SJSR Singapore Journal of Scientific Research

OPEN ACCESS

In vitro Assessment of Antioxidant Enzymes in *Oreochromis niloticus* Induced with Linear Alkyl Benzene Sulfonates

[1]*Thomas Ohwofasa Ikpesu and* [2]*Christina Ngozi Opara*
[1]*Department of Biology, Federal University Otuoke, Otuoke, Bayelsa, Nigeria*
[2]*Department of Microbiology, Federal University Otuoke, Otuoke, Bayelsa, Nigeria*

ABSTRACT

Background and Objective: Linear Alkyl Benzene Sulfonates is one of the most overused surfactants and one of the most challenging emerging poisons that wastewater treatment plants and other users constantly release into the environment. This study evaluated the toxicity of Linear Alkyl Benzene Sulfonates (LABS) in post-juveniles of Nile tilapia, a common wetland fish. **Materials and Methods:** The fish were self-bred in a serene atmosphere and were induced with low doses of LABS for thirty days after which changes in clinical indices were evaluated in the major tissues in the fish. The glutathione S-transferase (GST), superoxide dismutase (SOD), catalase (CAT), globulin and albumin. GST and SOD activities in the red blood corpuscles were assessed at 7-day intervals, CAT was accessed in the liver, kidney and gills on days 2nd and 30th. Similarly, the globulin and albumin in the sera were measured on days 2nd and 30th. The enzyme activities were evaluated with standardized methods. **Results:** A duration and concentration-dependent rise of GST activity was seen in the erythrocytes of the circhlid induced with different toxicant concentrations. The SOD activity did not markedly differ between treatments ($p>0.05$) and they were indistinguishable from the reference treatment. The activity of catalase was in the order of, liver>gills>kidney and when compared with the untreated fish, the enzyme activities in the organs were significantly ($p<0.05$) higher. Compared to the control, albumin and globulin were suppressed over time and at different doses, although the decrease was significant ($p<0.05$) in all treatments except 0.20 mg L^{-1} of LABS treatment. **Conclusion:** The present investigation has attempted to provide a framework for the implication of one of the frequently readily available surfactants in coastal counties, observations that call for the precautionary use of detergents in this region.

KEYWORDS

Linear alkyl benzene sulfonates, *Oreochromis niloticus*, liver, kidney, erythrocytes, biomarkers

INTRODUCTION

Surfactants are routinely dumped into sewers after usage and predominantly enter the environment through the discharge of treated or untreated wastewater, they are one of the most significant organic chemicals in the environment. When it comes to a wide range of industries from healthcare and cosmetics to oil extraction and processing, surfactant management has evolved into a key concern[1-5]. A system's

ISSN: 2152-0461 (Online)
ISSN: 1816-4919 (Print)
https://doi.org/10.3923/sjsr.2023.88.97

Received: 27 Apr. 2023
Accepted: 21 Aug. 2023
Published: 2 Sep. 2023

non-covalent interactions, such as dipole-dipole, hydrogen bonding and dispersion forces, can drive these surfactants to form a variety of self-assembly structures. Surfactants include cleansers, foam boosters, solubilizers, emulsifiers and suspending agents.

The influence of nonionic surfactants on environmental qualities has become increasingly important to regulators and downstream users in recent years[6,7]. As a result, the momentous surfactant disintegration as a fundamental performance factor has grown and numerous industry-led initiatives, both voluntary and mandated, are being undertaken to limit the amount of chemicals discharged into the environment[8,9]. The implementation of severe requirements for industrial applications, as well as initial and final biodegradability, was required to establish standardized performance characteristics of surfactants that have had an impact on the worldwide detergents industry. Surfactants with good safety profiles are in high demand as a result of the broad adoption of the globally harmonised system of classification[10].

There is a need to measure the levels of surfactants in different compartments due to the large amounts that are consumed (roughly 10 million tonnes per year worldwide) and released into wastewaters, as well as their toxicity to aquatic organisms (LC_{50} values in the mg L^{-1} range). Future growth of this demand is anticipated as surfactants are increasingly used globally.

Linear alkyl benzene sulfonates are all high production volume (HPV) and "down-the-drain" compounds in laundry and personal care items, which are used extensively worldwide. Low levels of these compounds are eventually discharged into the environment through wastewater effluent. These and other surfactants are the main cleaning components common in both home and personal care products. LABS, which has a hydrophobic alkyl chain and a hydrophilic head with a benzene ring and a sulfonate group, is only made from petrochemical by-products. It isn't just one substance, rather, it's preferably a blend of 20 substances, including isomers and homologs of the same molecule.

Tilapia are major fish species raised in Southern Nigeria. These pisces are very significant in aquaculture, thus research into their biological responses to toxins is unavoidable. Furthermore, tilapia species have certain functional properties that allow them to adapt to a variety of environmental circumstances. In this research, the fish's blood, tissues and sera were examined as indications of their physiological status. These measures are increasingly being used in environmental monitoring as useful indicators of toxicant-induced physiological changes in living systems[11,12]. Therefore, this study was conducted to assess the long-term effects on *Oreochromis niloticus*, a common freshwater fish in Nigeria, of a low concentration of linear alkyl benzene sulfonates

MATERIALS AND METHODS

Study area: This study was carried out at the Ecotoxicology Unit, Department of Biology, Federal University Otuoke, Nigeria. The investigation started in April, 2022 and ended in February, 2023.

Materials/reagents: Linear alkyl benzene sulfonates (96% purity) were procured from the Indian company Manuchar, ethyl 3-aminobenzoate methanesulfonate salt was bought from Sigma Aldrich and all other reagents were of technical quality.

The tools, reagents and kit used to analyse the GST were purchased from Sigma Aldrich.com, Saint Louis Missouri 63103, United States of America e.g., 1-Chloro-2,4-dinitrobenzene (CDNB), 10 mM glutathione buffer: 50 mM Tris, 10 mM reduced glutathione, pH 8.0 (make fresh daily), temperature controlled UV/visible spectrophotometer with UV light, quartz cuvette 1 mL, catalog number S10SM. UV 96-well plate, water-17 MW, from., PBS/EDTA/PMSF: 1×PBS, 5 mM EDTA, 0.15 mM PMSF, pH 7.4 purchased from Sigma Aldrich.com, Saint Louis Missouri 63103, United States of America.

SOD assay kits (purchased from 152 Grove Street Itham, MA 02453, United States of America): Reaction initiation solution, SOD detection buffer, a water-soluble tetrazolium salt, enzyme solution, reaction initiation solution (40×).

Catalase assay kits (obtained from14780 Memorial Drive, Suite 108, Houston, Texas, 77079, United States of America): 1.0 M potassium phosphate dibasic solution,1.0 M potassium phosphate monobasic solution, potassium hydroxide, hydrochloric acid, hydrogen peroxide 30% (w/w), digital calliper, cuvettes and thermostatted spectrophotometer (obtained from14780 Memorial Drive, Suite 108, Houston, Texas, 77079, United States of America).

Pre-analytical stage: One hundred and fifty adult *O. niloticus* of mean weight (1.2±0.03 kg) and length (19.25±0.05 cm) were used for spawning and were obtained from a private fish from Delta State, Nigeria. The fish were cultured in a well-made shallow clayey pond with one male to five females between April and May, 2021 and a total of five shallow ponds were built, with a total of 5 males and 25 female breeding stocks used.

The breeding stocks were fed protein- and vitamin-rich feeds throughout the experiment and the spawning aquariums were cleaned every four to 5 days.

Males often select the most developed female to mate with before chasing the other females to the furthest part of the aquarium. It can take up to 2 hrs for the entire spawning and fertilization process. The female quickly leaves the aquarium's nesting area while holding the fertilized eggs in her mouth. No feeding throughout incubation and until the born fry has finished absorbing the yolk sac because their mouths are already full-first with eggs, then with young and tender fries. The male selects and mates with a different female soon after the female departs the nest and the cycle is repeated.

The fries were carefully placed in a nursing pond to mature once the yolk sac was absorbed. They grow to a length of 1-2 cm, eat by themselves and resemble fish. Fingerling is a fry that has reached 10 to 15 cm, or around the size of a finger. In 45 to 60 days, the fry will grow to fingerling size. A specifically formulated diet composed of rice bran and soybean powder in a 1:1 ratio was given to the fry and fingerlings 4-5 times each day. The nursery pond was netted out after 2 to 3 weeks and advanced fries were transferred to the rearing pond until the fingerlings stage was obtained.

A greenhouse was constructed and regularly maintained to mimic the fish's natural environment. The clayey loam soil was used to build the fifteen earthen ponds, each of which was 27 1/4×24 1/8×29/12 inches in size. For 12 weeks, ten fingerlings in the last ponds were each fed with rice bran and finely ground cake. After cleaning the aquariums, syphoning the old water with a hand pump. Fresh water was added every other day.

Analytical stage: In the final ponds, the fish were subjected to a range of LABS concentrations (0.00, 1.0, 1.5, 2.0 and 2.5 mg L^{-1}) after the completion of the 12th week for a total of 30 days. Fish in the control and experimental groups received twice-daily meals at 3% of their body weight throughout the experiment. The earthen ponds were kept as clean as possible and the water and poisonous substances were changed every 24 hrs.

Regular measurements of the water's physicochemical characteristics were taken throughout the experiment. A fish from each pond is taken out after each research period, brought into the lab in a vented container and quickly put to sleep with MS222. Ethyl 3-aminobenzoate methanesulfonate salt, Sigma.

Glutathione S-transferase and superoxide dismutase responses to various stresses were assessed every seven days. On days 2nd and 30th, albumin and globulin in the serum as well as catalase activity in the liver, kidney, gills and globulin were determined. Because each organ reacts differently, variations in evaluation times were attributed to this and absorption, distribution, metabolism and excretion were utilized as a baseline.

Tissue paper was used to dry the mucous and water from the fish's body. To improve grip, the fish head was wrapped in a towel. Using insulin syringes, blood was drawn from a vein extending ventrally down the spinal column. The tubes were then centrifuged at 3000 rpm for five minutes to remove the serum and they were then kept at -80°C for later examination. Fish were slain soon after blood was collected and the relevant organs (liver, kidney and gills) were removed for enzymatic and biochemical tests.

To evaluate GST and SOD activities were analyzed spectrophotometrically, using their respective assay kits. For catalase determination, 100 liters of sample or standard were incubated with 100 liters of 1% Triton-X and 100 L of 30% hydrogen peroxide. A digital calliper was used to measure the height of the O_2 foam generated after 15 min. The reaction's specificity was tested with samples containing 10 mM sodium azide. The activity of catalase is measured in U mg^{-1} protein units.

The most abundant protein component of serum is albumin. It is produced in the liver and is known for its capacity to change the configuration. Because of its steric affinity, albumin may transport a variety of chemicals, including bilirubin, fatty acids, uric acid, different medicines and antibiotics. Albumin is also involved in maintaining adequate osmotic pressure. Serum albumin levels that are elevated are linked to dehydration. Low serum and albumin levels can signal malnutrition, liver disease, kidney problems and rheumatoid arthritis. The BCG (Bromocresol Green) albumin assay kit (MyBioSource, Inc., San Diego, Canada 92195-3308, United States of America) was used to determine the albumin contents in the serum of the fish and absorbance was measured at 628 nm against a blank. Albumin was subtracted from the total protein to get the globulin content.

Ethical consideration: The study received ethical approval from the Federal University Otuoke Ethical Committee (FUOEC), Nigeria. The research was also carried out following the policies specified in the Guide for the Care and Use of Laboratory Animals.

Statistical analysis: The three technical replicates of each treatment were used for all analyses (n = 3). For each experimental group, the standard error values (SE) and individual enzyme activity were determined using the average values. GraphPad Prism was used to conduct the statistical analysis (Version 9.0.0, GraphPad Software, Inc., La Jolla, Canada, United States of America) and One-way ANOVA was used to compare the groups (control and exposed fish). A posthoc Bonferroni test was used to analyze the results and statistical significance was defined as $p<0.05$ and $p<0.01$, respectively.

RESULTS

In this investigation, GST activity in the erythrocytes of *O. niloticus* exposed to various concentrations of LABs showed duration and concentration-dependent elevation. The enzyme activity in the fish samples at 0.05 mg L^{-1} on days 2nd to 30th was 34.11±0.12, 43.23±0.30, 49.30±0.31, 52.60±0.70 and 27.50±0.60 umg^{-1} protein. At 0.10, it ranges between 37.00-58.91 mg mL^{-1}, at 0.15, it ranges between 39.11-66.11 mg mL^{-1} and at 0.20, the range was 45.30-69.40 mg mL^{-1}.

On day 2, no significant difference ($p>0.05$) between the enzyme activity and various treatments, except for the 2.50 treatment which was slightly significant ($p<0.05$). However, on days 9th, 16th, 23rd and 30th the enzyme activity in the erythrocyte of the fish varied significantly ($p<0.05$ and $p<0.01$) in all the treatments and the control (Fig. 1).

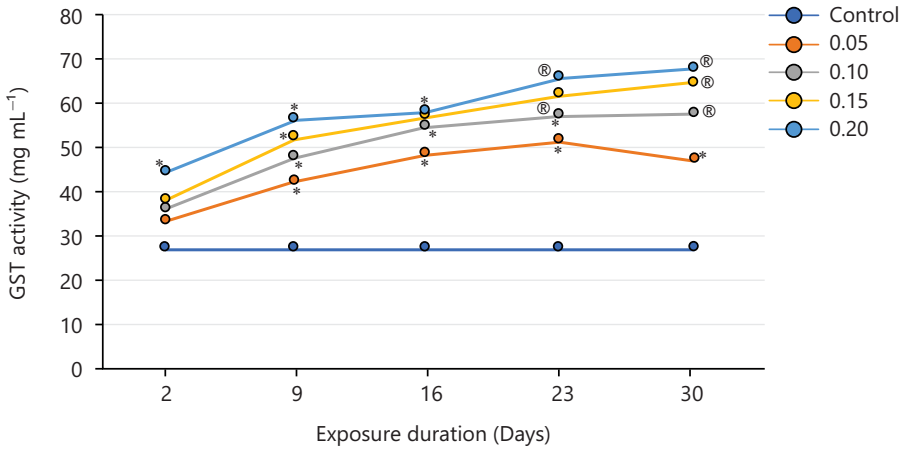

Fig. 1: GST activities in the erythrocytes of *O. niloticus* exposed to sublethal concentrations of linear alkyl benzene sulfonates

A symbol indicates a significant different *($p<0.05$) and ®($p<0.01$) between the control and various exposures

Table 1: SOD activity (u/mgHb) in the erythrocytes of *O. niloticus* exposed sublethal concentrations of Linear Alkyl Benzene sulfonates

	Mean±SE				
Concentration	Days 2	Days 9	Days 16	Days 23	Days 30
0.000	5.40±0.37[a]	5.26±0.03[a]	5.39±1.12[a]	5.20±0.50[a]	5.48±0.32[a]
0.050	5.70±0.53[a]	5.13±0.30[a]	5.15±0.79[a]	5.30±0.28[a]	5.80±1.10[a]
0.100	6.80±0.28[a]	4.83±0.10[a]	4.80±0.30[a]	4.20±0.10[a]	4.08±0.80[a]
0.150	6.82±0.01[a]	4.88±0.30[a]	4.40±0.40[a]	4.10±0.60[a]	4.02±0.25[a]
0.200	9.88±0.17[a]	5.20±0.13[a]	4.11±0.10[a]	3.80±0.30[a]	3.80±0.01[a]

*[a]Not significant ($p>0.05$)

The SOD activities in *O. niloticus* erythrocytes subjected to sublethal doses of linear alkyl benzene sulfonates. Though the activities did not differ significantly ($p>0.05$) between treatments and were virtually similar to the control group, there was some variance (Table 1). The enzyme activities decline as the toxicant concentration and exposure time rise. The enzyme activities on day 2 vary from 5.40±0.37 to 9.88±0.17 u/mgHb. On day 9, SOD activity varies between (5.13±0.30) and (5.26±0.03) u/mgHb, on day 16, between (4.11±0.10) and (5.39±1.12) u/mgHb, on day 23, between (3.80±0.30) and (5.30±0.28) u/mgHb and on day 30, between (3.80±0.01) and (5.80±1.10) u/mgHb.

Responses of catalase in the examined tissues of *O. niloticus* exposed to different concentrations of LABS are shown in Table 2. With the control as the reference, the toxicant induced the enzyme in all the tissues and was dose and time-dependent. Results of this study showed that catalase activity significantly ($p<0.05$ and $p<0.01$) increased in the liver, gills and kidney compared to control at the higher concentrations. At lower doses and early periods of the study, the enzyme activity was comparable with the control.

The albumin and globulin contents in the plasma of *O. niloticus* exposed to various concentrations of LABS are shown in Table 3. Both markers were inhibited with time and the concentrations when compared with the control. The decreasing order with exposure concentrations in albumin level (mg/100 mL) on day 2 are, 5.30±0.35 (control)>4.92±0.02 (0.05)>3.68±0.30 (0.10)>3.20±0.10 and (0.15)>2.50±0.35 (0.20). Though, the decrease was conspicuous but was not significant ($p>0.05$) in all the treatments except 0.20 which was significant at a 0.05 level.

Also, on day 30th, the range of albumin level (mg/100 mL) in the treated group was 2.60±0.04-1.20±0.05 mg/100 mL. Unlike day 2, The prolonged exposure at 0.05 mg/L of LABS was not significant ($p>0.05$), but other concentrations were highly significant ($p<0.05$) and the level of significance is concentrations dependent (Table 3).

Table 2: Changes in the activity of catalase (units/min/mg protein) in the liver, gills and kidney of *O. niloticus* on exposure to linear alkyl benzene sulfonates

	0.00 ($\bar{x}$±SE)		0.05 ($\bar{x}$±SE)		0.10 ($\bar{x}$±SE)		0.15 ($\bar{x}$±SE)		0.20 ($\bar{x}$±SE)	
Concentration	2 days	30 days	2 days	30 days	2 days	30 days	2 days	30 days	2 days	30 days
Liver	17.20±0.02[a]	17.40±0.01[a]	23.90±0.17[a]	29.10±0.13[b]	27.11±0.24[b]	32.40±0.05[b]	29.60±0.30[b]	37.10±0.60[b]	29.50±0.10[c]	43.30±0.80[c]
Gills	7.10±0.20[a]	7.13±0.12[a]	11.90±0.50[a]	16.10±0.30[b]	13.20±1.02[a]	18.70±0.70[b]	15.10±0.20[b]	21.40±0.70[c]	15.60±0.10[b]	28.11±1.18[c]
Kidney	13.20±1.20[a]	13.30±0.28[a]	15.20±0.80[a]	21.19±0.50[b]	15.80±0.06[a]	21.80±0.20[b]	17.80±0.070[a]	25.40±1.05[b]	21.10±2.19[b]	33.12±0.48[c]

*[a]Not significant ($p<0.05$) [b]significant ($p<0.05$), [c]highly significant ($p<0.01$) and ±: Plus or minus standard error

Table 3: Variation in albumin and globulin contents in the serum of *O. niloticus* exposed to sub-lethal concentrations of sodium linear alkyl benzene sulfonates

	Albumin (mg/100 mL)		Globulin (mg/100 mL)	
Concentration (mg L^{-1})	2 day ($\bar{x}$±SE)	30 day ($\bar{x}$±SE)	2 day ($\bar{x}$±SE)	30 day ($\bar{x}$±SE)
0.00	5.30±0.35*	5.28±0.70*	7.15±0.18*	7.20±0.42*
0.05	4.92±0.02*	2.60±0.04$^{\beta}$	6.12±0.32*	4.80±0.73*
0.10	3.68±0.30*	2.10±0.02$^{\beta}$	3.80±0.20$^{\beta}$	2.90±0.02$^{\beta}$
0.15	3.20±0.10*	1.98±0.30®	3.20±0.40®	1.90±0.08®
0.20	2.50±0.35$^{\beta}$	1.20±0.05®	3.10±0.03®	1.30±0.05®

Not significant ($p<0.05$), β: Significant ($p<0.05$)$^{\beta}$, ®: Highly significant ($p<0.01$)®

Globulin level decreases with exposure duration and increases in the concentrations of the toxicant. On day 2, the globulin content at 0.05, 0.10, 0.15 and 0.20 mg L^{-1} of LAB's toxicity were, 7.15±0.18, 6.12±0.32, 3.80±0.20, 3.20±0.40 and 3.10±0.03 mg/100 mL, respectively. The globulin content varies significantly ($p<0.05$ and $p<0.01$) in all the treatments except 0.05 treated fish when compared with the control. On day 30th, globulin level ranges from 4.80±0.73 to 1.30±0.05 in fish treated with 0.05 to 0.20 mg L^{-1} of LABS and the level of significance ($p<0.05$ and $p<0.01$) is concentration dependent (Table 3).

DISCUSSION

In this investigation, the biomarkers examined were either induced or inhibited and were concentrations and time-dependent. Biochemical biomarkers are being used more frequently in ecological risk assessments of the environment to determine the occurrence and effects of xenobiotics. This is due to the possibility that it could act as an immediate early warning indicator of possibly adverse stressor outcomes. Biochemical biomarkers should be able to identify subcellular effects before they become apparent at larger levels of biological organization[13].

Fish responses to stress were revealed to rely on both time and stress across all analyzed biological components. By catalyzing their conjugation with glutathione, GST actively contributes to the detoxification of both ROS and lipophilic compounds, such as pesticides, in living organisms. The induction of GST in this study is similar to the findings of other research in fish exposed to agricultural pesticides[14,15] and this investigation's increase in GST activity suggested a rise in ROS production as a result of the surfactant exposure. The increase in the GST activity may indicate that the liver detoxification mechanism has been induced[16].

The SOD is one of the essential enzymes that serve as the first line of defense against pro-oxidants and catalyzes and the transition of superoxide radicals to H_2O_2 and O_2[17]. It is known that toxic stress alters the activity of SOD in the important tissues of fish. In the current investigation, fish exposed to sublethal concentrations of LABs showed an initial rise in SOD activity up to 48 hrs, followed by a decline as the experiment concluded. Superoxide radical anion synthesis was indicated by a spike in SOD activity at the beginning of the experiment and the suppression at the end may have resulted from more oxyradical formation than the enzyme was able to neutralize. Additionally, in a few rare cases, the superoxide radical has been shown to significantly oxidize the cysteine in the enzyme and reduce SOD activity, either by itself or after being converted to H_2O_2[18].

To participate in their detoxification processes, catalase catalyzes the reduction of ROS[19,20]. Increased level of CAT in the fish exposed to low doses of linear alkyl benzene sulfonates in this study, both in the concentrations and durations may be due to the removal of ROS from the cell generated by the surfactant's exposure[21] and is consistent with results of previous studies in fish exposed to other contaminants such as herbicides[21-23]. In contrast to this finding Oruc and Usta[24] observed in

Heteropneustes' skeletal muscle and brain's CAT activity was significantly reduced after exposure to chlorpyrifos and *Ctenopharyngodon idellus'* gills, kidney and liver's CAT activity were also significantly decreased after exposure to chlorpyrifos[25,26]

Primary factors influencing the synthesis of albumin in the liver include protein and amino acid nutrition, colloidal osmotic pressure, hormones, disease states and stress. Enhanced fluid retention in tissue spaces caused by edoema in the fish led to a shift of fluid from the intravascular to the interstitial space, resulting in intravascular volume depletion. In this study, the decrease in albumin in fish treated with low doses of sodium lauryl sulphate may be attributed to a drop in osmotic pressure, which leads to increased fluid retention in tissue spaces, resulting in a shift of fluid from the intravascular to the interstitial space, resulting in intravascular volume depletion and edoema formation. Proteinuria caused by kidney disease, which is characterized by tubular epithelial degeneration and hyaline casts in the lumen[27], may be attributed to serum albumin decreases. The serum albumin results from these findings are comparable with those obtained by Velisek *et al.*[28] and they emphasize that hypoalbuminemia in fish can occur as a result of pollution exposure and other stressful situations. The plasma globulin levels were significantly decreased in the fish treated with different doses of sodium lauryl sulfate. Reduced globulin was most likely caused by lower levels of immune-related proteins such as lysozyme, complement components, antimicrobials and peptides. Another study found that fish exposed to various pollutants and pesticides had lower globulin levels[29,30]. Nkpondion *et al.*[31] indicate a rise in the liver globulin level of fish subjected to detergent, which contradicts these findings.

Water naturally can neutralize contamination, but when contamination gets out of hand, water loses its ability to generate itself. Therefore, regular monitoring and control of pollutant discharge into the local aquatic environment are necessary. By imposing permissible quantities of pollutants and preventing their release into water resources, water quality laws can help conserve aquatic ecosystems. Use detergents with little to no phosphate because the eutrophication of lakes is caused by high phosphate content. Manage the runoff of stormwater. Stormwater runoff gathers trash, sediments, chemicals and other pollutants when it flows over impermeable surfaces. If the runoff is not treated, these pollutants may have a negative impact on the water quality.

CONCLUSION

This study has shown that fish biochemical enzymes can significantly change as a result of exposure to surfactants. These enzymes are useful biomarkers that can be utilized to detect surfactant pollution in aquatic systems. The findings also suggested that aquatic creatures' health may be harmed by the presence of surfactants in water bodies. From the results of the current investigation, it can be inferred that the fish are experiencing significant oxidative stress because of the reaction of the antioxidant enzymes. Water naturally can neutralize contamination, but when contamination is above the carrying capacity, water loses its ability to generate itself. Therefore, regular monitoring and management of pollution discharge into the local aquatic environment are necessary. By enforcing permissible quantities of pollutants and limiting their discharge into water resources, water quality laws can aid in the protection of aquatic ecosystems.

SIGNIFICANCE STATEMENT

One of the most challenging pollutants that wastewater treatment facilities routinely release into the environment is surfactants. This study has demonstrated that exposure to LABs can ultimately result in substantial harm to aquatic life and may have similar effects on human health. Since anthropogenic factors are the primary sources of surfactant exposure, concerned authorities must enact stringent mitigation measures to curb this underappreciated risk.

REFERENCES

1. Chaudhari, S.P. and R.P. Dugar, 2017. Application of surfactants in solid dispersion technology for improving solubility of poorly water soluble drugs. J. Drug Delivery Sci. Technol., 41: 68-77.
2. Hayes, D.G., 2017. Fatty Acids-Based Surfactants and Their Uses. In: Fatty Acids: Chemistry, Synthesis, and Applications, Ahmad, M.U. (Ed.), AOCS Press, Urbana, Illinois, ISBN: 9780128095218, pp: 355-384.
3. Schramm, L.L., 2000. Surfactants: Fundamentals and Applications in the Petroleum Industry. Cambridge University Press, Cambridge, UK., ISBN: 9780521640671, Pages: 621.
4. Echaide, M., C. Autilio, R. Arroyo and J. Perez-Gil, 2017. Restoring pulmonary surfactant membranes and films at the respiratory surface. Biochim. Biophys. Acta (BBA)-Biomembr., 1859: 1725-1739.
5. Romsted, L.S., 2014. Surfactant Science and Technology: Retrospects and Prospects. CRC Press, Boca Raton, Florida, ISBN: 9781439882955, Pages: 593.
6. Lacasse, K. and W. Baumann, 2012. Textile Chemicals: Environmental Data and Facts. Springer Science & Business Media, Berlin/Heidelberg, Germany, ISBN: 9783642188985, Pages: 1184.
7. Ohwofasa, I.T., 2022. Impact surfactants on aquatic resources with references to neurological inhibition, antioxidant defenses, and endocrine disruption in *Oreochromis niloticus* induced with alpha-olefin sulfonate. J. Toxicol. Risk Assess., Vol. 8. 10.23937/2572-4061.1510050.
8. Wenzel, A., W. Böhmer, J. Müller, H. Rüdel and C. SchrÖter-Kermani, 2004. Retrospective monitoring of alkylphenols and alkylphenol monoethoxylates in aquatic biota from 1985 to 2001: Results from the German environmental specimen bank. Environ. Sci. Technol., 38: 1654-1661.
9. Tenji, D., B. Micic, S. Sipos, B. Miljanovic, I. Teodorovic and S. Kaisarevic, 2020. Fish biomarkers from a different perspective: Evidence of adaptive strategy of *Abramis brama* (L.) to chemical stress. Environ. Sci. Eur., Vol. 32. 10.1186/s12302-020-00316-7.
10. Johnson, P., A. Trybala, V. Starov and V.J. Pinfield, 2021. Effect of synthetic surfactants on the environment and the potential for substitution by biosurfactants. Adv. Colloid Interface Sci., Vol. 288. 10.1016/j.cis.2020.102340.
11. Mazon, A.F., E.A.S. Monteiro, G.H.D. Pinheiro and M.N. Fernandes, 2002. Hematological and physiological changes induced by short-term exposure to copper in the freshwater fish, *Prochilodus scrofa*. Braz. J. Biol., 62: 621-631.
12. Schjolden, J., J. Sørensen, G.E. Nilsson and A.B.S. Poléo, 2007. The toxicity of copper to crucian carp (*Carassius carassius*) in soft water. Sci. Total Environ., 384: 239-251.
13. Dey, S., P. Samanta, S. Pal, A.K. Mukherjee, D. Kole and A.R. Ghosh, 2016. Integrative assessment of biomarker responses in teleostean fishes exposed to glyphosate-based herbicide (Excel Mera 71). Emerging Contam., 2: 191-203.
14. Iwase, T., A. Tajima, S. Sugimoto, K. Okuda and I. Hironaka *et al*., 2013. A simple assay for measuring catalase activity: A visual approach. Sci. Rep., Vol. 3. 10.1038/srep03081.
15. Olsen, T., L. Ellerbeck, T. Fisher, A. Callaghan and M. Crane, 2001. Variability in acetylcholinesterase and glutathione *S*-transferase activities in *Chironomus riparius* meigen deployed *in situ* at uncontaminated field sites. Environ. Toxicol. Chem., 20: 1725-1732.
16. Jin, Y., X. Zhang, L. Shu, L. Chen and L. Sun *et al*., 2010. Oxidative stress response and gene expression with atrazine exposure in adult female zebrafish (*Danio rerio*). Chemosphere, 78: 846-852.
17. Blahová, J., L. Plhalová, M. Hostovský, L. Divišová and R. Dobšíková *et al*., 2013. Oxidative stress responses in zebrafish *Danio rerio* after subchronic exposure to atrazine. Food Chem. Toxicol., 61: 82-85.
18. Santos, D., M. Melo, D. Mendes, I. Rocha, J. Silva, S. Cantanhêde and P. Meletti, 2014. Histological changes in gills of two fish species as indicators of water quality in Jansen Lagoon (São Luís, Maranhão State, Brazil). Int. J. Environ. Res. Public Health, 11: 12927-12937.
19. Alves, S.R., P.C. Severino, D.P. Ibbotson, A.Z. da Silva, F.R. Lopes, L.A. Saenz and A.C. Bainy, 2002. Effects of furadan in the brown mussel *Perna perna* and in the mangrove oyster *Crassostrea rhizophorae*. Mar. Environ. Res., 54: 241-245.

20. Regoli, F. and M.E. Giuliani, 2014. Oxidative pathways of chemical toxicity and oxidative stress biomarkers in marine organisms. Mar. Environ. Res., 93: 106-117.
21. Stara, A., J. Machova and J. Velisek, 2012. Effect of chronic exposure to simazine on oxidative stress and antioxidant response in common carp (*Cyprinus carpio* L.). Environ. Toxicol. Pharmacol., 33: 334-343.
22. Guilherme, S., I. Gaivão, M.A. Santos and M. Pacheco, 2012. DNA damage in fish (*Anguilla anguilla*) exposed to a glyphosate-based herbicide-elucidation of organ-specificity and the role of oxidative stress. Mutat. Res. Genet. Toxicol. Environ. Mutagen., 743: 1-9.
23. Xing, H., S. Li, Z. Wang, X. Gao, S. Xu and X. Wang, 2012. Oxidative stress response and histopathological changes due to atrazine and chlorpyrifos exposure in common carp. Pestic. Biochem. Physiol., 103: 74-80.
24. Oruc, E.O. and D. Usta, 2007. Evaluation of oxidative stress responses and neurotoxicity potential of diazinon in different tissues of *Cyprinus carpio*. Environ. Toxicol. Pharmacol., 23: 48-55.
25. Kaur, M. and R. Jindal, 2017. Oxidative stress response in liver, kidney and gills of *Ctenopharyngodon idellus* (Cuvier & Valenciennes) exposed to chlorpyrifos. MOJ Biol. Med., 1: 103-112.
26. Obianime, A.W. and I.I. Roberts, 2009. Antioxidants, cadmium-induced toxicity, serum biochemical and the histological abnormalities of the kidney and testes of the male wistar rats. Niger. J. Physiol. Sci., 24: 177-185.
27. Chen, C.Y., G.A. Wooster and P.R. Bowser, 2004. Comparative blood chemistry and histopathology of tilapia infected with *Vibrio vulnificus* or *Streptococcus iniae* or exposed to carbon tetrachloride, gentamicin, or copper sulfate. Aquaculture, 239: 421-443.
28. Velisek, J., Z. Svobodova and J. Machova, 2009. Effects of bifenthrin on some haematological, biochemical and histopathological parameters of common carp (*Cyprinus carpio* L.). Fish Physiol. Biochem., 35: 583-590.
29. Banaee, M., A. Sureda, A.R. Mirvaghe and K. Ahmadi, 2011. Effects of diazinon on biochemical parameters of blood in rainbow trout (*Oncorhynchus mykiss*). Pestic. Biochem. Physiol., 99: 1-6.
30. Atli, G. and M. Canli, 2010. Response of antioxidant system of freshwater fish *Oreochromis niloticus* to acute and chronic metal (Cd, Cu, Cr, Zn, Fe) exposures. Ecotox. Environ. Safe., 73: 1884-1889.
31. Nkpondion, N.N., O.A. Ugwumba and I.K. Esenowo, 2016. The toxicity effect of detergent on enzymatic and protein activities of African mud catfish (*Clarias gariepinus*). J. Environ. Anal. Toxicol., Vol. 6. 10.4172/2161-0525.1000361.