IV. Material resources and methods of calculation:
Atomic absorption spectroscopy is extensively applied in qualitative and quantitative analyzes, based on the calibration curve, which is based on the determination of the concentrations of the analyzed element from the absorbance; the line is based on the beer-lambert law.

Least squares correction: It is used to correct any anomalies made on the calibration curves.

V. Results and interpretations:

Table 1 : copper standards solutions

[ppm]	0	1	2	3	4	5
Abs	00.00	0.033	0.060	0.107	0.126	0.176

Table 2 : Determination of absorbances of plazma samples by SAA

Samples	S1	S2	S3	S4	S5	S6	S7	S8
Abs	0.004	0.007	0.004	0.002	0.003	0.004	0.002	0.002

Table 3 : Determination of Absorbance of Serum Samples by SAA:

Samples	S1	S2	S3	S4	S5	S6	S7	S8
Abs	0.007	0.004	0.006	0.006	0.003	0.002	0.003	0.002

Table 4: Concentrations of copper in diseased subjects (Plasma)

Samples	Absorbances	Concentrations (ppm)
Sample 1	0.004	1.30
Sample 2	0.007	1.60
Sample 3	0.004	1.50
Sample 4	0.002	0.90
Sample 5	0.003	1.56
Sample 6	0.005	1.60
Sample 7	0.002	1.20
Sample 8	0.002	1.45

Table 5: Concentrations of Copper in Diseased Subjects (Serum)

Samples	Absorbances	Concentrations (ppm)
Sample 1	0.007	2.67
Sample 2	0.004	1.17
Sample 3	0.006	1.76
Sample 4	0.006	1.76
Sample 5	0.003	0.88
Sample 6	0.002	0.58
Sample 7	0.003	0.88
Sample 8	0.002	0.68

Table 6: Concentartions of Copper in Control Subjects (Plasma and Serum)

Samples	$[Cu^{2+}]$ in Serum (ppm)	$[Cu^{2+}]$ in Plasma (ppm)
Sample 1	1.11	1.12
Sample 2	1.06	0.80
Sample 3	0.88	0.84
Sample 4	0.90	0.84
Sample 5	0.46	0.42
Sample 6	0.70	0.56
Sample 7	0.94	0.68
Sample 8	0.42	0.67

VI.1 Interpretation of results:
According to the results obtained during the investigation it was concluded that:

Before dialysis (Serum):
Values are higher than standard (1.7 to 2.67 ppm)
Three values are in the standards (0.8 to 1.17 ppm)
Two values are below standard (0.5 to 0.67ppm) See Tables of results .

After dialysis (Plasma):
Six values are greater than the standard, ranging from (1.45 to 1.60 ppm)
Two values are normal, ranging from (0.92 to 1.2 ppm) See Tables of results .
According to the results of the investigation it was possible to note by comparison of the results in the sick subjects and the control subjects in the case of the Serum, that the sick subjects that five values exceed the norm which is of the order of [0.70-1.4]; however, in control subjects, all concentrations are normalized.
With regard to the results of the concentrations of the sick and control subjects in the plasma, it is noted that the concentrations of all patients are raised by standards after the dialysis operation, this

can be explained by the copper overload after the dialysis [4], this overload is due to the contamination of the membrane of the device as the fibers of the membrane are cuprophane, in addition to the diet that is not balanced [2].

The high level of copper concentrations in patients with CKD may be explained by the copper concentration of the blood after dialysis following the removal of water from the blood before dialysis [3]. This extra copper can kill liver cells and cause nerve damage.

In hemodialysis patients, the ionic copper can be released by semi permeable membranes made of copper or by copper tubes or heating coils of the dialysis device, particularly when the dialysate has become acidic [7]. In two hemodialysis patients, copper intoxication was characterized by marked hemolysis, acidosis, and methemoglobinemia. It can also be pointed out through these investigations that Acute-positive proteins include C-reactive protein (CRP), a protein associated with secondary amyloid formation, serum amyloid A protein (SAA), fibrinogen, members of the complement cascade, and protein carrier of copper, or ceruloplasmin [5] . Negative proteins in the acute phase include albumin, pre-albumin, transferrin, and apoliprotein AI (apo AI) [15] . While malnutrition, such as inflammation, causes a decrease in the latter group of proteins, it does not increase the level of positive proteins in the acute phase, which makes it possible to differentiate between these two conditions [8]. Bioincompatible membranes such as Cuprophane activate leucocytes [10] and complement [1] and may even have effects on residual renal function.Cytokine activation is observed during hemodialysis using Cuprophane® membranes [11], unlike biocompatible membranes [12, 13]. It can also be concluded that the use of the cuprophane-based membrane is at the origin of ceruloplasmin is a copper-carrying protein that can oxidize both HDL and LDL. SAA moves apo AI from HDL. During inflammation, the activity of PAF-AH and paraoxonase is reduced, leading to an inability of HDL to reduce HDL oxidation [9] . In addition to the effect of

ceruloplasmin, inflammation also causes oxidation of LDL by the action of leukocyte myeloperoxidase [6].

Conclusion:

At the end of our investigation based essentially on a toxicological evaluation of the copper element in the subjects with chronic renal insufficiency, it appears that: the concentrations of the copper detected in the plasma and the serum at the patients are not similar, and that the concentrations of the copper element are very high in the subjects taken as study samples, in addition the potassium level is high by contribution to the control subjects.

These data suggest that the state of health of the patients can worsen according to the dialysis sessions by the mechanism of gradual accumulation, if corrective measures do not follow and more particularly of medical order, studied diet of on the one hand and instrumental on the other.

Bibliography

[1]-Anzi RE, Petrukhin K, Chernov I, et al. (1993). "The Wilson disease gene is a copper transporting ATPase with homology to the Menkes disease gene". Nat. Genet. 5 (4): 344–50

[2]-Balamurugan K, Schaffner W (2006) Copper homeostasis in eukaryotes: teeteri tightrope. Biochim Biophys Acta 1763(7):737–746

[3]- Chambers A, Krewski D, Plunkett L (2010) An exposure-response curve for c excess and deficiency. J Toxicol Environ Health B Crit Rev 13(7–8):546–578

[4]-Christodoulou J, Danks DM, Sarkar B, Baerlocher KE, Ca-sey R, Horn N, Tümer Z, Clarke JT. Early treatment of Menkes

disease with parenteral copper-histidine: long-term follow-up of four treated patients. Am. J. Med. Genet. 1998;76(2):154–164

[5]-Cobine PA, Pierrel F, Winge DR. Copper trafficking to the mitochondrion and assembly of copper metalloenzymes. Biochim Biophys Acta. 2006;1763:759–72. doi: 10.1016/j.bbamcr.2006.03.002

[6]- Culotta, V.C. and Gitlin, J.D.: Disorders ofcopper transport. In The Metabolic andMolecular Bases of Inherited Disease,(ed.Scriver et al.) 8th ed., vol II, McGraw Hill,New York, 2000; pp. 3105–3126.

[7]-Guclu BK, Kara K, Beyaz L, Uyanik F, Eren M, Atasever A. Influence of dietary copper proteinate on performance, selected biochemical parameters, lipid peroxidation, liver, and egg copper content in laying hens. Biol Trace Elem Res. 2008;125:160–9. doi: 10.1007/s12011-008-8164

[8]-8-La Fontaine S, Mercer JF. Trafficking of the copper-ATPases, ATP7A and ATP7B: role in copper homeostasis. Arch. Biochem. Biophys. 2007;463(2):149–167

[9]-8-Manser WW, Khan MA, Hasan KZ. Trace element studies on Karachi population. Part IV: blood copper, zinc, magnesium and lead levels in psychiatric patients with depression, mental retardation and seizure disorders. J Pak Med Assoc. 1989;39:269–274

[10]-Müller T, van de Sluis B, Zhernakova A, et al. (2003). "The canine copper toxicosis gene MURR1 does not cause non-Wilsonian hepatic copper toxicosis". J. Hepatol. 38 (2): 164–8

[11]-Sendovski M, Kanteev M, Ben-Yosef VS, Adir N, Fishman A. First structures of an active bacterial tyrosinase reveal copper

plasticity. J Mol Biol. 2011;405:227–37. doi: 10.1016/j.jmb.2010.10.048

[12]-Shaver WA, Bhatt H, Combes B (1986). "Low serum alkaline phosphatase activity in Wilson's disease".

[13]-Suttle N, Mills C. Studies of the toxicity of copper to pigs. Br J Nutr. 1966;20:149–61

[14]-Turnlund JR. Human whole-body copper metabolism. Am. J. Clin. Nut 1998;67(Suppl):S960–S964.

[15]-Van De Sluis B, Rothuizen J, Pearson PL, van Oost BA, Wijmenga C. Identification of a new copper metabolism gene by positional cloning in a purebred dog population. Hum. Mol. Genet. 2002;11(2):165–73.