Biospectra : ISSN: 0973-7057, Vol. 15(2), September, 2020, pp. 167-172
An International Biannual Refereed Journal of Life Sciences

34(AS).
Animal Science

ISSN : 0973-7057

Int. Database Index: 663 www.mjl.clarivate.com

Effect of Calcium Hypochlorite and Sodium Hypochlorite on the Glomerulus and Bowman's Capsule of Mice

Joyita Das[a]* & Doris D' Souza[b]

[a]P. C. Vigyan College, J.P.University, Chapra, Bihar, India

[b]Patna Women's College, Patna University, Patna, Bihar, India

Received : 25th April, 2020 ; Revised : 24th May, 2020

Abstract-The effect of chlorine salts calcium hypochlorites and sodium hypochlorite commonly used for disinfection of water were seen in Swiss albino mice. Mice were given oral doses of 50, 100, 150 and 200 ppm per kg of body weight per day. Significant changes were observed in the glomerulus and Bowman's capsule after four months treatment with 200 ppm oral dose of both the chemicals. Changes were evident even in lower doses and duration. The decrease in size was significant in both glomerulus and Bowman's capsule at $P < 0.05$ for the highest dose of 200 ppm and longest duration of four months for both calcium hypochlorite and sodium hypochlorite.

Keywords : Calcium hypochlorite, sodium hypochlorite,kidney,mice.

INTRODUCTION

Water used for drinking has to be disinfected prior to use. According to Krenkel and Novotny(1980)[1] water borne disease causes some 10 million deaths per year and 500 million cases of illness in the developing nations. The disinfection process may be either physical or chemical. Physical processes involve boiling, reverse osmosis, ultraviolet radiation treatment etc. which are costly and usually not undertaken for public or community supply. Chemical processes like chlorination, ozonation etc. are prominent. Ozonation also has mutagenic effects as per Cotruvo *et al.* (1977)[2]. Chlorination is a common practice in many parts of the world. It requires the addition of chlorine or its compound in water. The commonly used process is to add chlorine salts like calcium hypochlorite or sodium hypochlorite. But if it exceeds the chlorine demand or if organic impurities are present in water it may be toxic to the body. The possible involvement of chlorine in the etiology of heart disease has also been suggested. Benditt (1974)[3] provided clinical evidence to support a link between plaque formation in the arteries with chlorinated water. Chemical substances that are ingested undergo detoxification, metabolism and excretion with the liver and kidneys being mainly involved as explained by Makita *et al.* (2000)[4]. The water industry concern stems from the proven link between widely used disinfection methods and drinking water organic level.[5]

*Corresponding author :
Phone :
E-mail : @gmail.com

Organic impurities present in water react with chlorine salts to form harmful bi-products which are mutagenic and carcinogenic.The kidney is a target for toxic chemicals besides the liver as it is the major organ for excretion and homeostasis for water-soluble molecules. It can concentrate certain substances actively. These properties of the kidney make it susceptible to certain chemicals which may prove toxic to it. Even Khalil-Manesh *et al.* (1992)[6] have ascertained these aspects of the vulnerability of the kidney to toxic chemicals. Ibrahim *et al.* (2018)[7] have reported mortality besides histopathological changes in the in the kidney, spleen and thymus of mice exposed to gold nanoparticles. Peck *et al.* (2014)[8] have shown acute kidney injury induced by sodium hypochlorite often used as a dental irrigant in humans.

Therefore the this work was undertaken to ascertain the toxicity of chlorine salts to the kidney of mice and extrapolate it to that of the human beings since they are exposed to the chlorine salts or its harmful bi-products through drinking water.

METHODOLOGY

Swiss albino mice were taken as the test animal. Male mice in the age group of 3-4 months were taken with an average weight of 32.5±2.0 gm. Two groups each consisting of ten mice for each dose were taken. One group was treated and the other corresponding control group was not treated i.e. not administered the dose of the chlorine salts. They were given equal amount of distilled water. The mice were given oral doses of 50, 100, 150 and 200 ppm of calcium hypochlorite and sodium hypochlorite, each, orally through gastric tubes. Corresponding control group were maintained for the respective doses and duration. Both the groups, treated as well as control were sacrificed at the end of one, three and four months. The kidney was cut into pieces and preserved in Bouin's fixative. Blocks were prepared and sections were cut. These were stained using standard HE (Hematoxylin-Eosin)stain. The stained sections were then examined under the microscope. The changes in the structure of the treated ones as compared to the control were analysed. The dimensions of the different subdivisions of the kidney were taken with the help of ocular micrometer and stage micrometer. Twenty observations were taken randomly throughout the experimental sets. Then the average values were calculated and subjected to statistical analysis. Standard deviation was calculated. The diameter of the cup of Bowman's capsule and glomerulus were analyzed taking its mean± standard deviation. The data were subject to student's t-test (taking the differences of the two means of independent samples). A 5% level of significance ($P<0.05$) was used throughout the analysis.

RESULTS AND DISCUSSION

Chemical insult to the kidney may involve several changes. The dimensions of the different subdivisions of the kidney are prime among them. The Bowman's capsule and the glomerulus are basically involved in ultrafiltration or filtration under immense pressure. Any toxin therefore leads to substantial changes in their dimensions besides structural changes. The changes in dimensions of the glomerulus for different doses and durations of calcium hypochlorite and sodium hypochlorite were as follows:

Table 1.Showing diameter of glomerulus (in mm)in kidneys of mice treated orally with calcium hypochlorite and sodium hypochlorite.

Duration→ Dose ↓	1 month	2 months	3 months
Control	0.1072871 ±0.0013416	0.0983091 ±0.0013416	0.0938201 ±0.00134
50 ppm Calcium hypochlorite	0.1005536 ±0.0004466	0.0821487 ±0.0108166	0.080802 ±0.109909
100 ppm Calcium hypochlorite	0.1072871 ±0.0013416	0.076313 ±0.0194627	0.0834954 ±0.102547
150 ppm Calcium hypochlorite	0.085291 ±0.0120415	0.0758641 ±0.0201171	0.848421 * ±0.0148087
200 ppm Calcium hypochlorite	0.0816998 ±0.0082643	0.0929223 * ±0.009423	0.070477 * ±0.0156843
50 ppm Sodium hypochlorite	0.0915756 ±0.0078166	0.0816998 ±0.008264	0.0830465 ±0.010281
100 ppm Sodium hypochlorite	0.0861888 ±0.0121737	0.0700284 ±0.0236833	0.0754152 ±0.16155
150 ppm Sodiumhypochlorite	0.1014514 ±0.0057358	0.0754152 ±0.0248314	0.0834954* ±0.102547
200 ppmSodiumhypochlorite	0.0902289 ±0.0110724	0.0664365 * ±0.02045	0.0682328 * ±0.0534097
*P< 0.05			

Table 2. Showing the diameter of the Bowman's capsule (in mm) of the kidneys of mice treated orally with Calcium hypochlorite and Sodium hypochlorite.

Duration→ Dose ↓	1 month	2 months	3 months
Control	0.1207541 ±0.0013416	0.1306599 ±0.0013416	0.1351189 ±0.0013416
50 ppm Calcium hypochlorite	0.1108783 ±0.0066708	0.1185096 ±0.0040987	0.1242453 ±0.0131643
100ppm Calcium hypochlorite	0.107736 ±0.0114324	0.1162651 ±0.0099247	0.1144695 ±0.020199
150 ppm Calcium hypochlorite	0.1019003 ±0.0137658	0.1216519 ±0.0110724	0.1095316 ±0.0040987
200 ppm Calcium hypochlorite	0.1005536 ±0.0090166	0.1144687 ±0.0.0067305	0.1229986* ±0.017977
50 ppm Sodium hypochlorite	0.1104294 ±0.0064031	0.1158162 ±0.0082219	0.1279365 ±0.009252
100 ppm Sodium hypochlorite	0.1086338 ±0.0095812	0.1153673 ±0.0069641	0.1274876 ±0.0094498
150 ppm Sodium hypochlorite	0.1001047 ±0.0128569	0.1140206 ±0.0085556	0.1252431 ±0.011606
200 ppm Sodium hypochlorite	0.1149184 * ±0.008792	0.1090827 ±0.0072456	s0.1279368 * ±0.0367219
*P<0.05			

The above table shows that the different oral doses of calcium hypochlorite and sodium hypochlorite have profound effect on the dimensions of glomerulus and Bowman's capsule. Since the changes in the diameter of the glomerulus and Bowman's capsule serves as an important parameter indicating the toxicity of these two chemicals. There were significant decrease in the diameter of the glomerulus with the doses 150 ppm per day for four months and 200 ppm per day of treatment for three and four months with calcium hypochlorite and sodium hypochlorite at $P<0.05$ (Table 1).

Similarly there were significant changes in the diameter of the Bowman's capsule on treatment with 200 ppm per day for four months treatment with calcium hypochlorite and sodium hypochlorite as well (Table 2). These changes indicate the stress induced by the two chemicals on the glomerulus and Bowman's capsule of the kidney. In the control or untreated mice i.e. mice not administered with the chlorine salts the diameter of the glomerulus was found to be varying between 0.0938201 mm ±0.0013416 after four months duration. In them there is a gradual increase in the diameter. As compared to that on treatment with 150 ppm calcium hypochlorite for four months the diameter of the glomerulus has decreased significantly to 0.0848421 mm ±0.0148087 at $P<0.05$ (Table 1). Similarly the dose of 200 ppm calcium hypochlorite for four months caused decrease of the diameter to 0.070477mm ±0.0156843 which was also quite significant at $P<0.05$(Table 1). The changes were even significant after treatment with 200 ppm per day for three months only. The changes induced by oral treatment with sodium hypochlorite caused still greater decrease in the diameter of the glomerulus. There was significant decrease after four months oral treatment of 150 ppm per day of sodium hypochlorite. The diameter of the glomerulus decreasedto 0.0834954 mm ±0.102547 and with 200 ppm per day treatment for four months to 0.0682328 mm ± 0.0534097 as compared to that of the control which was 0.0938201 mm ±0.0013416 (Table 1). Therefore, these significant changes in the diameter of the glomerulus indicate the irreversible damage induced to the structure of the kidney by the two chemicals calcium hypochlorite and sodium hypochlorite. The dynamics of the glomerular ultrafiltration makes it susceptible to the increasing load of the chemicals according to Brenner *et al.* (1971)[9]. The hypochlorite's potential to cause toxicity is related to its oxidizing capacity and the pH of the solution as per the reviews of Slaughter *et al.* (2019)[10] on the toxic effects of sodium hypochlorite. Thus the chemical injury to the subdivisions of the kidney is quite evident. Ibrahim *et al.* (2018)[7] have shown the pathological changes in the form of diminished and distorted glomeruli in the kidney of mice treated with gold nanoparticles .

Similarly the changes induced on treatment with the two chemicals on the Bowman's capsule were also profound. Calcium hypochlorite caused significant changes after four months dose of 200 ppm per day. Whereas, the dose of 200 ppm of sodium hypochlorite caused significant changes in the diameter of the Bowman's capsule even after one month treatment as well as four months treatment. Thus sodium hypochlorite induced greater changes as compared to calcium hypochlorite. According to Estrella *et al.* (2002)[11], sodium hypochlorite combines with water to generate hypochlorous acid which in turn generates superoxide radicals, resulting in oxidative injury. It is this property therefore that is responsible for the toxic effect on the renal cells when used in excess of the required amount. Even Gernhadt *et al.* (2004)[12] have provided evidence that sodium hypochlorite is a cytotoxic agent. Hence it becomes increasingly important that the amount of chlorine salts added to the water for disinfection process should be strictly according to the requirement. Even potassium bromate used as food additive acts as a potent nephrotoxic agent and causes serious kidney damage as reported by *Bao et al.* (2008)[13]. Obliteration of the Bowman's space with irregularity of Bowman's membrane were also observed in male albino rats treated with antimony by Rashedy *et al.* (2013)[14]. The injury induced to the glomerulus by sodium oxalate has also been shown by deAraujo *et al.* (2020)[15]. Thus the glomerulus and Bowman's capsule are susceptible to injury by a vast array of chemicals. The chlorine salts are capable of inducing irreversible damage to the kidney as revealed by the changes in dimensions of the glomerulus and Bowman's capsule by the two chlorine salts calcium hypochlorite and sodium hypochlorite in Swiss albino mice. The chemicals in food and water prove to be toxic in most of the cases. Fahmy (2017)[16] has shown severe kidney damage induced by Aflatoxin B1 when administered through contaminated food. Even in the humans the dietary phthalates used in food packaging, flooring etc cause low grade albuminaria according to Trasande *et al.* (2014)[17]. Chemicals in food and water have the potential to cause great damage to the kidney.

CONCLUSION

The effect of sodium hypochlorite was greater as compared to calcium hypochlorite. Sodium hypochlorite caused damage even at a shorter duration. Substantial damage has been induced by calcium hypochlorite and sodium hypochlorite on both the glomerulusandthe Bowman's capsule of the kidney of Swiss albino mice. This study can be extrapolated to kidney of human beings. Further statistical data on the histopathological and biochemical aspects will definitely prove the damage induced by these two chemicals used in the disinfection of water. Since water is taken in at regular interval, therefore, if this water has excess of chlorine salts or its by products it has the capacity of causing damage to the kidney in the long run as seen in the glomerulus and Bowman's capsule of mice. Therefore, alternative methods of disinfection of water should be used such as reverse osmosis, commonly used in household purification system. Even for public systems, drinking water supply should be separated from water used for household chores.

REFERENCES

1. **Krenkel PA and Novotny V. 1980.** Water quality management. Academic Press, New York; 671.
2. **Cotruvo JA, Simmon VF and Spanggord RJ. 1977.** Investigation of mutagenic effects of products of ozonation reactions in water. *Annals of New York Academy of Sciences*. **298(1):** 124-140.
3. **Benditt EP. 1974.** Atherosclerosis may start with cell proliferation. *Journal of American Medical Association*. **227(7):** 734.
4. **Makita Y and Inoue N. 2000.** Acute hepatic and renal disorders. *Asian Medical Journal*. **43(7):** 337- 342.
5. **Page T, Harris RH and Epstein SS. 1976.** Drinking water and cancer mortality in Louisiana. *Science*. **193:**55-57.
6. **Khalil –Manesh F, Gonick HC, Cohen AH, Alinovi BE, Mutti Aand Rosen VJ. 1992.** Experimental model of lead nephropathy, continuous high dose lead administration. *Kidney International*. **41:** 1192- 1203.
7. **Ibrahim KE, Ghaleb M, Mutary A and Khan HA. 2018**. Histopathology of the liver, kidney and spleen of mice exposed to gold nanoparticles. *Multidisciplinary Digital Publishing Institute*. **23(8):** 1848.
8. **PeckBW, Workeneh B, Kadikoy H, Abdellatif A. 2014**. Sodium hypochlorite induced acute kidney injury; *Saudi Journal of Kidney Disease and Transplantion*. **25:** 381- 384.
9. **Brenner BM, Troy JL and Daugharty TM. 1971**. The dynamics of glomerular ultrafiltration in the rat. *Journal of Clinical Investment*. **50:** 1776-1780.
10. **Slaughter RJ, Watts M, Allister V, Grieve JR, Schep L J. 2019.** The Clinical Toxicology of Sodium Hypochlorite. *Clinical Toxicology* (*Philadelphia*). **57(5):** 303-311.
11. **Estrela C, Estrela CR, Barbin EL, Spano JC, Marchesan MA, Pecora JD. 2002.** *Brazilian Dental Journal*. **13:** 113- 117.
12. **Gernhardt CR, Eppendrof K, Kozlowski A and Brandt M. 2004.** Toxicity of sodium hypochlorite seed

as an endodontic irrigant. *International Endodontic Journal*. **37:** 272- 280.

13. **Bao L, Yao XS, Tsi D, Yau CC, Chia CS, Nagai H, Kurihara H. 2008.** Protective Effects of Bilberry (*Vaccinium myrtillus* L) Extract on KBrO? Induced Kidney Damage in Mice. *Journal of Agricultural and Food Chemistry*. **56(2):** 420-425.

14. **Rashedy AH, Solimany AA, Ismail AK, Wahdan MH and Saban KA. 2013.** Histopathological and functional effects of antimony on the renal cortex of growing albino rat. *International Journal of Clinical Pathology*. **6(8):** 1467- 1480.

15. **deAraujo L, Costa- Pessoa JM, dePonte MC, Oliveira-Souza. 2020.** Sodium oxalate induced acute kidney injury associated with glomerular and tubulointerstitial damage in rats. *Frontiers in Physiology*. **11:**1076.

16. **Fahmy A. 2017.** Histological and immuno histochemical study on the possible protective effects of curcumin and garlic against Aflatoxin B1 induced toxicity on the renal cortex of adult male guinea pig. *Journal of American Science.* **13(3):** 115-124.

17. **Trasande L, Sathyanarayana S and Trachtman H. 2014.** Dietary phthalates and low grade albuminaria in US children and adolescents. *Clinical Journal of the American Society of Nephrology*. **9(1):** 100- 109.

***