WORLD JOURNAL OF PHARMACEUTICAL RESEARCH
SJIF Impact Factor 8.084
Volume 9, Issue 14, 621-630. Research Article ISSN 2277– 7105

HISTOPATHOLOGICAL IMPACT OF ACETAMINOPHEN ON KIDNEY OF *OREOCHROMIS NILOTICUS* (NILE TILAPIA)

[1]*Dr. Rema L. P. and [2]Liji C. B.

[1]Associate Professor, Post Graduate and Research Department of Zoology, Maharaja's College Ernakulam, Kerala, India.

[2]Research Scholar, Post Graduate and Research Department of Zoology, Maharaja's College Ernakulam, Kerala, India.

Article Received on
15 Sept. 2020,

Revised on 05 Oct. 2020,
Accepted on 25 Oct. 2020

DOI: 10.20959/wjpr202014-19009

***Corresponding Author**
Dr. Rema L. P.
Associate Professor, Post Graduate and Research Department of Zoology, Maharaja's College Ernakulam, Kerala, India.

ABSTRACT

Acetaminophen is one of the most emerging pollutant worldwide. It is a non- prescription analgesic and antipyretic drug. It was a safe drug used in prescribed dose, but overdose may lead to renal failure. It was detected in our water bodies and also has harmful effects on aquatic organisms and experimental animals. In the present study fishes were exposed to sub lethal concentration of $1/5^{th}$ and $1/10^{th}$ of acetaminophen for 96 hours. A control was run along with it. Effect of acetaminophen on kidney tissues increased with overdose and long-term exposure. Fishes exposed to $1/5^{th}$ of acetaminophen showed more tissue damage in kidney, compared to $1/10^{th}$ of sub lethal concentration. Drug induced renal injury has become a major challenge. The result indicates that the acetaminophen at overdose cause nephrotoxicity in experimental fish. The major pathological changes observed in the regions of kidney are presence of pyknotic nuclei, necrosis, congestion, glomerular edema with decrease in bowman's space etc.

KEYWORDS: pharmaceutical pollution, acetaminophen, nephrotoxicity.

INTRODUCTION

In a global context, pharmaceutical pollution is an emerging type of water pollution. The discharge of pharmaceuticals in freshwater bodies is increasing day by day. These compounds enter into aquatic compartment by many ways including excretion after therapeutic use, disposal of domestic surplus drugs directly, and inadequate treatment of

effluents mainly from manufacturing industry and domestic sewage.[1] In addition to the dispersion of liquid manure and its ultimate leaching into water streams, specific compounds used for veterinary treatment or growth promoter in aquaculture activities and live stocks are also a major source of such chemicals.[2] The accumulation of pharmaceuticals in the environment can have adverse ecological impacts and affect human and animal health, especially in aquatic organisms.

Fishes are the important tool for evaluating the degree of pollution. Gill, liver and kidney of fishes are considered as important biomarker for determining the health of fishes. The pollutant induced histological changes in the various organs of fish throw light to its adverse impact on a given aquatic ecosystem. In eco toxicological studies (behavioral, morphological, histological assays) fishes are used as an ideal indicator for various stressors and other harmful compounds, due to their direct interaction with aquatic habitat, ecological importance and tolerance capacity.[3-5]

Kidneys are important organ which function as endocrine, hematopoietic and excretory. In addition to this, kidney perform other functions such as osmoregulation, balancing of water and electrolyte, maintaining the balance of internal environment, excretion of waste products such as ammonia, urea and creatine.[6,7]

Histological examination is a useful method for evaluating the toxic effect of pollution. It is an indicator of exposure to pollutants.[8] Histopathological examinations have long been recognized to be reliable biomarkers in fish due to stress. In this context, a histological examination carried out on the fish to determine the health status of fish as well as the aquatic ecosystem.[9]

MATERIALS AND METHODS

The male *Oreochromis niloticus* of average length 15 ± 1 cm and weight about 100 g were collected from fish farm at Maliankara, Ernakulum district in Kerala, India. The fishes were brought to the laboratory and were acclimatized for a week. After the period of acclimatization, fishes were divided into three groups of 10 each. Group I served as control group. Group II and III fishes were exposed to $1/10^{th}$ and $1/5^{th}$ of 96 hr. LC_{50} of Acetaminophen respectively. For histopathological examination, the kidney was carefully dissected out from the fish. It was washed and wiped thoroughly with blotting paper to remove blood and other body fluids. Then the sample was fixed in Bouins fluid for 24 hours

and further processed to form the wax block. Block with the tissue is cut into thin section of 5micrometer thickness using a microtome. The sections were stained using Haematoxylin and Eosin.

The sections are observed under phase contrast microscope with attached photomicrography machinery (NIKON ECLIPSE 80 i) under 400 X magnification.

RESULT

Acetaminophen-induced histopathological changes in kidney of Nile tilapia observed, are as follows. In control group, kidney of fishes showed normal arrangement of cell. Kidneys have normal tubules. Both proximal and distal convoluted tubules have normal architecture. Each tubule begins from cup like Bowman's capsule. Normal arrangement of glomerulus with normal Bowman's capsule (Fig 1. A)

Fishes treated with $1/10^{th}$ of sub-lethal concentration of Acetaminophen showed almost normal architecture of tubules. But mild congestion in the region of kidney parenchyma was visible. Mild expansion of Bowman's capsule in the glomerulus is also noticed (Fig 1. B)

In fishes treated with $1/5^{th}$ of sub-lethal concentration of Acetaminophen, kidney showed advanced degenerative changes with cellular details. Glomerular oedema with decrease in bowman's space was observed. Glomerular edema is a sign of kidney failure. Kidney tubules showed degenerative changes. Necrosis and presence of debris in the tubular lumen. In the kidney parenchyma blood vessels are filled with blood i.e., blood vessels are highly engorged. A major pathological change is the nuclear pyknosis. It is the condition in which disappearance of nucleus. Some cells are in the stage of cell death (Fig 1.C)

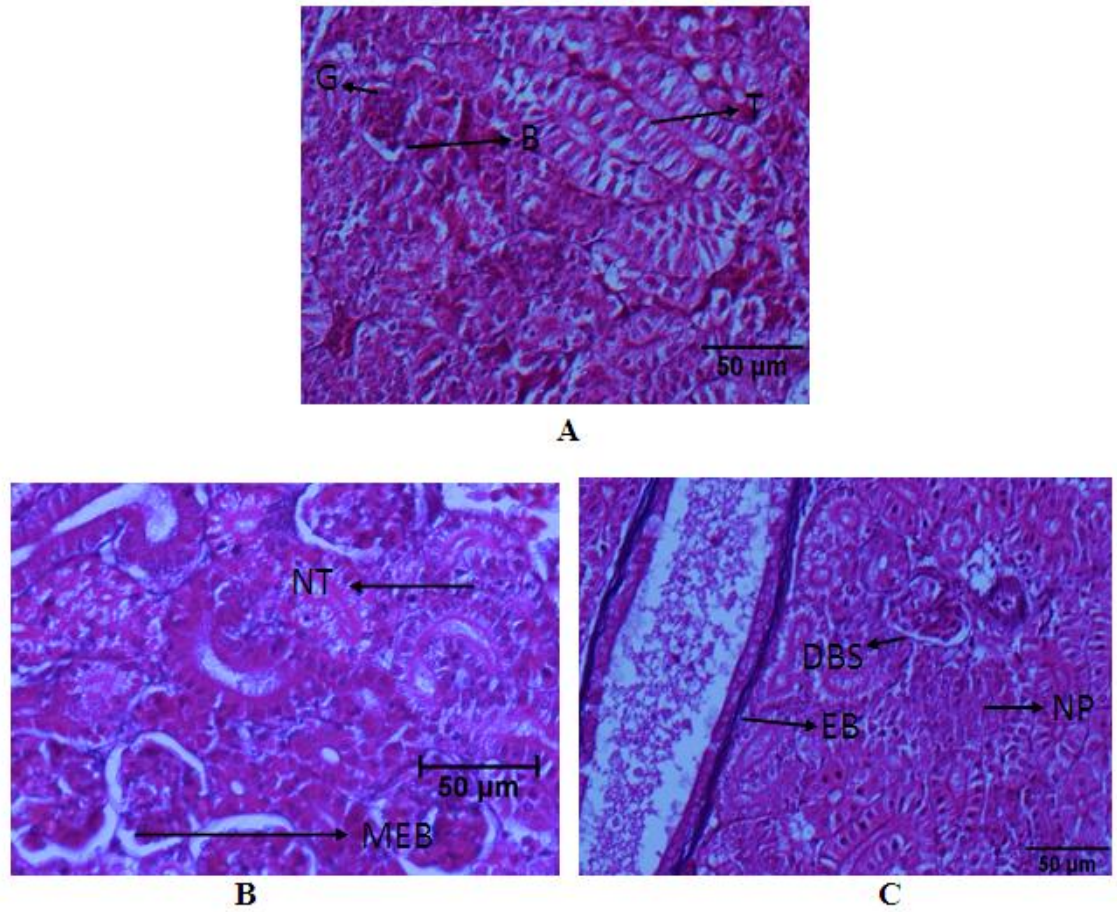

Fig. 1: Histopathological changes in kidney of Nile tilapia treated with Acetaminophen. A. Section of kidney from the control fish: shows normal arrangement of Bowman's capsule (B), normal glomerulus (G) and normal kidney tubules (T). B. Section of kidney from fish treated with 1/10th of 96 hr. $LC_{50:}$ shows mild expansion of Bowman's capsule (MEB) and normal kidney tubules (NT). C. Section of kidney from fish treated with 1/5th of 96 hr. $LC_{50:}$ shows decreased bowman's space (DBS), expanded blood vessel (EB) and nuclear pyknosis (NP).

DISCUSSION

Histological studies in fishes are extremely relevant, since they have direct contact with the polluted habitat. Thus, fishes are considered as an important model for assessing aquatic contamination. They play a major role in food web, bio-accumulating and biomagnifying harmful substances and also responding to low concentrations of mutagens.[10] Thophon et al[11] observed that the teleostean kidney is one of the first organs to be affected by contaminants present in the water.

Many authors have previously documented Acetaminophen induced nephrotoxicity.[12-14] When compared to hepatotoxicity, nephrotoxicity is less common in acetaminophen overdose. But even in the absence of liver injury, renal damage and acute renal failure can occur and lead to death in humans and experimental animals.[15,16] Toxicity of Acetaminophen is associated mainly with liver damage. The nephrotoxicity or kidney dysfunction may occur as a secondary effect to hepatotoxicity. Vermeulen *et al*[17] reported that in rare cases acute renal tubular necrosis due to acetaminophen also occurred. Recently more studies have been incorporated the kidney in biomarker research. Histological studies have also used to identify in the renal tissue after exposure to toxicants.[18,19]

Histologically, teleost kidney consists of two parts - renal parenchyma and interstitial tissues. In renal parenchyma functional unit is nephron and interstitial tissue consist of hematopoietic tissues.[20] In fresh water teleost nephron is divided into many parts such as renal corpuscle (malpighian body) consisting of glomerulus and Bowman's capsule; renal tubule divided into a neck segment, and proximal convoluted tubule consisting of two parts P 1 and P2; an intermediate segment and a distal convoluted tube.[20] Kidney of control fishes showed all the parts intact and with normal cells. Arrangement of glomerulus with normal Bowman's capsule and normal tubules are noted. Both proximal and distal convoluted tubules have normal architecture.

Fishes treated with $1/10^{th}$ 96 hr. LC_{50} of Acetaminophen showed almost normal architecture of tubules. But congestion in the region of kidney parenchyma and mild expansion of Bowman's capsule in the glomerulus was observed. Various drugs and chemicals cause nephrotoxicity. It occurs as a result of disturbances in renal function.[21] Rat treated with cisplatin a chemotherapeutic drug at dose >5 mg/Kg body weight showed signs of nephrotoxicity.[22,23]

Fishes treated with $1/5^{th}$ 96 hr. LC_{50} of Acetaminophen showed advanced degenerative changes with respect to cellular details. According to Jones and Vale[24] even in situations where hepatotoxicity is not observed with toxic dose intakes, nephrotoxicity is reported. In the present study kidney showed glomerular edema with decrease in Bowman's space. Glomerular edema is a sign of kidney failure. Kidney tubules showed degenerative changes. Additional glomerular damage could be observed at the ultra-structural level.[25] Paracetamol and gentamicin - the two drugs associated with renal tubular damage in man,[26,27] have been

proved to cause morphological and/or functional alterations in the zebra fish pronephros also.[28 - 32]

In the present study, kidney showed necrosis and presence of debris in tubular lumen. Das and Mukherjee[33] reported dilation of renal tubules and necrotic changes characterized by karyorrhexix and karyolysis in hexachloro-cyclohexane exposed *Labeo rohita.* In the kidney parenchyma blood vessels are highly engorged. A major pathological change noticed is the nuclear pyknosis. Degeneration in the epithelial cells of renal tubules, pyknotic nuclei in the hematopoietic tissue, dilation of glomerular capillaries, degeneration of glomerulus, intra cytoplasmic vacuoles in the epithelial cells with hypertrophied cells and narrowing of the tubular lumen were observed in the kidney of fish exposed to deltamethrin.[34] Nasser *et al*[35] observed that nephrotoxicity due to over dose of acetaminophen causes changes in the cytoarchitecture of the glomeruli, proximal, distal convoluted tubules, as well as interstitium. Tilak *et al*[36] confirmed severe necrosis, cloudy swelling in the renal tubules, cellular hypertrophy, granular cytoplasm and vacuolization in kidney tissues of *Ctenopharyngodon idella* exposed to fenvalerate.

Many pathological changes associated with kidney are reported by various scientists. It includes dilation or inflammation of blood capillaries in glomerulus and thickening of their walls, damage and fibrosis of glomerulus capillaries sclerosis, abnormal increase of epithelial cells in Bowman's capsule, cloudy swelling of epithelial cells present in renal tubules, degeneration of hyaline droplet in epithelial cells of renal tubule, epithelial cell necrosis and glycogen infiltration of tubular epithelium.[20]

CONCLUSIONS

Many pharmaceutical drugs are reported in considerable amount in water bodies. The accumulation of pharmaceuticals in water bodies have adverse ecological impact. It may harmfully affect human and animal health. Acetaminophen have toxic effect on kidney of Nile Tilapia when exposed to sub lethal concentrations. Kidneys of fishes are very sensitive to Acetaminophen in a dose dependent manner. Acetaminophen is a widely used drug and detected in the environment. Its continuous exposure lead to harmful effect on fishes and also affect the ecosystem.

REFERENCES

1. Lapworth DJ, Baran N, Stuart, ME, Ward, RS. Emerging organic contaminants in groundwater: a review of sources, fate and occurrence. Environmental pollution, 2012; 163: 287-303.
2. Sui Q, Cao X., Lu S, Zhao W, Qiu Z, Yu G. Occurrence, sources and fate of pharmaceuticals and personal care products in the groundwater: a review. Emerging Contaminants, 2015; 1(1): 14-24.
3. Kane AS, Salierno JD, Brewer SK. Fish models in behavioral toxicology: automated techniques, updates and perspectives. Methods in aquatic toxicology, 2005; 2: 559-590.
4. Kaur R, Dua A. Scales of freshwater fish *Labeo rohita* as bio indicators of water pollution in Tung Dhab Drain, Amritsar, Punjab, India. Journal of Toxicology and Environmental Health, Part A, 2015 b; 78(6): 388-396.
5. Kaur R, Dua A. Colour changes in *Labeo rohita* (Ham.) due to pigment translocation in melanophores, on exposure to municipal wastewater of Tung Dhab drain, Amritsar, India. Environmental Toxicology and Pharmacology, 2015 c; 39(2): 747-757.
6. Jiraungkoorskul, W, Upatham, ES, Kruatrachue M, Sahaphong S, Vichasri-Grams S, Pokethitiyook P. Biochemical and histopathological effects of glyphosate herbicide on Nile tilapia (*Oreochromis niloticus*). Environmental Toxicology: An International Journal, 2003; 18(4): 260-267.
7. Ortiz JB, de Canales, MLG, Sarasquete C. Histopathological changes induced by lindane (?-HCH) in various organs of fishes. Scientia Marina, 2003; 67(1): 53-61.
8. Bernet D, Schmidt H, Meier W, Burkhardt-Holm P, Wahli T. Histopathology in fish: proposal for a protocol to assess aquatic pollution. Journal of fish diseases, 1999; 22(1): 25-34.
9. Van der Oost R, Beyer J, Vermeulen NP. Fish bioaccumulation and biomarkers in environmental risk assessment: a review. Environmental toxicology and pharmacology, 2003; 13(2): 57-149.
10. Al-Sabti K, Metcalfe CD. Fish micronuclei for assessing genotoxicity in water. Mutation Research/Genetic Toxicology, 1995; 343(2-3): 121-135.
11. Thophon, S, Kruatrachue M, Upatham ES, Pokethitiyook P, Sahaphong S, Jaritkhuan S. Histopathological alterations of white seabass, Lates calcarifer, in acute and subchronic cadmium exposure. Environmental Pollution, 2003; 121(3): 307-320.
12. Mitchell JR, McMurtry RJ, Statham CN, Nelson SD. Molecular basis for several drug-induced nephropathies. The American journal of medicine, 1977; 62(4): 518-526.

13. McMurtry RJ, Snodgrass, WR, Mitchell JR. Renal necrosis, glutathione depletion, and covalent binding after acetaminophen. Toxicology and applied pharmacology, 1978; 46(1): 87-100.
14. Newton JF, Yoshimoto M, Bernstein J, Rush GF, Hook JB. Acetaminophen nephrotoxicity in the rat: I. Strain differences in nephrotoxicity and metabolism. Toxicology and applied pharmacology, 1983; 69(2): 291-306.
15. Cekmen, Mustafa, Ilbey YO, Ozbek E, Simsek A, Somay A, Ersoz C. Curcumin prevents oxidative renal damage induced by acetaminophen in rats. Food and Chemical Toxicology, 2009; 47(7): 1480-1484.
16. Palani S, Raja S, Kumar RP, Jayakumar S, Kumar BS. Therapeutic efficacy of Pimpinella tirupatiensis (Apiaceae) on acetaminophen induced nephrotoxicity and oxidative stress in male albino rats. Int J PharmTech Res, 2009; 1(3): 925-34.
17. Vermeulen NPE, Bessems JGM, Van de Straat R. Molecular aspects of paracetamol-induced hepatotoxicity and its mechanism-based prevention. Drug metabolism reviews, 1992; 24(3): 367-407.
18. Schmidt-Posthaus H, Bernet D, Wahli T, Burkhardt-Holm P. Morphological organ alterations and infectious diseases in brown trout *Salmo trutta* and *Rainbow trout Oncorhynchus mykiss* exposed to polluted river water. Diseases of Aquatic Organisms, 2001; 44(3): 161-170.
19. Bernet D, Schmidt-Posthaus H, Wahli T, Burkhardt-Holm P. Evaluation of two monitoring approaches to assess effects of waste water disposal on histological alterations in fish. Hydrobiologia, 2004; 524(1): 53-66.
20. Takashima F, Hibiya T. An atlas of Fish histology: Normal and Pathological features: 1995.
21. Watkins PB, Kaplowitz N, Slattery JT, Colonese CR., Colucci SV, Stewart PW, Harris S C. Aminotransferase elevations in healthy adults receiving 4 grams of acetaminophen daily: a randomized controlled trial. Jama, 2006; 296(1): 87-93.
22. DeWoskin RS, Riviere JE. Cisplatin-induced loss of kidney copper and nephrotoxicity is ameliorated by single dose diethyldithiocarbamate, but not mesna. Toxicology and applied pharmacology, 1992; 112(2): 182-189.
23. Babu E, Gopalakrishnan VK, Sriganth, INP, Gopalakrishnan R, Sakthisekaran, D. Cisplatin induced nephrotoxicity and the modulating effect of glutathione ester. Molecular and cellular biochemistry, 1995; 144(1): 7-11.

24. Jones AF, Vale JA. Paracetamol poisoning and the kidney. Journal of clinical pharmacy and therapeutics, 1993; 18(1): 5-8.
25. Triebskorn R, Casper H, Heyd A, Eikemper R., Köhler HR, Schwaiger J. Toxic effects of the non-steroidal anti-inflammatory drug diclofenac: Part II. Cytological effects in liver, kidney, gills and intestine of rainbow trout (*Oncorhynchus mykiss*). Aquatic toxicology, 2004; 68(2): 151-166.
26. Mazer M, Perrone J. Acetaminophen-induced nephrotoxicity: pathophysiology, clinical manifestations, and management. Journal of Medical Toxicology, 2008; 4(1): 2-6.
27. Lopez-Novoa JM, Quiros Y, Vicente L, Morales AI, Lopez-Hernandez FJ. New insights into the mechanism of aminoglycoside nephrotoxicity: an integrative point of view. Kidney international, 2011; 79(1): 33-45.
28. Hentschel DM, Park KM, Cilenti, L, Zervos AS, Drummond I, Bonventre JV. Acute renal failure in zebra fish: a novel system to study a complex disease. American Journal of Physiology-Renal Physiology, 2005; 288(5): F923-F929.
29. Peng HC, Wang YH, Wen CC, Wang WH, Cheng CC, Chen YH. Nephrotoxicity assessments of acetaminophen during zebra fish embryogenesis. Comparative Biochemistry and Physiology Part C: Toxicology & Pharmacology, 2010; 151(4): 480-486.
30. Rider SA, Mullins LJ, Verdon RF, MacRae CA, Mullins JJ. Renin expression in developing zebra fish is associated with angiogenesis and requires the Notch pathway and endothelium. American Journal of Physiology-Renal Physiology, 2015; 309(6): F531-F539.
31. Cosentino CC, Skrypnyk NI, Brilli LL, Chiba T, Novitskaya T, Woods … Davidson AJ. Histone deacetylase inhibitor enhances recovery after AKI. Journal of the American Society of Nephrology, 2013; 24(6): 943-953.
32. Westhoff JH, Giselbrecht S, Schmidts M, Schindler S, Beales PL, Tönshoff B, ... Gehrig J. Development of an automated imaging pipeline for the analysis of the zebra fish larval kidney. PLoS One, 2013; 8(12): e82137.
33. Das BK, Mukherjee SC. A histopathological study of carp (Labeo rohita) exposed to hexachlorocyclohexane. Veterinarski Arhiv, 2000; 70(4): 169-180.
34. Cengiz, EI. Gill and kidney histopathology in the freshwater fish Cyprinus carpio after acute exposure to deltamethrin. Environmental Toxicology and Pharmacology, 2006; 22(2): 200-204.

35. Nassar I, Pasupati T, Judson JP, Segarra I. Histopathological study of the hepatic and renal toxicity associated with the co-administration of imatinib and acetaminophen in a preclinical mouse model. Malays J Pathol, 2010; 32(1): 1-11.
36. Tilak KS, Veeraiah, K, Yacobu K. Studies on histopathological changes in the gills liver and kidney of *Ctenopharyngodon indellus* (Valen.) exposed to technical fenvalerate and EC 20%. Pollution Research, 2001; 20(3): 387-393.