International Journal of Engineering, Science and Mathematics
Vol. 15 Issue 8,Aug 2026,
ISSN: 2320-0294 Impact Factor: 8.215
Journal Homepage: http://www.ijesm.co.in, Email: ijesmj@gmail.com
Double-Blind Peer Reviewed Refereed Open Access International Journal - Included in the International Serial Directories Indexed & Listed at: Ulrich's Periodicals Directory ©, U.S.A., Open J-Gage as well as in Cabell's Directories of Publishing Opportunities, U.S.A

Ameliorative Effect of Selenium on Endosulfan Induced Nephrotoxicity in Swiss Albino Mice: Histological and Biochemical Analysis

Dr. Sunita Kumari*[1], Mrs. Bhawana Singh[2]

*[1]Assistant Professor, Department of Zoology, Ram Krishna Dwarika College, Patliputra University; s.k.patna26@gmail.com

[2]Assistant Professor, Department of Zoology, Ram Krishna Dwarika College, Patliputra University; bhawana15bhu@gmail.com

ABSTRACT

The widespread application of agrochemical specially organochemical in agriculture has contributed to serious health concerns for humans and has also negatively impacted the environment. Endosulfan, a widely known organochemical pesticide, possess serious health risks as it induces long term harmful detrimental effect on reproductive, immune and endocrine system. Although it has been banned globally, its use persists in some regions of the world.

This study aimed to assess Endosulfan induced nephrotoxicity by conducting histological examinations and biochemical analysis of serum creatinine level. Swiss Albino mice were selected for study. It also evaluates ameliorative effect of selenium as an antioxidant. Swiss albino mice were orally administered Endosulfan at its maximum tolerated dose for four weeks. This was followed by selenium dissolved in water at its maximum tolerated dose for an additional four weeks. Histological as well as biochemical analysis of kidney tissues were done at certain intervals using light microscopy to examine structural lesions and biochemical changes. The finding revealed several prominent histological lesions in renal cortex and tubules, after Endosulfan exposure. Treatment with Selenium demonstrated protective effect, reducing necrosis, clumping of glomerulus cells and the serum creatinine level was also observed normal. These findings demonstrate the potential of the antioxidant selenium as a nephroprotective agent against the toxicity induced by endosulfan.

Key words: Endosulfan, Swiss albino mice, Selenium, Nephrotoxicity, Antioxidant

INTRODUCTION

Approximately 1/3rd of agricultural production is lost annually due to pest infestation. The severity of these losses continues to increase each year with emergence of new pests and plant diseases. To mitigate these challenges pesticides are widely used in modern agriculture. However, while these chemicals are effective in controlling pests, they also pose significant risk to human health and environment. Prolong exposure to such substances can lead to toxic effect in human and their persistence in soil and water ecosystems further contributes to environmental degradation.

Endosulfan, an organochlorine pesticide, tends to persist in regions where it is applied due to its chemical stability and low biodegradability. As a result, it accumulates in the environment. Despite restrictions and ban in many countries, its extensive consistent use has contributed to its widespread presence in soil, water and biological system. Endosulfan has been linked to multiple adverse effects inducing endocrine disruption, immune suppression, reproductive toxicity and genotoxicity (Pandey et al.). Studies has also reported its potential to induce kidney diseases of unknown etiology (Ghosh et al.) with nephrotoxicity being one of the significant manifestation impact of endosulfan on kidney of Swiss albino mice were highlighted by Singh and Pandey (Singh and Pandey).

Selenium is one of the micronutrient that plays an important role in the health of animal (Mehdi and Dufrasne). It protects the cell against excess ROS & regulation of immune system (El-Boshy et al.). In Selenium deficit, results oxidative stress and apoptosis in chicken were highlighted by Yao et al. (Yao et al.). Studies also shown that selenium has protective effect on ischemia reperfusion injury in rat testis (Kara et al.). Endosulfan toxicity and its reducing effect by selenium has been reported (Shafiq-ur-Rehman). As, selenium is already established strong antioxidant, it may also act against nephrotoxicity. The results of the study add to our understanding of the effects of selenium on nephrotoxicity. This study highlights selenium's potential to prevent pesticide-induced nephrotoxicity, highlighting its importance in creating substitute tactics to counteract kidney damage brought on by environmental pollutants.

MATERIALS AND METHOD

Experimental animal

The study was conducted using healthy male mice (*Mus musculus*), Swiss albino mice, each weighing approximately 28 ± 2 grams. The animals were procured from Molecular Cell Biology Laboratory, Department of Zoology, Patna University, Patna. Standard laboratory

conditions were maintained throughout the experiment. The mice were housed in well aerated cages with rice husk bedding. Water was provided ad libitum through nozzles and bottles. A 12 hours light and dark cycle was maintained with temperature 22 ± 2°C.

***Chemicals*:** Endosulfan (35% emulsifiable concentrate, EC) was used as pesticide. It was procured from Excel India Private Limited, Mumbai.

***Antioxidant*:** Sodium selenite was purchased from market for experiment. It was dissolved in drinking water.

Methodology

Phase 1*: *Determination of Maximum Tolerated Dose (MTD) of Endosulfan

To establish MTD of endosulfan mice were divided into six groups and kept in separate cages. Each group received separate dose of endosulfan concentration with drinking water (0.6 ppm, 0.48 ppm, 0.96 ppm, 1.92 ppm, 3.84 ppm) following a doubling manner.

Observations

1.Within the 1st week four mice exposed to a dose of 3.84 ppm, dies.

2. Mice exposed to 1.92 ppm survived for up to two weeks.

3. In contrast, mice treated with lower dose up to 0.96 ppm survived for one month without any observable adverse effects.

Based on these observations 0.96 ppm was established maximum tolerated dose.

Phase 2: Determination of maximum tolerated dose of selenium

To establish MTD of selenium sodium selenite was dissolved in drinking water making concentration of 0.5 ppm, 1 ppm and 2 ppm and was orally administered for one, two and four weeks.

Observations

1. Mice were found dead when exposed to 2 ppm.

2. Mice survived when treated up to 1ppm without any adverse effect for 1 month.

1 ppm concentration found to be effective without any lethal effect, so selected for maximum tolerated dose.

Experimental design: 30 mice were housed in three cages separately in a group of 10 mice each.

1. ***Control group:*** Received normal feed and water.
2. ***Endosulfan treated:*** Administered 0.96 ppm daily dose orally for 4 weeks.
3. ***Selenium treated group:*** Following endosulfan administration, the treated mice received 1 ppm sodium selenite daily dose orally with drinking water for 7, 14 and 28 days to assess ameliorative effect of selenium.

Histopathological Examination

Mice were anesthetized with chloroform and subsequently sacrificed with minimal stress at interval for 7, 14 and 28 days post treatment. Kidney tissues were excised, rinsed with normal saline and cut into small pieces.

The tissues were then fixed in 10% neutral buffered formalin for at least 24 hours. At fixation, sample were rinsed for 24 hours running water, dehydrated gradually with graded alcohol, cleared in xylene and finally paraffin embedding was done. It was cut in 3.4 μm section using microtome and stained with hematoxylin and eosin for histopathological examination.

Biochemical Analysis

Creatinine levels were estimated in Swiss albino mice to assess kidney function following treatment. A designated time interval (7,14 and 28 days post treatment) mice were anesthetized and sacrificed. Blood samples were collected and allowed to clot, followed by centrifugation to obtain serum.

Serum creatinine levels were measured using a standard biochemical assay kit based on the Jaffe's reaction method. Creatinine concentration was calculated using standard calibration curve. Results were expressed in mg/dl.

Histological Assessment

The stained kidney section after examining under light microscope, structural changes were assessed. The parameter taken for examination were renal cortex and tubules, cellularity, necrosis and clumping of glomerulus.

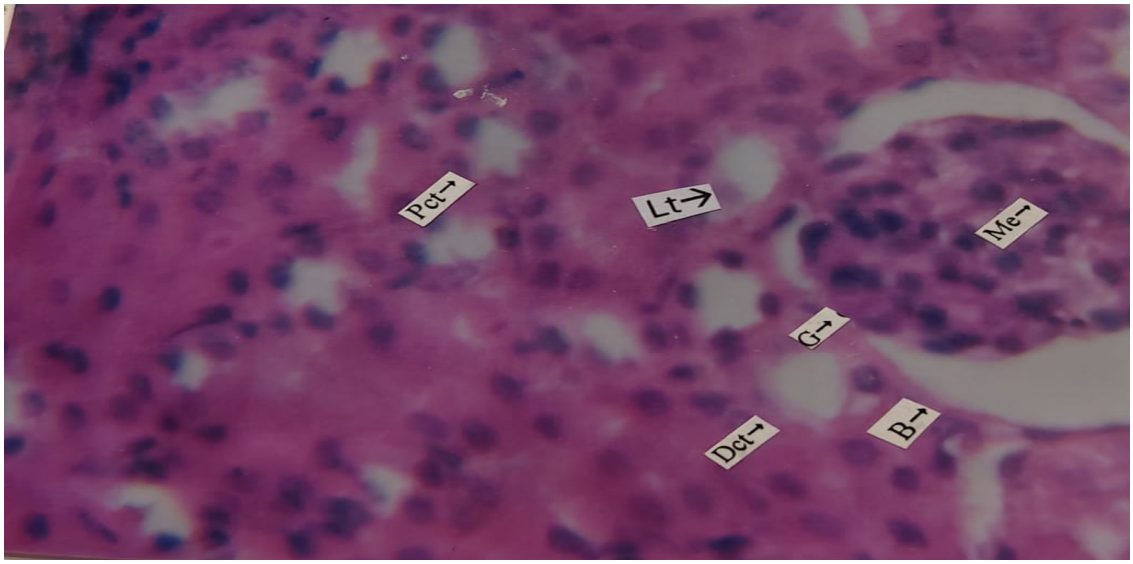
Ethical Statement: All experimental procedures were conducted following the guidelines of committee and approved by the Institutional Animal Committee of Patna University.

RESULTS

Histological examinations of renal corpuscles and tubules

Histological examination of renal corpuscles was observed using light microscopy. The structural changes observed were of both endosulfan treated, selenium treated as well as well compared with normal. In the control group (Figure 1), kidney tissue observed normal, with no significant structural changes with no lesions. In the endosulfan treated group (Figure 2), histopathological lesions were apparent increasing with duration of exposure.

- After 7 days of endosulfan administration at 0.96 ppm, minor histopathological lesion were observed.
- After 14 days, the lesions appeared more pronounced, glomeruli also appeared clumped.



- After 28 days, the changes were more drastic, and extensive. Glomeruli appeared wire loop like due to acellularity, necrosis in bowman's capsule, vacuolation, cellular infiltrates were present in proximal convoluted tubule and distal convoluted tubule.

Figure 1: Normal kidney tissue showing normal glomerulus (G), Bowman's Capsule (B), Proximal convoluted tubule (Pct), Distal convoluted tubule (Dct), Mesangial cells (Me) and Lumen tubule (Lt) at 200X.

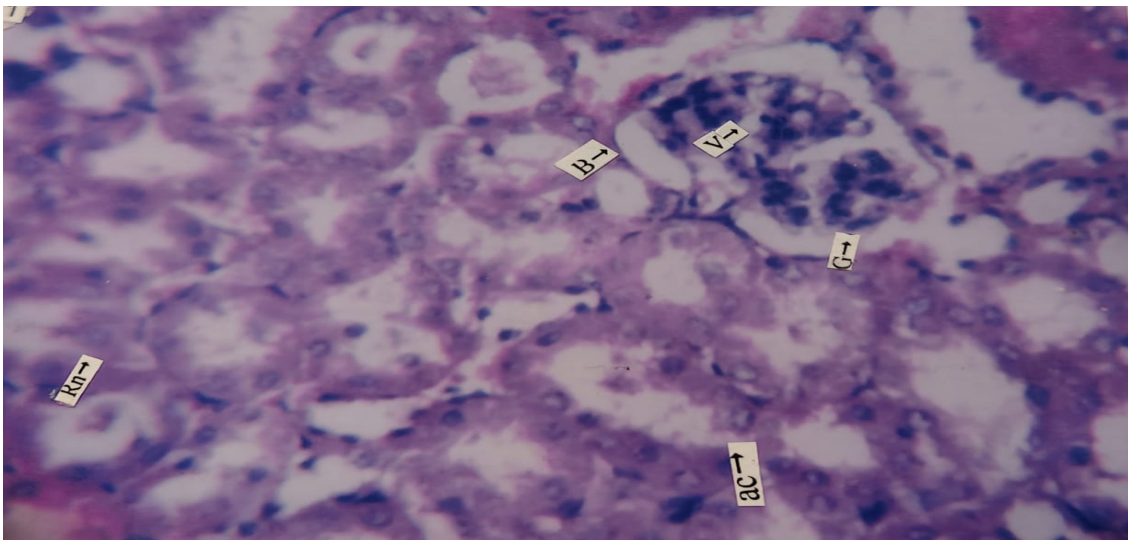


Figure 2: Kidney tissue from endosulfan treated mice showing vacuolation (V) and necrosis abnormalities in glomerulus (G), and pycnotic nuclei in Renal tubule (Rn), acellularity in Bowman's Capsule (B) and abnormal cell of Proximal convoluted tubule & Distal convoluted tubule (ac) at 200X

Biochemical examination and statistical analysis

Serum creatinine level was observed after post treatment and compared with normal level, represented in Table 1, Figure 3. After 7 days of endosulfan treatment at 0.96 ppm creatinine level increased by 8.5% which were significant (P<0.0001). After 14 days of endosulfan treatment at 0.96 ppm, creatinine level increased by 18.3% which were highly significant (P<0.0001). After 28 days of endosulfan treatment, 22.01% increase was observed indicating a highly significant change (P<0.001).

TABLE 1: Selenium (1 ppm) induced changes in creatinine level in Endosulfan treated mice (value expressed in mg/dl). Mice were treated with 0.96 ppm endosulfan for 7 days (EN7), 14 days (EN14) and 28 days (EN28).

	% CHANGE	*MEAN*	*P*
Control		0.668	
EN7	8.5%	0.72	**
EN7 + Selenium	2.1%	0.710	NS
EN14	+18.5%	0.79	**
EN14 + Selenium	9.87%	0.712	*
EN28	22.01%	0.815	**
EN28 + Selenium	12.64%	0.71	**

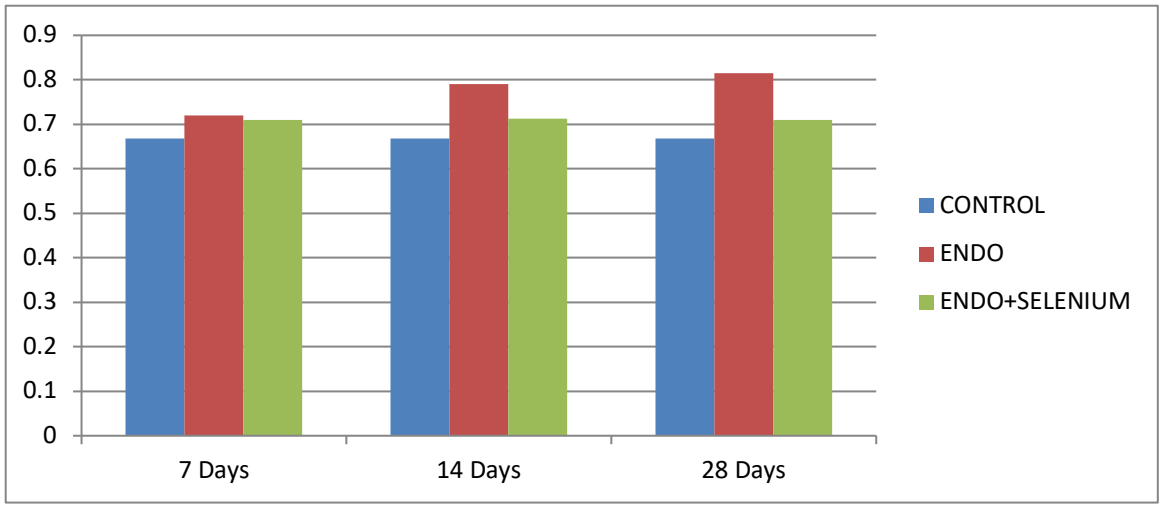


Figure 3: Selenium (1 ppm) induced changes in creatinine level in Endosulfan treated mice for 7 days, 14 days and 28 days.

Ameliorative effects of selenium:

In selenium treated group (Figure 4), the administration of 1 ppm selenium daily for 7, 14 and 28 days demonstrated noticeable change in serum creatinine level. It was observed noticeable decrease in serum creatinine level with ameliorative effect.

- After 7 days selenium treated group: The administration of 1 ppm selenium for 7 days demonstrates ameliorative effect on endosulfan treated nephrotoxicity. Creatinine level decreases by 2.1 %, (P< 0.01) which is less significant.
- After 14 days: Percentage decreases by 9.87%, (P<0.01) which is significant.
- After 28 days: Percentage decreases by 12.64%, (P<0.001) which is highly significant.

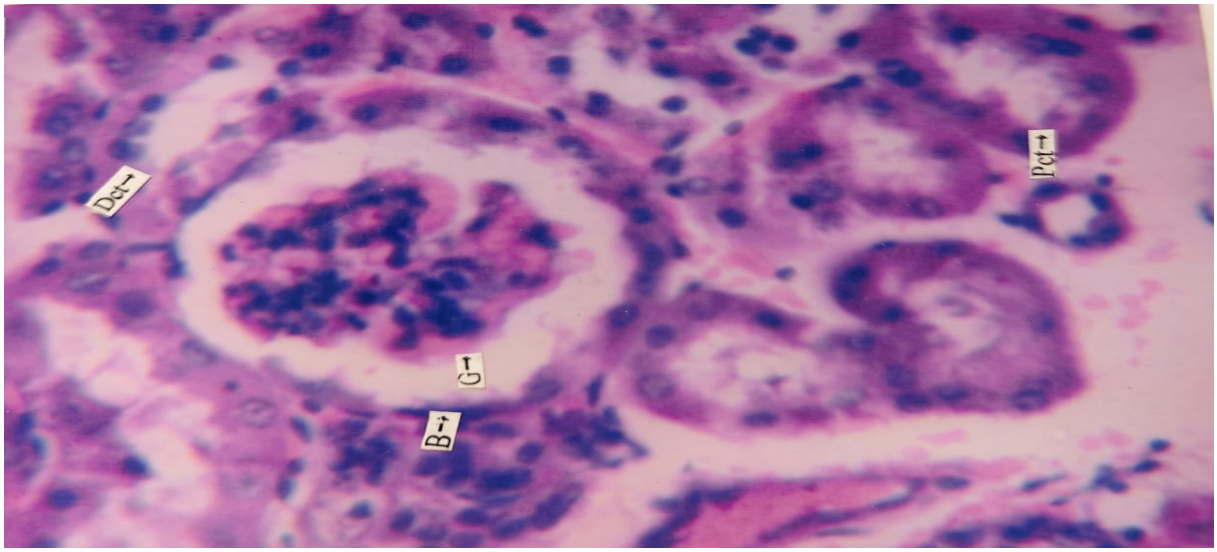


Figure 4: Kidney tissue structure showing reduced necrosis and improved cellular integrity of glomerulus (G), Bowman's Capsule (B), Proximal convoluted tubule (Pct) and Distal convoluted tubule (Dct) at 200X.

DISCUSSION

In the present investigation showed that sub lethal treatment of endosulfan induces pronounced lesions in kidney of mice. These lesions were visible in renal cortex as well as tubules of Swiss albino mice. Along with the present and previous study, the research highlights the nephrotoxic effect of endosulfan which induces severe histological lesions such as necrosis of bowman's capsule, glomerulus and vacuolation (Khan). Prolonged exposure to endosulfan revealed the histopathological lesions more pronounced such as wire loop like glomerulus, clumped PCT, DCT, acellularity in renal cortex and tubules.

Mechanism of endosulfan induced nephrotoxicity

Several studies reveal that endosulfan exposure are associated with heavy oxidative stress in rats' heart (Enas). It results to generate ROS, causing oxidative stress leading to apoptosis. These findings were reported and it was revealed that there were ultrastructural damages in mice kidney due to endosulfan exposure (Caglar et al.). Further, Singh and Pandey highlighted the effect of endosulfan exposure on plasma gonadotropin, testosterone of

androgen biosynthesis which likely compounds its nephrotoxic effect (Singh and Pandey).

Protective effect of selenium

The administration of selenium following endosulfan treatment significantly and noticeable relived the histopathological lesions, as evidence by normal integrity of renal cortex as well as tubules (figure 4).The structural restoration of bowman's capsule, glomerulus, PCT & DCT shows the efficacy of selenium against it.

Comparative study

Selenium was reported to show efficacy against oxidative stress in various studies (Klotz et al.). It protects cell membrane from damage caused by harmful free radicals and plays a role in liver to detoxify harmful compounds, so that they can be removed from body. Selenium dependent cellular glutathione peroxidase protects mice against a pro-oxidant induced oxidation of NADH, NADPH & lipids (Xuliang et al.). The endosulfan toxicity and its reduction by selenium has been studied extensively (Shafiq-ur-Rehman). The protective effect of Vit. E and selenium on renal morphology have been reported by Gonca et al. (Gonca et al.) in rat fed with high cholesterol diets .

Implication and recommendation

The ameliorative effect of selenium in this study underscores its potential as an effective antioxidant in mitigating the pesticide induced nephrotoxicity. Given its availability and safety profile, selenium may serve viable therapeutic agent for individual exposed to endosulfan.

CONCLUSION

The study confirms that endosulfan causes histopathological lesions in kidney and high creatinine level in Swiss albino mice. The lesions caused by endosulfan are glomerulus and bowman's capsule, acellularity, clumping of cells, necrosis as well as appearance of eosinophil cells. The administration of selenium as an antidote showed efficacy against pathological lesion and restores the renal cortex and tubules to normal and reduces oxidative stress.

Further research is recommended to explore the molecular pathway unsolved in selenium protective effects to validate these findings in other models. Additionally, long-term research could explore the potential of selenium as protective supplement for population, who are at risk of endosulfan exposure.

REFERENCES

1. Caglar, Y., et al. "Ultrastructural evaluation of the effect of endosulfan on mice kidney." *Histology and histopathology* 18.3 (2003): 703-708.

2. El-Boshy, Mohamed E., et al. "Protective effects of selenium against cadmium induced hematological disturbances, immunosuppressive, oxidative stress and hepatorenal damage in rats." *Journal of Trace Elements in Medicine and Biology* 29 (2015): 104-110.

3. Enas, A. Abbas. "Preventive role of selenium against toxicological and pathological effects of ochratoxin A in rabbits Enas, A. Abbas*; Amany, M. Salama*; Eman, IM Ismail**; Abdalla, SH***; Sahar, N. Mohamedy*** and Naglaa, S. Soliman."
4. Ghosh, Rishila, et al. "Organochlorine pesticide level in patients with chronic kidney disease of unknown etiology and its association with renal function." *Environmental health and preventive medicine* 22.1 (2017): 49.
5. Gonca, Süheyla, et al. "Protective effects of vitamin E and selenium on the renal morphology in rats fed high-cholesterol diets." *Pathobiology* 68.6 (2001): 258-263.
6. Kara, Önder, et al. "Effects of selenium on ischaemia–reperfusion injury in a rat testis model." *Andrologia* 48.10 (2016): 1267-1273.
7. Khan, Sabiha. "Study of histopathological changes induced by endosulfan in kidney of albino rats." *Journal of Drug Delivery & Therapeutics* 4.3 (2014): 125-128.
8. Klotz, Lars-Oliver, et al. "Role of copper, zinc, selenium and tellurium in the cellular defense against oxidative and nitrosative stress." *The Journal of nutrition* 133.5 (2003): 1448S-1451S.
9. Mehdi, Youcef, and Isabelle Dufrasne. "Selenium in cattle: a review." *Molecules* 21.4 (2016): 545.
10. Pandey, Nina, et al. "Studies on the genotoxicity of endosulfan, an organochlorine insecticide, in mammalian germ cells." *Mutation Research/Genetic Toxicology* 242.1 (1990): 1-7.

11. Shafiq-ur-Rehman. "Evaluation of malonaldialdehyde as an index of chlorpyriphos insecticide exposure in Apis mellifora: Ameliorating role of melatonin and α-tocopherol against oxidative stress." *Toxicological and Environ Chemistry* 91.6 (2009): 1135-1148.
12. Singh, S. K., and R. S. Pandey. "Effect of sub-chronic endosulfan exposures on plasma gonadotrophins, testosterone, testicular testosterone and enzymes of androgen biosynthesis in rat." *Indian Journal of Experimental Biology* 28.10 (1990): 953-956.

13. Xuliang, Zhang, et al. "The protective effect of selenium on T-2-induced nephrotoxicity is related to the inhibition of ROS-mediated apoptosis in mice kidney." *Biological trace element research* 200.1 (2022): 206-216.
14. Yao, Linlin, et al. "Roles of oxidative stress and endoplasmic reticulum stress in selenium deficiency-induced apoptosis in chicken liver." *Biometals* 28.2 (2015): 255-265.

ACKNOWLEDGEMENT

The author sincerely acknowledges the molecular cell biology laboratory, Dept. of Zoology, Patna University for providing resources & laboratory facilities to conduct this research.

The author deeply appreciates the support & encouragement given throughout research to Ram Krishna Dwarika Mahavidyalaya,PPU, Patna, 800026.

Conflict of interest

The author declares no conflict of interest concerning the research & publication of this manuscript.