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Effect of DDT on Haematological and Histopathological Parameters in *Clarias batrachus*

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Abstract

The current work aims to study the effect of DDT on the haematological and histological characteristics of freshwater fish Clarias batrachus. A persistent organochlorine insecticide, DDT, can build up in aquatic environments and negatively impact organisms that are not its intended targets. Four groups of sixteen healthy fish were created: low dose (3.62 mg/L), medium dose (6.80 mg/L), high dose (13.62 mg/L), and control. Initially, the fish tanks comprised of four fishes each. As a consequence of DDT exposure, which lasted for 21 days, two fish were left in each group at the conclusion of the exposure period. Red blood cell (RBC), white blood cell (WBC), and haemoglobin (Hb) counts were among the haematological markers that were examined. The findings showed that fish exposed to DDT had significantly lower RBC counts and haemoglobin concentrations than the control group, suggesting the development of anaemia. The WBC count, on the other hand, significantly increased, indicating that toxic stress had triggered the immunological response. The kidney, liver, and gill tissues' histopathological examination showed dose-dependent structural alterations. The gills displayed hyperplasia, degeneration, lamellar fusion, and epithelial elevation. Hepatic cord disorganisation, congestion, hepatocellular vacuolation, and necrotic changes were observed in the liver. Tubular degeneration, vacuolation, congestion, and disruption of normal tissue architecture were all seen or displayed in the kidney. As the DDT content rose, the lesions' severity gradually worsened. Analysis of Variance (ANOVA) was used for the statistical analysis, and differences were deemed significant at $p < 0.05$. Body weight, haematological parameters (Hb, RBC, and WBC), and histopathological features of Clarias batrachus exposed to different DDT doses showed substantial dose-dependent differences from the control group, according to the analysis. The results of this investigation show that DDT significantly damages Clarias batrachus at the physiological and tissue levels. Therefore, haematological and histopathological markers can be employed as reliable biomarkers to monitor pesticide toxicity in freshwater settings.

Keywords: *haematological, histological, Clarias batrachus, organochlorine, DDT exposure.*

Introduction

On Earth, aquatic environments or ecosystems are among the most biologically prominent and productive. A vast variety of living things are supported by rivers, ponds, lakes, reservoirs, wetlands, and streams, which also offer vital ecological services to people and wildlife. Freshwater ecosystems have an important role in food production, agriculture, fishing, transportation, pleasure, and industrial operations in addition to providing habitat for a wide variety of aquatic creatures. However, increased industrialization, urbanization, and agricultural intensification have resulted in serious contamination of aquatic habitats [1]. Due to their widespread use in agriculture or farm and community health management initiatives, pesticides hold a significant position among many environmental pollutants. Chemicals known as pesticides are used to eradicate or manage dangerous pests, insects, weeds, and organisms that cause illness. Despite the fact that pesticides play a major role in crop yield and disease management, their excessive usage has raised major ecological and health issues globally.

Dichloro-Diphenyl-Trichloroethane (DDT) is one of the most prominent insecticides in history. DDT is a synthetic organochlorine pesticide that became well-known due to its potent insecticidal properties, long-lasting effects, and comparatively inexpensive initial cost. Throughout the Twentieth Era, it was widely employed in agricultural pest management and mosquito control initiatives, particularly to lower the prevalence of vector-borne illnesses like typhus and malaria. Despite its efficacy, DDT's environmental durability and hazardous characteristics quickly made it a topic of international concern. DDT does not easily break down in the environment, in contrary to many biodegradable materials. It stays active for extended duration of time in living tissues, water, and soil. Due of its extremely stable chemical nature, DDT can accumulate in food chains or food webs and last for several years. Since DDT can spread through the air, soil, water, and slits after being inserted into the environment, its persistence poses a serious threat to the environment. DDT residues enter freshwater environments through air deposition, polluted wastewater discharge, agricultural runoff, and inappropriate disposal methods. Consequently, contaminated water, food sources, and natural earth materials, reveal aquatic creatures to DDT either explicitly or implicitly. Fish react rapidly to chemical stressors found in aquatic bodies, making them extremely sympathetic living indicators of aquatic pollution. Fish health is a good indicator of the state of their surroundings. Fish species may endure physiological, biochemical, behavioural, haematological, and histological alterations as a result of any change in the water's chemistry. As a result, fish are frequently utilised in environmental toxicology studies to assess the negative effects of contaminants and pesticides. *Clarias batrachus*, also referred to as walking catfish or magur fish, is a species of freshwater fish that plays a significant role in aquatic culture (aquaculture), fisheries, and experimental toxicity. This resilient freshwater teleost fish is found all over South and Southeast Asia. The species is highly valuable economically because of its nutritional significance, consumer demand, and capacity to adapt to a variety of environmental situations. Because of its exceptional ability to survive and withstand harsh environmental circumstances including low oxygen concentrations and varying water quality, *Clarias batrachus* is especially well-suited for toxicological research [2][3].

Fish pesticide toxicity research frequently concentrates on modifications in blood attributes and tissue architecture. Haematological indicators offer valuable insights into an organism's physiological state and overall health. Oxygen, nutrients, hormones, waste products from metabolism, and immune cells are all transported throughout the body via blood. As a result, any harmful material that alters blood composition has the potential to disrupt several biological processes. Packed cell volume (PCV), red blood cell count (RBC), white blood cell count (WBC), haemoglobin concentration, and associated erythrocytic indices are prominent haematological attributes that are frequently observed in fish toxicology. Stress reactions, anaemia, immunological disorders, metabolic problems, and toxic damage are frequently indicated by variations in these markers [4].

Histopathological study, in addition to haematological investigation, is a useful method for comprehending toxic effects at the tissue level. The microscopic study of structural changes in tissues brought on by illnesses, harmful substances, or environmental stresses is known as histopathology. Histopathological evaluation offers immediate proof of tissue damage and organ dysfunction because internal organs are often the target of hazardous substances[5]. The kidney, liver, and gills are some of the most significant organs that are frequently studied in aquatic toxicological studies. Fish use their gills as their major respiratory organ, and they are always in contact with the water around them. Gills are extremely susceptible to waterborne contaminants since they are directly exposed to the environment. Gas exchange systems or mechanism, ion control, and respiration may all be hampered by structural damage to gill tissues [6][7]. The primary metabolic and detoxifying organ in charge of breaking down harmful compounds that enter the body is the liver. When harmful substances like DDT build up in liver tissue, they may interfere with metabolism, enzyme activity, and cellular structure. Therefore, changes in liver histology are significant indicators of chemical toxicity. Similar to this, the kidneys carry out essential physiological functions related to excretion, osmoregulation, and ionic balance maintenance. Renal tissue damage can impede filtration functions and jeopardise fish physiological equilibrium in usual. These factors make it crucial from a scientific standpoint to look at how DDT affects *Clarias* batrachus's haematological and histological properties or characteristics. These investigations enhance environmental monitoring initiatives targeted at safeguarding aquatic biodiversity and advance knowledge of pesticide toxicity processes [8].

The detrimental effects of pesticides on aquatic life have drawn a lot of attention in recent decades due to the growing vulnerability of freshwater environments to chemical contaminants. Environmental pollution is largely caused by agricultural activities, especially when pesticides are used excessively and frequently. Pesticide residues from agricultural fields are transported into adjacent rivers, ponds, canals, and lakes by rainfall, irrigation runoff, and leaching processes. These substances interact with water, sediments, microbes, plants, and aquatic animals or organisms once they reach aquatic systems or environment, which has an impact on biological productivity and ecological equilibrium [9][10][11].

Fish hold a pivotal function in food chains or food webs among aquatic creatures or organisms and are crucial to preserving ecological stability. Fish are extremely vulnerable to harmful compounds dissolved or suspended in their surroundings because they rely directly on water for respiration, eating, reproduction, and survival. Numerous biological reactions, such as irregular behaviour, changed food habits, respiratory stress, stunted growth, impaired reproduction, immunological disorders, and cellular damage, can be brought on by exposure to chemical pollutants. Changes in behaviour are frequently seen as one of the first signs that fish have been exposed to toxins.

Fish exposed to pesticides may swim erratically, become hyperactive, lose their balance, secrete more mucus, eat less, and have irregular breathing patterns. These behavioural abnormalities typically reflect physiological stress brought on by disruption of metabolic and nervous system functions. Long-term exposure may eventually result in frailty, decreased adaptation, and higher mortality [13].

DDT's detrimental effects on aquatic life are mainly linked to its lipophilic and bioaccumulative characteristics. DDT easily builds up in biological tissues once it enters living or biological things because it is soluble in fats rather than water. DDT remains may gradually build up in muscles, fatty tissues, and organs as a outcome of repeated exposure. Biomagnification, a process where pollutant concentration rises at successive trophic levels of the food chain, makes this buildup more severe [14][15]. Because toxins that are originally present in water in moderate concentrations may later reach hazardous levels in higher organisms, the phenomenon of biomagnification poses a serious ecological risk. Fish that eat tainted food organisms eventually develop pesticide residues in their tissues. As a result, even when environmental concentrations seem relatively moderate, fish habitancy become susceptible to long-term harmful consequences. Fish toxic responses require research at several biological levels [16].It's possible that external behavioural observations won't fully capture the level of toxic damage. As a result, integrated assessment encompassing physiological, biochemical, haematological, and histological analyses is emphasised in contemporary aquatic toxicity. These methods aid scientists in recognising early indicators of toxicity and comprehending the mechanics behind biological changes brought on by pollutants. Haematological research has become more important in environmental toxicity because blood reflects the functional status of an organism. Chemical stress frequently causes changes in blood composition, which may be a sign of problems with respiration, immunity, metabolism, and tissue function. When it comes to physiological adaptation or the development of disease conditions in exposed organisms, blood parameters offer quick and accurate information[12][17][18].

One of the most crucial blood factors that is frequently examined during toxicological examinations is haemoglobin. It is in charge of moving oxygen from respiratory surfaces to bodily tissues and making the removal of carbon dioxide (CO_2) easier. A decrease in haemoglobin concentration may hinder the effectiveness of oxygen delivery and lower metabolic function. Pesticides and other toxic compounds frequently disrupt the production of haemoglobin or harm erythrocytes, which can lead to anaemia in fish. Erythrocytes, another name for red blood cells, are directly linked to the ability to deliver oxygen. A sign of toxic stress and physiological disturbance is often a change in the red blood cell count. Reduced erythrocyte counts can be caused by toxic damage to blood-forming organs, defective erythropoiesis, or the death of blood cells.

On the other hand, aberrant variations in blood parameters could be the result of compensatory physiological reactions brought on by stress in the environment. Another vital component of blood that is involved in immune defence and defence against diseases and foreign substances are white blood cells. The activation of defence mechanisms in response to hazardous exposure is frequently indicated by variations in WBC count. While a protracted disruption of immune function can jeopardise organisms health and disease resistance, a higher leukocyte count may be an adaptive reaction against physiological stress [18]. By offering microscopic proof of tissue-level changes brought on by harmful substances, histopathological examinations supplement haematological research. Because structural alterations typically indicate or represent underlying physiological dysfunction, tissue damage or not work properly is a reliable biomarker for assessing pollutant toxicity. Lesions, degenerative changes, necrosis, congestion, and cellular disarray that would not be apparent by external view alone can be found by researchers using microscopic investigation. Because of their close contact with the surrounding water, gill tissues are especially prominent in aquatic toxicology. Respiration, osmoregulation, acid-base balance, and the excretion of metabolic wastes are only a few of the vital tasks carried out by gills. Gills are extremely susceptible to chemical damage due to their delicate structure and constant contact to the environment. Pesticide exposure can cause lamellar fusion, epithelial lifting, hyperplasia, congestion, and necrosis, which can compromise respiratory function and cause physiological imbalance. Detoxification, metabolism, storage, and metabolic modification of xenobiotic chemicals are mostly handled by the liver. Hepatic tissues frequently handle the body's absorption of toxic substances. As a outcome, the liver becomes a primary target organ when exposed to pesticides. Hepatocellular degeneration, vacuolation, necrosis, and vascular congestion are examples of histological changes that often point to metabolic failure and toxic stress. Filtration, osmoregulation, excretion, and the preservation of internal physiological balance all depend on the kidneys, which are equally significant organs. Toxic damage to renal tissues can interfere with the regulation of ionic and water balance as well as disrupt regular filtration processes. Therefore, structural anomalies in kidney tissues offer important information about how chemical pollutants affect aquatic organisms systemically[19][20]. The ecological significance of freshwater habitats and the general concern about pesticide contamination make a thorough study of DDT toxicity in *Clarias batrachus* extremely pertinent. Thorough evaluation of haematological and histopathological characteristics might offer valuable insights into physiological disorders and tissue damage brought on by pesticide exposure. These results could be useful for monitoring the environment, evaluating fish health, conserving aquatic resources, and managing freshwater ecosystems sustainably [21].

The goal of the current study, "Effect of DDT on Haematological and Histopathological Parameters in *Clarias batrachus*", is to comprehend the harmful effects of DDT on the physiological and tissue health of freshwater fish. Analysing blood parameters and microscopic tissue changes can yield valuable information on the biological stress caused by pesticide exposure. Assessing blood and internal organs can be a useful method for spotting early indications of environmental pollution in aquatic creatures since they react quickly to hazardous substances.The necessity for thorough toxicological research with freshwater fish has been highlighted in recent years due to growing concerns about environmental contamination and diminishing aquatic biodiversity. It is crucial to comprehend the connection between fish biological reactions and pesticide revelation from a scientific standpoint as well as for fisheries management, aquaculture development, and environmental preservation. Studies concentrating on pesticide-induced modifications in fish health contribute considerably toward the development of pollution monitoring systems and sustainable conservation practices. Thus, the goal of this research is to examine how DDT exposure affects specific haematological and histological parameters in *Clarias batrachus* under well monitored experimental settings. The results of this study should contribute to our understanding of the toxicological effects of DDT in freshwater ecosystems and further our understanding of fish physiology, aquatic systems health, and pesticide toxicity assessment.

Statement of Research Problem

Rapid agricultural development, industrialisation, and careless chemical use expose aquatic habitats to a variety of environmental pollutants on a constant basis. Because of their detrimental impacts on aquatic life, persistence, and bioaccumulative nature, pesticides are among the most damaging groups of these pollutants. The organochlorine insecticide dichloro-diphenyl-trichloroethane (DDT) has a lengthy half-life in the environment and a high affinity for biological tissues. Even though many nations have banned the use of DDT in agriculture, its remains can still be found in sediments, water bodies, and aquatic food chains. Fish are extremely sensitive markers of aquatic pollution because their gills, skin, and feeding habits keep them in close proximity to contaminated water. Fish physiological (physical function), biochemical, haematological (blood related), and histological processes can be disrupted by exposure to pesticide contaminants. Blood parameter changes frequently indicate internal physiological stress, whereas histopathological changes show structural damage to important organs. The freshwater catfish species *Clarias batrachus* (Desi Magur) is highly significant in terms of nutrition, economy, and ecology. Because this fish species lives on the bottom and interacts closely with aquatic sediments, it is especially susceptible to pesticide contamination.

Detailed information about DDT-induced haematological and histological alterations in *Clarias batrachus* is still scarce, despite its significance.Thus, the current study was conducted to examine how "DDT affected the histopathological and haematological characteristics of *Clarias batrachus*" in vitro.

Methodology

For research, healthy Desi Magur (*Clarias batrachus*) specimens were gathered from Lalpur Market, Ranchi, Jharkhand. Fish were carefully chosen based on their bodily condition, swimming activity, outward look, and lack of obvious clinical symptoms. In the beginning, sixteen fish specimens were acquired for testing. However, because of the stress of transit and the high temperatures during acclimatisation, mortality was observed. In such experiments, handling disruption, crowding, sudden habitat change, oxygen fluctuation, and temperature variation are prominent causes of transportation stress. These elements may impair immunological response, change metabolic activity, disrupt physiological balance, and raise mortality. As a result, eight fish perished prior to the start of the dose phase trial, while the remaining eight healthy fish were kept for additional research. There were four tanks in the experiment, each holding around 25 litres of water. Before being used, the tanks were properly cleaned to minimise contamination and preserve hygienic laboratory conditions. The experimental setup was as follows:

- **Control Tank — No DDT treatment**
- **Tank A — Low Dose Treatment**
  DDT concentration: 3.3 ml / 25 L water
- **Tank B — Medium Dose Treatment**
  DDT concentration: 6.8 ml / 25 L water
- **Tank C — High Dose Treatment**
  DDT concentration: 13.62 ml / 25 L water

Table 1: Experimental Tank Arrangement

Tank	Treatment	DDT Dose	Water Volume
Control	No DDT	0 ml	25 L
Tank A	Low Dose	3.3 ml	25 L
Tank B	Medium Dose	6.8 ml	25 L
Tank C	High Dose	13.62 ml	25 L

The duration of the dosage phase experiment was 21 days. Water was replaced every 24 hours throughout the trial period. Fresh water was introduced into each tank and fresh DDT solution was again administered according to the designated concentration. During the 21-day exposure period, same procedure was repeated.Because environmental fluctuations may independently affect fish physiology and complicate toxicological interpretation, water temperature and quality were continuously monitored. In order to guarantee the fish's nutritional support and physiological stability, feeding was continued during the experiment.

Research Design and Workflow

Fish Collection
(*Clarias batrachus* sampling)
↓
Acclimatization
(Laboratory environment adaptation)
↓
Tank Grouping
(Allocation into Control, Tank A, B, and C)
↓
DDT Exposure
(Specific dosage administration in 25 L water)
↓
Blood Collection
(Sample extraction for analysis)
↓
Haematology & Histopathology

Statistical Analysis

The significance of changes seen in the haematological and histopathological characteristics of *Clarias batrachus* subjected to various DDT doses was assessed using statistical analysis. The RBC count, WBC count, body weight measurement, and haemoglobin estimation experimental data were all methodically documented and presented as mean values. The impact of DDT exposure was ascertained by comparing the outcomes of the treatment and control groups. Determining whether the observed variations between experimental groups are the result of treatment effects or random variation is made easier with statistical examination. The experimental results are more reliable and have greater scientific value when statistical analysis is performed appropriately. The collected data was totalled and shown using the proper tables, graphs, and charts for simpler interpretation. A comparison of the control, low-dose, medium-dose, and high-dose groups was conducted. The study's statistical findings were utilised to evaluate the degree of physiological changes brought on by DDT exposure and to determine correlations between pesticide concentration and biological reactions.

Table 2: Statistical Parameters

Parameter	Purpose of Analysis
Body Weight	Assessment of growth performance
RBC Count	Evaluation of oxygen carrying capacity
WBC Count	Assessment of immune response
Haemoglobin	Evaluation of blood physiology
Histopathology	Assessment of tissue damage

Observations

Table 3: Body weight across experimental groups

Experiment group	Mean body weight
Control	**52.7**
Low Dose (3.3 ml/25 L)	**74.5**
Medium Dose (6.8 ml/25L	**65.4**
High Dose (13.62 ml/25 L)	**62.1**

There was a discernible difference in body weight across the experimental groups. The low-dose group had the highest mean body weight (74.5 g) compared to the control group's 52.7 g. The high-dose group reported 62.1 g, while the medium-dose group recorded 65.4 g. The body weight of the fish (Clarias batrachus) gradually decreased from the low-dose group to the high-dose group, suggesting that the fish's normal growth and physiological function may be adversely affected by increased DDT concentrations.

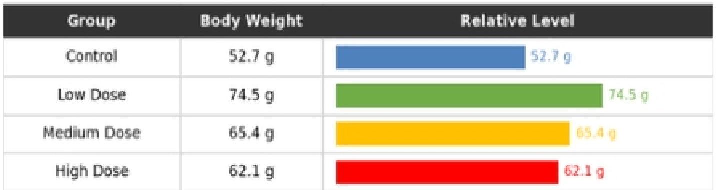

Fig. 1: Body weight measurement of control & DDT treated fish

Haematological Parameters

The effects of DDT exposure on *Clarias batrachus's* haematological parameters after 21 days are displayed in Table 4.

Table 4: Haematological Parameters of Clarias batrachus

Dose Group	RBC (cells/mm³)	WBC (cells/mm³)	Haemoglobin (g/dL)
Control	4.33×10^6	7.50×10^4	8.2
Low dose	4.10×10^6	8.90×10^4	8.1
Medium dose	4.05×10^6	9.87×10^4	7
High dose	1.45×10^6	2.50×10^5	6.5

As the DDT concentration rose, the RBC count progressively declined. The RBC count was 1.45×10^6 cells/mm^3 in the high-dose group and 4.33×10^6 cells/mm^3 in the control group.
On the other hand, the WBC count rose as the DDT concentration rose. The high-dose group recorded the highest value of 2.50×10^5 cells/mm^3, while the control group displayed a WBC count of 7.50×10^4 cells/mm^3. The control group's haemoglobin concentration was 8.2 g/dL, while the high-dose group's was 6.5 g/dL. The decrease was most noticeable for DDT concentrations that were medium and high. The overall haematological results show that exposure to DDT caused anaemic conditions marked by decreased haemoglobin concentration and red blood cell count, together with leukocytosis as shown by an increased white blood cell count.

Fig. 2: RBC Count Comparison (Data Chart)

Group	RBC Count	Relative Level
Control	4.33×10^6	████████████████████ 4.33×10^6
Low Dose	4.10×10^6	███████████████████ 4.10×10^6
Medium Dose	4.05×10^6	███████████████████ 4.05×10^6
High Dose	1.45×10^6	███████ 1.45×10^6

Fig. 3: WBC Count of Control and DDT-Treated Fish (Data Chart)

Group	WBC Count	Relative Level
Control	7.50×10^4	██████ 7.50×10^4
Low Dose	8.90×10^4	███████ 8.90×10^4
Medium Dose	9.87×10^4	████████ 9.87×10^4
High Dose	2.50×10^5	████████████████████ 2.50×10^5

Fig.4: Haemoglobin Comparison Among Experimental Groups (Data Chart)

Group	Haemoglobin	Relative Level
Control	8.2 g/dL	████████████████████ 8.2 g/dL
Low Dose	8.1 g/dL	████████████████████ 8.1 g/dL
Medium Dose	7.0 g/dL	█████████████████ 7.0 g/dL
High Dose	6.5 g/dL	████████████████ 6.5 g/dL

Table 5: Overall Comparison of Haemoglobin, RBC and WBC

Dose Group	Gill	Kidney	Liver	Overall Toxicity
Control	0	0	0	No toxicity
Low Dose	1	1	1	Mild toxicity
Medium Dose	2	2	2	Moderate toxicity
High Dose	3	3	3	Severe toxicity

Histopathological Analysis

The microscopic examination of tissues to assess structural and cellular changes brought on by illnesses, environmental contaminants, and hazardous materials is known as histopathology. It is one of the most crucial methods in toxicological research since it offers concrete proof of tissue damage brought on by chemical pollutants. Understanding the impact of toxicants at the organ and cellular levels through histopathological investigation yields important information about the degree of toxicity.Histopathological alterations are regarded as one of the most trustworthy indicators in toxicology since they show the organism's true tissue damage. Exposure to pesticides can result in structural anomalies in several organs that are visible under a microscope. Cellular degeneration, physiological disturbances, and impaired organ function are often indicated by these alterations.

DDT is a preserving organochlorine insecticide that has been shown to accumulate in aquatic life. Because DDT is lipophilic, it enters fish bodies through their gills, skin, and digestive tracts before travelling through the bloodstream to other organs. Necrosis, inflammation, cellular damage, tissue degradation, and disruption of normal organ architecture can result from long-term exposure to DDT. Histopathological analysis was used in the current study to assess how DDT exposure affected specific *Clarias batrachus* organs. Because of their critical functions in respiration, metabolism, detoxification, excretion, and osmoregulation as well as their extreme sensitivity to environmental contaminants, the gills, liver, and kidney were chosen for examination. Tissue samples from the experimental fish (*Clarias batrachus*) were collected and examined using standard histopathology techniques after the 21-day exposure period. The tissues underwent microtome sectioning, dehydration, clarification, paraffin wax embedding, hematoxylin and eosin staining, and compound microscope examination.

Results and Discussion

The current study shown that the gill, kidney, and liver tissues of *Clarias batrachus* experienced significant histopathological changes when exposed to increasing DDT concentrations. From the control group to the high-dose group, the degree of tissue damage gradually increased, demonstrating a distinct dose-dependent toxic impact.

- The tissue architecture of control fish was normal.
- Mild pathological alterations were caused by low-dose exposure.
- Moderate tissue damage resulted from medium-dose exposure.
- Exposure to high doses caused significant structural damage.

Table 6: Overall Comparison Table

Organ	Control	Low Dose	Medium Dose	High Dose	Severity
Gill	Normal lamellae	Mild thickening	Partial fusion	Severe fusion and clubbing	Severe
Kidney	Normal tubules	Mild degeneration	Vacuolation and congestion	Extensive degeneration	Severe
Liver	Normal hepatocytes	Mild oedema	Moderate vacuolation	Necrosis and severe damage	Severe

Table 7: Histopathological Severity Score

Dose Group	Gill	Kidney	Liver	Overall Toxicity
Control	0	0	0	No toxicity
Low Dose	1	1	1	Mild toxicity
Medium Dose	2	2	2	Moderate toxicity
High Dose	3	3	3	Severe toxicity

Score Key: *0 = Absent | 1 = Mild | 2 = Moderate | 3 = Severe*

Table 8: Total Percentage of Damage

Dose Group	Gill Damage (%)	Kidney Damage (%)	Liver Damage (%)
Control	0	0	0
Low Dose	25	20	20
Medium Dose	55	50	50
High Dose	90	85	80

Discussion

The gills were the first organ affected by DDT exposure because of their close proximity to the aquatic environment. While medium and high doses produced degeneration, lamellar fusion, curling, clubbing, and epithelial lifting, low levels produced modest epithelial hyperplasia and secondary lamellae thickening. These modifications decrease the surface area of the respiratory system and hinder gaseous exchange. As DDT levels increased, the kidney gradually underwent degenerative changes. There was a small amount of tubular congestion and hypertrophy after low-dose exposure.

While high-dose exposure led to significant tubular degeneration, dilatation, vascular congestion, and disruption of normal renal architecture, medium-dose exposure induced vacuolation and renal tubule degeneration. Nephrotoxicity is suggested by these data.
The primary organ for detoxification, the liver, showed hepatocellular changes as exposure to DDT increased. Fish with low doses displayed minor vacuolation and oedema. Fish given a medium dose showed signs of inflammatory infiltration and sinusoidal congestion. After high-dose exposure, hepatic cord disorganisation, localised necrosis, and widespread vacuolar degeneration were indicators of severe hepatotoxicity. All things considered, the pathological alterations that have been noted indicate that DDT causes dose-dependent toxicity in *Clarias batrachus* and impacts respiratory, excretory, and metabolic processes.

Table 9: Total Score Information

Dose Group	Gill	Kidney	Liver
Control	0	0	0
Low Dose	1	1	1
Medium Dose	2	2	2
High Dose	3	3	3

Conclusion

The current study unequivocally shows that DDT causes notable histological changes in the liver, kidney, and gill tissues of *Clarias batrachus*. As DDT concentrations rose, lesions became more severe over time. The kidneys revealed tubular degeneration and congestion, the liver showed vacuolation and hepatocellular necrosis, and the gills showed lamellar fusion and clubbing. The findings verify that DDT has a harmful effect that is dose-dependent and significantly impairs the fish's normal physiology. As a result or outcome, these organs' histopathological changes can be used as trustworthy biomarkers to evaluate the toxicity of pesticides in aquatic environments.

Order of Final Overall Severity

Control < Low Dose < Medium Dose < High Dose

Gill Damage: Control < Low < Medium < High
Kidney Damage: Control < Low < Medium < High
Liver Damage: Control < Low < Medium < High

Reflections

In the gill, kidney, and liver tissues of *Clarias batrachus*, DDT exposure results in progressive and dose-dependent histopathological damage; the high-dose group exhibits the greatest toxicity and tissue deterioration.

Novelty of the Current Research

The current study differs from earlier research on fish pesticide toxicity in a number of ways.

1. Histopathological Evaluation of Many Organs

A single organ, such the liver or gills, has been the subject of the majority of earlier research. The current study assessed the overall harmful effect of DDT by concurrently examining three key organs: the liver, kidney, and gills.

2. Analysis of Dose-dependent Comparisons

Four exposure groups—Control, Low, Medium, and High dose—were assessed in the study, enabling a thorough assessment of the progression of tissue damage. The progression of toxicity is clearly understood as a result.

3. Scoring for Comparative Histopathology

The development of a comparative severity scale allowed for the classification of lesions as mild, moderate, and severe. This rating method facilitates a more accurate assessment of pesticide toxicity.

Table 10: Practical Uses

Field	Application
Toxicology	Assessment of pesticide toxicity
Fisheries	Monitoring fish health
Ecology	Environmental pollution studies
Aquaculture	Water quality management
Environmental Science	Biomarker development

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