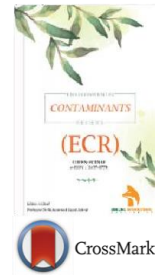


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REVIEW ARTICLE

NITRIC OXIDE AS A POTENTIAL BIOMARKER FOR MONITORING ENDOCRINE DISRUPTING POLLUTANTS IN AQUATIC ENVIRONMENTS

Vedastus W. Makene

Department of Biological and Food Sciences, The Open University of Tanzania. P.O. Box 23409, Dar es Salaam, Tanzania.

*Corresponding Author Email: vmakene@gmail.com

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ABSTRACT

The contamination of aquatic environments by endocrine-disrupting chemicals (EDCs) endangers both environmental and human health. Traditional detection methods, such as chromatography-mass spectrometry (GC-MS) or high-performance liquid chromatography (HPLC), though accurate, are expensive, time-consuming, and require sophisticated equipment. Consequently, there is increasing interest in identifying efficient, biology-based biomarker alternatives. Nitric Oxide (NO) has emerged as a promising biomarker for detecting EDCs because of its role in cellular signalling and oxidative stress responses to environmental pollutants. While research indicates NO's potential for water quality monitoring, its use as a standard biomarker remains to be explored and validated further. This review examines the biological mechanisms underlying NO's response to EDC exposure, its application in environmental monitoring, and the benefits and challenges of NO-based assessment methods. The paper also addresses existing knowledge gaps, including the absence of standardised methods for NO monitoring, limited understanding of species-specific responses, and uncertainties related to long-term environmental changes. Additionally, it suggests future research directions to incorporate NO as a biomarker for water quality evaluation. Using NO as a biomarker for EDCs could improve water pollution surveillance and help mitigate ecological and health risks associated with endocrine-disrupting pollutants in aquatic environments.

KEYWORDS

Nitric Oxide (NO), Endocrine-Disrupting Chemicals (EDCs), Water pollution, Biomarkers, Aquatic environment, Environmental Monitoring

1. INTRODUCTION

Aquatic pollution is a growing environmental concern worldwide. The main causes of aquatic pollution include heavy metals, industrial compounds such as bisphenol A (BPA), pesticides like atrazine, and pharmaceuticals such as ethinylestradiol. Among these pollutants are endocrine-disrupting chemicals (EDCs), which pose significant risks to aquatic ecosystems and human health (Encarnação et al., 2019; Pironti et al., 2021). In aquatic environments, EDCs interfere with reproductive and developmental processes in wildlife, leading to ecosystem imbalances (Kadhim et al., 2024). In humans, they disrupt endocrine function, potentially affecting fertility, triggering early puberty, and contributing to metabolic disorders (Yilmaz et al., 2020). Additionally, EDC exposure has been linked to impaired brain development, behavioural and learning difficulties, altered immune function, and an increased risk of hormone-sensitive cancers (Raja et al., 2022; Yilmaz et al., 2020).

Several techniques have been implemented to detect and monitor EDCs in aquatic environments. Traditional methods, such as chromatography-mass spectrometry (GC-MS) or high-performance liquid chromatography (HPLC), provide high accuracy but require sophisticated laboratory infrastructure, making large-scale environmental monitoring impractical (Wu et al., 2014). Consequently, there is growing interest in using biomarkers, which are measurable biochemical or cellular changes in organisms exposed to pollutants. They are considered cost-effective and sensitive indicators of environmental contamination. Bioassays, known for their rapid and efficient detection capabilities, are commonly used to assess biomarker alterations. Among these, nitric oxide (NO), a small reactive signalling molecule, has gained attention as a potential biomarker due to its role in oxidative stress, immune response, and physiological

regulation in aquatic organisms exposed to environmental pollutants (Yu et al., 2020).

Nitric oxide is naturally produced by various cells and is critical to cellular signalling pathways, including those involved in stress responses. Exposure to EDCs can disrupt NO synthesis, leading to altered NO levels that serve as early indicators of environmental toxicity (Xu et al., 2013). Studies have shown that pollutants such as bisphenol A (BPA) induce oxidative stress, triggering changes in NO metabolism and nitric oxide synthase (NOS) activity (Palacios-Valladares et al., 2024). These physiological changes provide valuable insights into pollutant exposure, making NO a promising candidate for biomonitoring applications in water quality assessment.

2. BIOLOGICAL BASIS OF NITRIC OXIDE AS A POTENTIAL BIOMARKER OF ENDOCRINE-DISRUPTING POLLUTANTS

Nitric oxide (NO) is primarily synthesised by the enzyme nitric oxide synthase (NOS), which converts L-arginine into NO and citrulline (Dutta and Sengupta, 2022). It is naturally produced by various cells and is critical to cellular signalling pathways, including those involved in immune and stress responses (Figure 1). In aquatic organisms, NO is involved in critical functions such as immune defence, neurotransmission, and vascular regulation (Fago and Jensen, 2015). Similarly, NO mediates oxidative stress and detoxification responses when organisms encounter environmental stressors, including EDC pollutants. EDCs disrupt NO metabolism, causing physiological and biochemical changes that serve as biomarkers for water quality assessment (Makene and Pool, 2015). Monitoring NO fluctuations in exposed cells or organisms provides insights into the presence and impact of pollutants in aquatic ecosystems.

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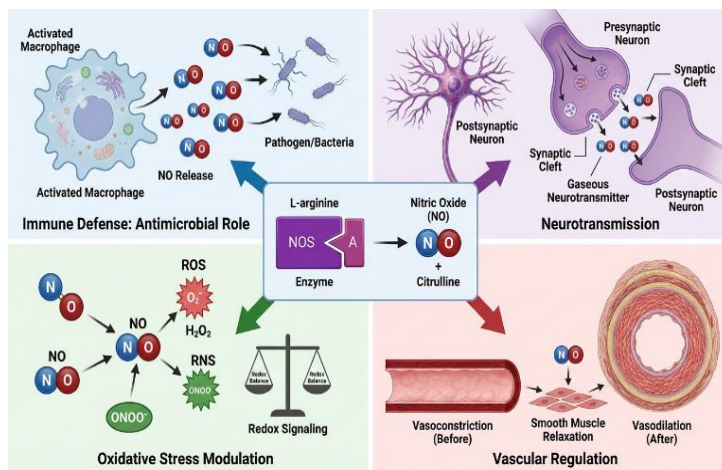


Figure 1: Nitric oxide (NO) synthesis and its biological roles

The synthesis and regulation of NO are highly sensitive to environmental stressors (Figure 2). EDCs such as bisphenol A (BPA), phthalates, and pesticides interfere with hormonal signalling pathways, indirectly affecting NO production. These chemicals modulate the expression of genes involved in oxidative stress regulation, leading to either excessive NO release, which contributes to cellular toxicity, or reduced NO production, impairing normal physiological functions (Guarnotta et al., 2022; Kim et al., 2022). The distinct NO response to various pollutants underscores its value as a biomarker for EDC contamination in aquatic environments.

Nitric oxide is also produced by immune cells, particularly macrophages,

to combat infections through antimicrobial and antiviral effects. It reacts with superoxide to form reactive nitrogen species that are responsible for pathogen destruction and the regulation of inflammation (Wink et al., 2011; Predonzani et al., 2015). However, exposure to EDCs such as BPA and phthalates can interfere with NO production and signalling (Hu et al., 2024; Palacios-Valladares et al., 2024). These pollutants alter NOS activity, leading to dysregulated immune responses, oxidative stress, and increased susceptibility to infections and inflammatory diseases (Popescu et al., 2021). While NO is essential for immune defense, environmental pollutants can disrupt its balance, contributing to immune dysfunction and broader health risks.

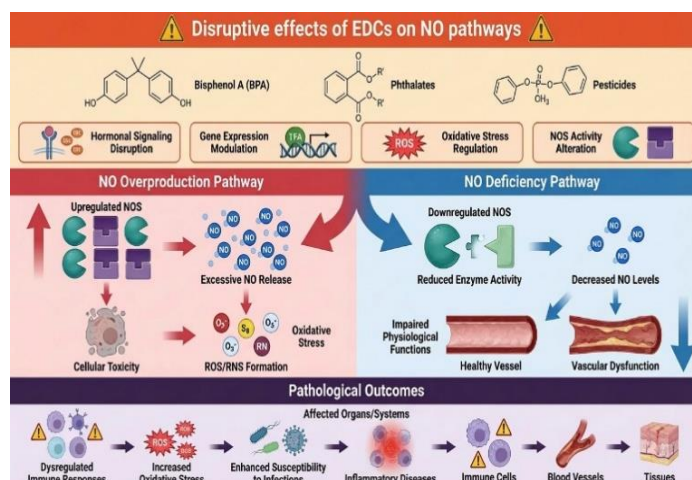


Figure 2: Disruptive effects of EDCs on NO pathways

3. POTENTIAL APPLICATION OF NO AS A BIOMARKER OF ENDOCRINE-DISRUPTING POLLUTANTS

The use of NO as a biomarker for EDCs in aquatic environments is supported by various analytical and bioassay techniques designed to measure NO levels. These methods leverage NO's highly reactive nature and its role in oxidative stress and physiological signalling pathways. Traditional EDCs detection techniques, such as GC-MS or HPLC, provide reliable quantification of EDCs. However, bioassays offer a more biologically relevant approach by assessing NO fluctuations in aquatic organisms exposed to EDCs. These assays utilise biological systems, including cell cultures (Makene and Pool, 2015; Makene and Pool, 2019) and whole aquatic organisms (Xu et al., 2013), to measure NO production and its physiological effects in response to pollutant exposure. Compared to conventional chemical analyses in water, bioassays provide insight into the real-time impact of EDCs on living organisms, enhancing environmental monitoring and risk assessment.

One of the most widely used NO bioassays is the Griess reaction, which measures NO breakdown products (nitrite and nitrate) in biological fluids. In this assay, NO reacts with oxygen and other cellular molecules to form nitrites, which are quantified using a colourimetric reaction with sulfanilamide and N-(1-naphthyl) ethylenediamine. This method is advantageous for large-scale environmental screening due to its simplicity, cost-effectiveness, and adaptability to different sample types.

However, its use is limited by its indirect measurement of NO and potential interference from other nitrogenous compounds.

Several studies have explored the relationship between NO production and exposure to EDCs (Table 1). For example, research on juvenile African catfish (*Clarias gariepinus*) demonstrated that exposure to bisphenol A (BPA) led to a concentration-dependent increase in NO synthesis (Samwel et al., 2022). The study measured NO levels and respiratory burst activity in fish exposed to different concentrations of BPA, bisphenol AP, and bisphenol P, revealing immune function impairment through NO alterations. Similarly, a study by Xu et al. (2013) investigated the effects of BPA and nonylphenol (NP) on oxidative stress and immune responses in zebrafish embryos. Their findings indicated a concentration-dependent increase in reactive oxygen species (ROS), upregulation of redox-sensitive transcription factors, and significant induction of NO levels, accompanied by increased nitric oxide synthase (NOS) activity and inducible NOS (iNOS) gene expression. These results suggest that NO production is a sensitive indicator of oxidative stress and immune response activation in aquatic organisms exposed to industrial EDCs.

Pesticides are also known to induce oxidative stress in aquatic organisms, leading to the production of reactive oxygen species (ROS) and reactive nitrogen species (RNS), including NO. This oxidative stress contributes to biochemical and histological changes, as well as neurological dysfunction in freshwater organisms (Saha and Dutta, 2024). Monitoring NO levels

could therefore serve as a biomarker for pesticide-induced toxicity in aquatic environments. Additionally, exposure to low doses of pesticides has been shown to induce an immune response and increase NO production in honeybees, suggesting a broader role for NO in immune responses to environmental pollutants (Bartling et al., 2021). While this study focuses on honeybees, it provides insights into how NO-based immune responses might apply to aquatic organisms.

Further evidence from *in vitro* studies supports the role of NO in EDC-related immune modulation. A study examining BPA's impact on carp phagocytic cells found that BPA altered respiratory burst activity, potentially affecting NO production and immune defence mechanisms (Gushiken et al., 2002). Similarly, a study investigating the effects of various EDCs on tumour necrosis factor- α (TNF- α) and NO production

in mouse macrophages revealed that certain EDCs modulate immune responses by altering NO levels (Hong et al., 2004). Another *in vitro* study assessed how EDCs, including estradiol (E2), 5 α -dihydrotestosterone (DHT), and BPA, influenced NO secretion, a key indicator of inflammation (Makene and Pool, 2019). The study demonstrated that exposure to these EDCs significantly decreased the secretion of NO and IL-6 in LPS-stimulated RAW264.7 cells, suggesting that EDCs can suppress inflammatory responses in immune cells. Collectively, these studies demonstrate that EDC exposure can significantly impact NO production in aquatic animals and *in vitro* models, resulting in oxidative stress and modulation of the immune system. The sensitivity of NO to EDC-induced physiological disruptions reinforces its potential as a biomarker for assessing environmental EDC contamination.

Table 1: Examples of applications of nitric oxide (NO) as a biomarker of EDCs

EDC Type	Model Organism / System	Exposure Context	Observed NO Response	Associated Biological Effects	Biomarker Application	Reference
Bisphenol A (BPA),	Juvenile African catfish (<i>Clarias gariepinus</i>)	<i>In vivo</i> exposure to increasing EDC concentrations	Concentration-dependent increase in NO synthesis	Impaired immune function; altered respiratory burst activity	NO as a sensitive biomarker of immune disruption and EDC exposure in freshwater fish	Samwel <i>et al.</i> , (2022)
Bisphenol A (BPA), Nonylphenol (NP)	Zebrafish embryos (<i>Danio rerio</i>)	<i>In vivo</i> embryonic exposure	Increased NO levels; elevated NOS activity; upregulated iNOS expression	Oxidative stress, immune activation, redox imbalance	NO as an indicator of oxidative stress and immune response activation by industrial EDCs	Xu <i>et al.</i> , (2013)
Pesticides	Freshwater aquatic organisms	Environmental pesticide exposure	Elevated NO and other reactive nitrogen species (RNS)	Biochemical, histological, and neurological dysfunction	NO as a biomarker of pesticide-induced oxidative and neurotoxic stress	Saha and Dutta, (2024)
Bisphenol A (BPA)	Carp phagocytic cells (<i>in vitro</i>)	Cellular exposure to BPA	Altered respiratory burst activity linked to NO modulation	Impaired immune defense mechanisms	NO as a cellular-level biomarker of immunotoxicity	Gushiken <i>et al.</i> , (2002)
Multiple EDCs	Mouse macrophages (<i>in vitro</i>)	Exposure to various endocrine disruptors	Altered NO production	Modulation of TNF- α -mediated immune responses	NO as a marker of EDC-mediated immune dysregulation	Hong <i>et al.</i> , (2004).
BPA, Estradiol (E2), 5 α -Dihydrotestosterone (DHT),	RAW264.7 macrophage cells (<i>in vitro</i>)	LPS-stimulated immune model	Decreased NO secretion	Suppressed inflammatory responses	NO as a biomarker of EDC-induced immunosuppression	Makene and Pool, (2019).

4. ADVANTAGES OF USING NO AS A BIOMARKER OF ENDOCRINE-DISRUPTING CHEMICALS

Using NO as a biomarker for assessing EDCs in aquatic environments offers several notable advantages, given its sensitivity to environmental stressors and its crucial role in physiological signalling pathways. One of the primary benefits is NO's early responsiveness to pollution-induced stress. Unlike some conventional water quality indicators, which require prolonged exposure to contaminants before measurable effects become apparent, NO levels can fluctuate rapidly in response to EDC exposure (Makene and Pool, 2019). This rapid response allows for early detection of endocrine disruption, offering a valuable early warning system for potential environmental and public health risks. For instance, studies have demonstrated that NO production in aquatic species such as fish can be implemented as an early biomarker for pollutants like bisphenol A (BPA) and phthalates, which often disrupt endocrine systems in aquatic organisms (Samuel *et al.* 2022).

The involvement of NO in oxidative stress, immune response, and hormonal regulation further supports its suitability for identifying biological disruptions caused by EDCs. As such, NO serves as a critical marker for a range of EDCs, including synthetic hormones, which are known to affect hormonal balance in aquatic organisms. The ability to measure NO production across various stages of the endocrine disruption process enhances its value as an indicator of chemical exposure and its effects on aquatic systems. Another significant advantage of NO as a

biomarker is its widespread presence in aquatic organisms, making it applicable across multiple bioindicator species, including fish (dos Santos Carvalho *et al.* 2012), amphibians (Chen *et al.*, 2023) and invertebrates (Ray *et al.*, 2015). NO is naturally produced as part of cellular metabolism and stress response mechanisms, making it a universal biomarker for different species.

By monitoring changes in NO concentration, researchers can assess the ecological impact of EDCs in diverse aquatic environments, ensuring the broad applicability of this biomarker in ecotoxicological studies. This universality improves our understanding of how EDCs affect entire ecosystems, as shifts in NO levels reflect the collective biological responses of multiple species to environmental stressors. Moreover, the involvement of NO in several interconnected biological pathways enables its integration with other biomarkers, such as oxidative stress markers, hormone level alterations, and reproductive toxicity indicators. This multi-pathway integration provides a more comprehensive understanding of the biological effects of EDC exposure, improving the accuracy of environmental risk assessments. For example, fluctuations in NO levels can be used alongside other biomarkers, such as hormonal levels, to assess the broader physiological impacts of EDCs on aquatic species, ultimately enhancing the quality of data used for decision-making.

Technological advancements in NO detection methods further contribute to its advantages as a biomarker for EDC monitoring. The development of electrochemical sensors (Brown and Schoenfish, 2019; Zhang, 2008) and fluorescence-based probes (McQuade and Lippard, 2010; Zhou *et al.*,

2023) has enabled real-time, highly sensitive, and cost-effective NO quantification. These modern methods are not only more accessible but also provide greater sensitivity compared to traditional analytical techniques, such as GC-MS or HPLC, which require expensive equipment and extensive sample preparation. For instance, NO biosensors allow rapid field-based detection without complex laboratory setups. This is especially beneficial in areas with limited laboratory infrastructure, where rapid environmental monitoring is crucial for managing pollution risks.

Furthermore, integrating NO detection with portable biosensor technologies offers miniaturised, automated systems capable of on-site, continuous pollution tracking. This reduces the reliance on labour-intensive and time-consuming chemical analyses, thus making environmental monitoring more efficient and practical. In addition to the practical advantages, the use of NO-based detection in water quality monitoring programs can significantly improve the monitoring and management of aquatic systems impacted by EDCs. By enabling real-time, continuous data collection, scientists and policymakers can more quickly monitor pollution events, track trends in water quality, and implement timely interventions. Furthermore, as these technologies become more widespread and accessible, they can contribute to global efforts in pollution regulation and aquatic ecosystem protection.

Overall, NO as a biomarker for monitoring EDCs in water offers significant advantages due to sensitivity, ecological relevance, and technological feasibility. Its ability to detect early physiological changes, its widespread applicability across aquatic organisms, and its compatibility with modern bio-sensing techniques make it a valuable tool for environmental monitoring. By incorporating NO-based detection strategies into water quality assessment programs, scientists and policymakers can enhance pollution surveillance and support evidence-based water quality regulations to safeguard aquatic ecosystems and human health from the harmful effects of endocrine-disrupting pollutants.

5. CHALLENGES, RESEARCH GAPS AND FUTURE DIRECTIVES

Despite the promising potential of NO as a biomarker for monitoring EDCs in aquatic environments, several challenges may hinder its widespread application in environmental monitoring. One of the most significant challenges is the instability and reactivity of NO, which makes its direct quantification difficult. NO is a short-lived molecule that readily interacts with oxygen and reactive oxygen species (ROS), forming secondary nitrogenous compounds such as nitrite, nitrate, and peroxynitrite (Pacher et al., 2007; Procházková et al., 2015). This rapid conversion complicates accurate measurement, often requiring indirect detection methods that may not provide precise real-time assessments. To overcome this challenge, integrating NO-based detection with multi-biomarker approaches, such as combining NO analysis with oxidative stress markers, gene expression studies, or hormone level assessments, can help differentiate EDC-related NO alterations from other environmental stressors.

Another challenge is the influence of external environmental factors, such as temperature, pH fluctuations, and water chemistry, on NO production and metabolism in bioindicator species (Giordano et al., 2022). These variations can lead to inconsistent results, complicating the establishment of standardised NO threshold levels for EDC monitoring across different aquatic ecosystems. To address this, establishing reference NO levels in various bioindicator species, correlating them with known EDC exposure data, and developing standardised NO measurement protocols across different aquatic environments are important. For instance, setting baseline NO concentrations in species like fish or amphibians under controlled conditions can provide valuable data for setting regulatory thresholds.

Another significant limitation is the lack of specificity in NO responses to EDC exposure. NO plays a role in various physiological processes, including vascular function, neurotransmission, and immune response. As a result, changes in NO levels may not be exclusively linked to EDC exposure but could also arise from heavy metal contamination, pathogen infections, or hypoxia (Donaghy et al., 2015). To improve specificity, future research should focus on differentiating NO fluctuations due to EDCs from those caused by other pollutants. This may involve identifying unique markers or patterns of NO disruption specific to endocrine disruption.

Furthermore, many EDCs exert their effects through complex hormonal signalling pathways, and the exact mechanisms they disrupt NO production remain incompletely understood. Expanding mechanistic research is crucial for refining the role of NO as a biomarker. In particular, research should focus on understanding the signalling pathways through which EDCs interfere with NO production, and how these mechanisms vary across different classes of chemicals. Advanced techniques such as proteomics and transcriptomics can help elucidate the molecular

mechanisms through which EDCs affect NO production. For example, transcriptomic analysis could identify genes involved in NO synthesis that are upregulated or downregulated in response to EDC exposure.

Through these combined efforts, a deeper understanding of the role of NO in EDC toxicity will emerge, enabling more accurate and efficient environmental monitoring strategies. Overall, refining the application of NO as a biomarker for monitoring EDC in aquatic environments will require addressing these challenges through both technological advancements and biological research. By improving NO measurement techniques, standardising protocols, and gaining deeper insights into the molecular pathways linking NO to EDC toxicity, researchers can enhance the effectiveness of NO as a tool for environmental pollution monitoring.

6. CONCLUSIONS

Nitric oxide (NO) has emerged as a promising biomarker for monitoring the effects of endocrine-disrupting chemicals (EDCs) in aquatic environments. Given its critical role in oxidative stress regulation, endocrine signalling, and immune responses, NO serves as an effective early indicator of pollutant exposure. NO-based detection methods, including electrochemical sensors and bioassays, offer several advantages over traditional chemical analyses, such as real-time monitoring, cost-effectiveness, and non-invasive application. However, to optimise the use of NO in environmental monitoring, challenges related to specificity, standardisation, and technological limitations must be addressed. For example, improving the selectivity of NO sensors to distinguish them from other reactive nitrogen species in complex aquatic matrices is crucial.

Additionally, establishing standardised protocols for NO detection across various aquatic environments will ensure the reliability and comparability of results. In general, NO-based biomonitoring has the potential to significantly enhance aquatic environmental assessment, thereby supporting the protection of aquatic ecosystems. Future efforts should focus on developing more sensitive, accurate, and widely applicable biosensors. Furthermore, integrating NO biomarkers into regulatory frameworks will be essential for the widespread adoption of these tools. Collaborative efforts between researchers, environmental agencies, and policymakers will be crucial in accelerating the adoption of NO-based monitoring systems and ensuring their implementation for monitoring EDCs in aquatic environments.

REFERENCES

- Bartling, M. T., Thümecke, S., Russert, J. H., Vilcinskis, A. and Lee, K. Z., 2021. Exposure to low doses of pesticides induces an immune response and the production of nitric oxide in honeybees. *Scientific reports*, 11(1), 6819. <https://doi.org/10.1038/s41598-021-86293-0>.
- Brown, M. D. and Schoenfish, M. H., 2019. Electrochemical nitric oxide sensors: principles of design and characterization. *Chemical Reviews*, 119(22), 11551-11575.
- Chen, H., Pang, Y., Wei, Y., He, X., Zhang, Y., Xie, L., 2023. Nitrate and sodium nitroprusside alter the development of Asian black-spined toads' embryos by inducing nitric oxide production. *Environ Sci Pollut Res*. 30, 23060-23069. <https://doi.org/10.1007/s11356-022-23821-z>
- Donaghy, L., Hong, H. K., Jauzein, C. and Choi, K. S., 2015. The known and unknown sources of reactive oxygen and nitrogen species in haemocytes of marine bivalve molluscs. *Fish & shellfish immunology*, 42(1), 91-97. <https://doi.org/10.1016/j.fsi.2014.10.030>
- dos Santos Carvalho, C., Bernusso, V. A., de Araújo, H. S. S., Espíndola, E. L. G. and Fernandes, M. N., 2012. Biomarker responses as an indication of contaminant effects in *Oreochromis niloticus*. *Chemosphere*, 89(1), 60-69. <https://doi.org/10.1016/j.chemosphere.2012.04.013>
- Dutta, S. and Sengupta, P., 2022. The role of nitric oxide on male and female reproduction. *The Malaysian journal of medical sciences: MJMS*, 29(2), 18. <https://doi.org/10.21315/mjms2022.29.2.3>
- Encarnaç o, T., Pais, A. A. and Campos, M. G. and Burrows, H. D., 2019. Endocrine disrupting chemicals: Impact on human health, wildlife and the environment. *Science Progress*, 102(1), 3-42. <https://doi.org/10.1177/0036850419826802>
- Fago, A. and Jensen, F. B., 2015. Hypoxia tolerance, nitric oxide, and nitrite: lessons from extreme animals. *Physiology*, 30(2), 116-126. <https://doi.org/10.1152/physiol.00051.2014>
- Giordano, D., Verde, C. and Corti, P., 2022. Nitric oxide production and regulation in the teleost cardiovascular system. *Antioxidants*, 11(5), 957. <https://doi.org/10.3390/antiox11050957>

- Guarnotta, V., Amodei, R., Frasca, F., Aversa, A. and Giordano, C., 2022. Impact of chemical endocrine disruptors and hormone modulators on the endocrine system. *International journal of molecular sciences*, 23(10), 5710. <https://doi.org/10.3390/ijms23105710>
- Gushiken, Y., Watanuki, H. and Sakai, M., 2002. In vitro effect of carp phagocytic cells by bisphenol A and nonylphenol. *Fisheries science*, 68(1), 178-183.
- Gusti, A. M., Qusti, S. Y., Alshammari, E. M., Toraih, E. A. and Fawzy, M. S., 2021. Antioxidants-related superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GPX), glutathione-S-transferase (GST), and nitric oxide synthase (NOS) gene variants analysis in an obese population: a preliminary case-control study. *Antioxidants*, 10(4), 595. <https://doi.org/10.3390/antiox10040595>
- Hong, C. C., Shimomura-Shimizu, M., Muroi, M. and Tanamoto, K. I., 2004. Effect of endocrine disrupting chemicals on lipopolysaccharide-induced tumor necrosis factor- α and nitric oxide production by mouse macrophages. *Biological and Pharmaceutical Bulletin*, 27(7), 1136-1139.
- Hu, C., Lu, L., Guo, C., Zhan, T., Zhang, X. and Zhang, H., 2024. Bisphenols and brominated bisphenols induced endothelial dysfunction via its disruption of endothelial nitric oxide synthase. *Environmental Pollution*, 346, 123600. <https://doi.org/10.1016/j.envpol.2024.123600>
- Kadhim, S.J., Sahib, M.A., Khlaif, S.M., AL-Azzawi, M.K. and Zubaidi, N.A., 2024. Evaluating the Impact of Endocrine-Disrupting-Chemicals on Fertility in Wildlife: From Amphibians to Mammals. *JMGCB*, 1, pp.216-228. <https://doi.org/10.61796/jmgcb.v1i8.841>
- Kim, K., Kwon, J. S., Ahn, C. and Jeung, E. B., 2022. Endocrine-disrupting chemicals and their adverse effects on the endoplasmic reticulum. *International Journal of Molecular Sciences*, 23(3), 1581. <https://doi.org/10.3390/ijms23031581>
- Makene, V. W. and Pool, E. J., 2015. The assessment of inflammatory activity and toxicity of treated sewage using RAW 264.7 cells. *Water and Environment Journal*, 29(3), 353-359. <https://doi.org/10.1111/wej.12127>
- Makene, V. W. and Pool, E. J., 2019. The effects of endocrine-disrupting chemicals on biomarkers of inflammation produced by lipopolysaccharide-stimulated RAW264. 7 macrophages. *International Journal of Environmental Research and Public Health*, 16(16), 2914. <https://doi.org/10.3390/ijerph16162914>
- McQuade, L. E. and Lippard, S. J., 2010. Fluorescent probes to investigate nitric oxide and other reactive nitrogen species in biology (truncated form: fluorescent probes of reactive nitrogen species). *Current opinion in chemical biology*, 14(1), 43-49. <https://doi.org/10.1016/j.cbpa.2009.10.004>
- Pacher, P., Beckman, J. S. and Liaudet, L., 2007. Nitric oxide and peroxynitrite in health and disease. *Physiological reviews*, 87(1), 315-424. <https://doi.org/10.1152/physrev.00029.2006>
- Palacios-Valladares, J. R., Martinez-Jimenez, Y. I., Morillon-Torres, V., Rivera-Maya, O. B., Gómez, R. and Calderon-Aranda, E. S., 2024. Bisphenol A and Its Emergent Substitutes: State of the Art of the Impact of These Plasticizers on Oxidative Stress and Its Role in Vascular Dysfunction. *Antioxidants*, 13(12), 1468. <https://doi.org/10.3390/antiox13121468>
- Pironti, C., Ricciardi, M., Proto, A., Bianco, P. M., Montano, L. and Motta, O., 2021. Endocrine-disrupting compounds: An overview on their occurrence in the aquatic environment and human exposure. *Water*, 13(10), 1347. <https://doi.org/10.3390/w13101347>
- Popescu, M., Feldman, T. B. and Chitnis, T., 2021. Interplay between endocrine disruptors and immunity: Implications for diseases of autoreactive etiology. *Frontiers in Pharmacology*, 12, 626107. doi: 10.3389/fphar.2021.626107
- Predonzani, A., Cali, B., Agnellini, A. H. and Molon, B., 2015. Spotlights on immunological effects of reactive nitrogen species: when inflammation says nitric oxide. *World journal of experimental medicine*, 5(2), 64. DOI: <http://dx.doi.org/10.5493/wjem.v5.i2.64>
- Procházková, D., Wilhelmová, N. and Pavlík, M., 2015. Reactive nitrogen species and nitric oxide. *Nitric Oxide Action in Abiotic Stress Responses in Plants*, 3-19.
- Raja, G. L., Subhashree, K. D. and Kantayya, K. E., 2022. In utero exposure to endocrine disruptors and developmental neurotoxicity: Implications for behavioural and neurological disorders in adult life. *Environmental Research*, 203, 111829. <https://doi.org/10.1016/j.envres.2021.111829>
- Ray, S., Mukherjee, S., Bhunia, N. S., Bhunia, A. S. and Ray, M., 2015. Immunotoxicological threats of pollutants in aquatic invertebrates. In *Emerging pollutants in the environment-current and further implications*. IntechOpen. DOI: 10.5772/60216
- Saha, R. and Dutta, S. M., 2024. Pesticides' mode of action on aquatic life. *Toxicology Reports*, 101780. <https://doi.org/10.1016/j.toxrep.2024.101780>
- Samuel, O. D., Adeyemi, J. A., Bamidele, O. S., Barbosa Jr, F. and Adedire, C. O., 2022. Cytotoxicity, redox and immune status in African catfish, *Clarias gariepinus* (Burchell, 1822) exposed to bisphenol A (BPA) and its analogues. *Environmental Science and Pollution Research*, 29(49), 74185-74196. <https://orcid.org/0000-0003-2250-5862>
- Singh, P., Singh, M. K., Beg, Y. R. and Nishad, G. R., 2019. A review on spectroscopic methods for determination of nitrite and nitrate in environmental samples. *Talanta*, 191, 364-381. <https://doi.org/10.1016/j.talanta.2018.08.028>
- Wink, D. A., Hines, H. B., Cheng, R. Y., Switzer, C. H., Flores-Santana, W., Vitek, M. P., Ridnour, L. A. and Colton, C. A., 2011. Nitric oxide and redox mechanisms in the immune response. *Journal of leukocyte biology*, 89(6), 873-891. <https://doi.org/10.1189/jlb.1010550>
- Wu, A., Duan, T., Tang, D., Zheng, Z., Zhu, J., Wang, R., He, B., Cheng, H., Feng, L. and Zhu, Q., 2014. Review the application of chromatography in the analysis of nitric oxide-derived nitrite and nitrate ions in biological fluids. *Current Analytical Chemistry*, 10(4), 609-621.
- Xu, H., Yang, M., Qiu, W., Pan, C., and Wu, M., 2013. The impact of endocrine-disrupting chemicals on oxidative stress and innate immune response in zebrafish embryos. *Environmental Toxicology and Chemistry*, 32(8), 1793-1799. DOI: 10.1002/etc.2245
- Yang, M., Qiu, W., Chen, B., Chen, J., Liu, S., Wu, M. and Wang, K. J., 2015. The in vitro immune modulatory effect of bisphenol A on fish macrophages via estrogen receptor α and nuclear factor- κ B signaling. *Environmental science & technology*, 49(3), 1888-1895. <http://dx.doi.org/10.1021/es505163v>
- Yilmaz, B., Terekeci, H., Sandal, S. and Kelestimur, F., 2020. Endocrine-disrupting chemicals: exposure, effects on human health, mechanism of action, models for testing and strategies for prevention. *Reviews in endocrine and metabolic disorders*, 21, 127-147. <https://doi.org/10.1007/s11154-019-09521-z>
- Yu, Z., Quan, Y. N., Huang, Z. Q., Wang, H. H. and Wu, L. F. (2020). Monitoring oxidative stress, immune response, Nrf2/NF- κ B signaling molecules of *Rhynchocypris lagowski* living in BFT system and exposed to waterborne ammonia. *Ecotoxicology and Environmental Safety*, 205, 111161. <https://doi.org/10.1016/j.ecoenv.2020.111161>
- Zhang, X., 2008. Nitric oxide (NO) electrochemical sensors. In *Electrochemical sensors, biosensors and their biomedical applications* (pp. 1-29). DOI: 10.1016/B978-012373738-0.50003-9
- Zhou, Y., Yang, X., Xu, S., Li, J., Wang, W. and Yan, M., 2023. Small molecule fluorescent probes for the detection of reactive nitrogen species in biological systems. *Coordination Chemistry Reviews*, 493, 215258. <https://doi.org/10.1016/j.ccr.2023.215258>

