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THE ROLE OF OXIDATIVE STRESS BIOMARKERS IN ANIMAL HEALTH, TOXICOLOGY, AND ENVIRONMENTAL MONITORING

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ABSTRACT

The oxidative stress biomarkers have gained significance in veterinary medicine, animal production, toxicology and environmental biomonitoring because of their link between redox imbalance and clinically as well as ecologically relevant results. RONS are produced during metabolism, immune defence, reproduction, growth and adaptation but excess production or a lack of antioxidants can lead to damage of lipids, proteins, DNA, mitochondria and cellular membranes. This review includes an integrated biomarker framework, practical interpretation criteria and original explanatory diagrams which detail the use of oxidative stress markers within animal health, toxicological evaluation and environmental monitoring, incorporating a One Health perspective. The biomarkers reviewed are those involved in lipid peroxidation (malondialdehyde), antioxidants (superoxide dismutase, catalase, glutathione peroxidase, and glutathione S-transferase), and molecular damage markers (protein carbonyls and 8-hydroxy-2'-deoxyguanosine). The oxidative stress biomarkers reviewed in this report are not diagnostic biomarkers of their own, but rather are indicators of early warning and should be combined with pathology, performance indicators, contaminant data and longitudinal surveillance.

Keywords: Animal Health; Antioxidant Enzymes; Environmental Monitoring; Oxidative Stress; Sentinel Species; Toxicology

1 INTRODUCTION

Oxidative stress is generally accepted to be an imbalance between the production of pro-oxidant molecules and the antioxidant defenses that results in a change in redox signaling and molecular damage. In addition to being toxic by-products, both ROS and RNS play roles in immune defence, vascular regulation, mitochondrial adaptation, reproduction and cellular signalling in animals. When the production of oxidants exceeds the capacity of enzymatic and non-enzymatic protection, a progressive damage to lipids, proteins, nucleic acids and membranes starts to occur, which can lead to disease and reduction in the ability to perform (Jomova et al., 2023).

Oxidative stress is becoming a biological link in veterinary and animal-production research between metabolism, inflammation, nutrition and immunity, toxic exposure and infection, ageing and welfare. Significant redox imbalance has been reported in farm animals under heat stress, high production demand, reproductive pressure, transport and dietary imbalance, and in companion animals with chronic renal, endocrine, cardiovascular, neoplastic, gastrointestinal and inflammatory diseases (Jain & Shakkarpude, 2024). The main scientific importance of oxidative stress biomarkers is their use to identify subclinical biological disturbances before gross lesions on the tissues, mortality or significant loss of productivity is observed. Traditional veterinary assessment relies heavily on clinical signs, haematology, serum biochemistry, histopathology and production indices, whereas oxidative biomarkers provide an earlier layer of information on whether cells are experiencing damage, or are responding to cell protection through antioxidant mechanisms (De La Riva De La Riva et al., 2022).

2 SOURCE OF THE BIOLOGICAL BASIS AND TYPES OF BIOMARKERS.

The oxidative stress biomarkers can be classified into three broad categories: Oxidant burden markers, Antioxidant defense markers, and Molecular damage markers. Oxidants measurement markers are total oxidant status and tests for reactive oxygen metabolites while antioxidants-defense markers test include superoxide dismutase, catalase, glutathione peroxidase, glutathione reductase, glutathione S-transferase, reduced glutathione, vitamin related antioxidants capacity, total antioxidants status, and total antioxidants capacity (Cordiano et al., 2023). One of the most commonly used marker of lipid peroxidation is malondialdehyde, which can be analyzed in serum, plasma, tissue homogenates, milk, urine and other biological fluids. It is very useful but the interpretation needs to be made with caution, since lipid oxidation could be overestimated when there is a potential analytical interference, hemolysis, lipid content or inadequate sample handling (Perez-Montero et al., 2025). Antioxidants do not offer a direct picture of molecular injury, but rather of cellular defence. Superoxide dismutase: superoxide radicals to hydrogen peroxide, catalase: hydrogen peroxide to water and oxygen, and glutathione peroxidase: peroxides, glutathione. Such enzymes can increase during adaptive protection, but may decrease when antioxidant stores are depleted or when damage to tissues is quite extensive (Formicki et al., 2025). A molecular dimension is given to the interpretation of oxidative stress due to the presence of markers related to DNA oxidation and to telomeres. 8-hydroxy-2'-deoxyguanosine is a biomarker of oxidative DNA damage, and is important for aging, chronic disease, reproduction, toxicology and conservation biology. The correlation of 8-OHdG, telomere length and antioxidant capacity in psittacine birds helps to understand the need of considering redox biomarkers in relation to lifespan strategy and species biology (Dominguez-de-Barros et al., 2024).

3 OXIDATIVE STRESS BIOMARKERS IN FARM ANIMAL HEALTH

Oxidative stress has a strong link with the transition period and early lactation in dairy cattle, with negative energy balance, immune suppression and metabolic disease. Periparturient period is characterized by increased production of oxygen, mobilisation of lipids, hormonal modification and inflammation, resulting in increased generation of reactive species and reduced antioxidant levels (Abuelo et al., 2019). High milk production during early lactation will accentuate oxygen demand and metabolic load. Increased malondialdehyde, altered glutathione peroxidase, modified catalase activity, and changes in total antioxidant capacity may be useful redox biomarkers to detect animals that are under metabolic strain before severe clinical disease is observed (Tufarelli et al., 2023). There is also an association between oxidative imbalance and transition cow disorders. It is hypothesized that oxidative stress biomarkers and metabolic parameters may vary in parallel in cows with abomasal displacement, implying that the use of redox parameters can be used in addition to biochemical and clinical data to assess the severity of the disease and the metabolic stress during the transition period (Mirzaei et al., 2025). Oxidative imbalance is a major cause of heat stress in ruminants. The biological cost of thermal exposure can be assessed through oxidative biomarkers, to help select heat resistant animals, as high ambient temperature and humidity lead to decreased feed intake, altered respiration and endocrine regulation, increased inflammatory burden and impaired immune function. (Abdelnour et al., 2019).

In poultry, oxidative stress has impacts on growth performance, feed efficiency, integrity of the intestine, immune responses, reproductive performance, quality of meat and disease resistance. In modern production, birds are exposed to rapid growth, high stocking density, heat stress, vaccination stress, mycotoxins, transport and infectious challenges which make the use of biomarker panels important markers for their nutritional and welfare assessment (Oke et al., 2024). Due to the interactions between mitochondrial ROS and the regulation of cytoprotective genes during heat stress, the antioxidant response related to Nrf2 is of special significance in heat-stressed chickens. This helps with interventions with vitamins, selenium, phytogenic compounds, probiotics, prebiotics and antioxidants from plant-based sources, which are based on practical results and should be evaluated based on biomarkers, not on general assumptions of benefit (Aryal et al., 2025). Environmental control, water management, stocking-density adjustment, feeding strategies, early conditioning and genetic selection are all important aspects of poultry heat-stress mitigation. When performance measures don't reveal early onset of cell damage, oxidative stress biomarkers can be employed as mechanistic endpoints to compare these interventions (Mangan & Siwek, 2024).

The importance of dietary antioxidants is related to their effect on immune competence, oxidative stability of meat and milk, reproductive performance and product quality, and hence is related to all farm species. The antioxidant support by vitamin E, selenium, carotenoids, polyphenols, flavonoids and other bioactive compounds should be targeted, but may be considered as supplements that complement a basal diet, production phase, physiological stage, and exposure conditions (Ponnampalam et al., 2022).. This is the reason why nutrition programs that are integrated are more scientifically valid than antioxidant supplementation isolated programs (Ayemele et al., 2021).

4 COMPANION ANIMAL MEDICINE

Oxidative stress biomarkers are used more and more in companion animal medicine for a better understanding of the mechanisms of chronic diseases and monitoring clinical burden. In dogs, oxidative stress has been reported in renal, cardiovascular, endocrine, gastrointestinal, neurologic, oncologic, infectious, parasitic, and inflammatory diseases, all of which have mitochondrial dysfunction, inflammation, immune activation, and tissue remodeling (Perez-Montero et al., 2024a). One of the greatest drawbacks to clinical applications is the lack of validated reference intervals for numerous species and assays. In recent years, reference intervals for serum TAS have been determined in dogs, and factors that may alter results have been tested, both biological and analytical, so it is a first step towards implementing TAS in clinical practice (Perez-Montero et al., 2024b).

An example of this is chronic kidney disease in dogs and cats. Oxidative stress is elevated with uremic toxins, inflammation, anemia, mitochondrial dysfunction, and changed antioxidant status, but must be correlated with the results of biomarkers, creatinine, symmetric dimethylarginine, urinalysis, blood pressure, proteinuria, hydration status, and clinical signs (Chen & Segev, 2024). Surgical operations can cause oxidative stress by damaging tissues, under anesthesia, inflammation, and ischemia reperfusion. Studies have been conducted comparing the effects of celiotomy and laparoscopy in dogs, which indicate that oxidative stress markers can be used alongside pain scores, vital parameters, and assessment of clinical recovery as a tool to measure perioperative stress (Naghavi et al., 2025).

5 TOXICOLOGICAL APPLICATIONS

Oxidative stress can be triggered by toxicants by mechanisms of redox cycling, inhibition of mitochondria, activation of inflammatory pathways, disruption of membranes, depleting glutathione, and disrupting antioxidant enzymes. Oxidative stress biomarkers are useful for toxicological pesticide monitoring and early detection of toxic effects of heavy metals, pharmaceuticals, mycotoxins, nanoparticles and plastics, as these effects can occur before the appearance of visible lesions (Anetor et al., 2022). Mycotoxins are a significant toxicological issue in animal production because of their ability to produce lipid peroxidation, liver toxicity, immune suppression, mitochondrial damage, and depletion of antioxidants, such as aflatoxins, fumonisins, ochratoxin A, deoxynivalenol, and zearalenone. Biomarkers are thus used to assess toxin exposure and the protective effects of the binders, probiotics, vitamins, selenium and plant-based interventions (Mavrommatis et al., 2021).

Pharmaceutical pollutants are growing in importance in aquatic systems. Biomarker panels are more informative than chemical detection alone, as they can provide information on antioxidant enzyme, lipid peroxidation, inflammatory, neurotoxic, endocrine disruptive, and histopathological changes in fish exposed to environmentally realistic concentrations (Gradinariu et al., 2025). Non-steroidal anti-inflammatory drugs may influence oxidative stress biomarkers in fish as revealed by a meta-analysis with some exposures showing high sensitivity for glutathione peroxidase. This is consistent with the inclusion of panels such as lipid peroxidation, glutathione peroxidase, catalase, superoxide dismutase, glutathione S-transferase and histopathology (Justi et al., 2025). Heavy metals are well-known inducers of oxidative stress in water organisms. Redox-active metals can directly produce ROS, whereas non-redox metals can inactivate antioxidant enzymes, sequester other metals, and chelate sulfhydryl groups, all of which can cause changes in antioxidant enzymes, metallothionein, lipid peroxidation, and DNA damage in tissue (Zeng et al., 2025).

Microplastics and nanoplastics are emerging stressors as they can lead to physical irritation, changes in gut microbiota, can be carrying adsorbed pollutants, disrupt immunity, and cause oxidative stress in aquatic animals. The effects of antioxidant, immune and histopathological endpoints on assessing biological burden of polypropylene microplastics has been demonstrated in African catfish, L. anales (Eid et al., 2025). Pharmaceutical pollutants produce antioxidant and histopathological responses in environmentally relevant sentinel species (common carp) in experimental evidence. Regarding the use of oxidative biomarkers in combination with tissue pathology and exposure information for ecological risk assessment, it is found that oxidative biomarkers will provide a better basis for ecological risk assessment than contaminant concentration alone (Shaalan et al., 2025).

6 ENVIRONMENTAL MONITORING AND SENTINEL SPECIES

Animals are used as sentinels in environmental monitoring due to their ability to integrate exposure across time and space. Chemical analysis determines concentrations of contaminants in water, sediment, soil or tissue, while biomarkers provide information on bioeffects at a cellular and physiological level (Cunningham et al., 2022). Fish are valuable sentinel species in aquatic monitoring because they come into contact with contaminated water via gills, skin, intestine, and food; and their liver, gill, kidney, brain, blood, and muscle tissues reflect exposure and damage to tissues. Traditionally, liver has been a preferred tissue for detox studies, while gills show the effects of water-borne exposure and muscle is of interest for food safety (Formicki et al., 2025). Wild birds are important monitors of environmental conditions: they are sensitive to pollution, disease, migration, reproductive efforts, nutritional stresses and climate stress. Reproduction, survival, immune competence and longevity may all be altered by oxidative stress, so the measurement of malondialdehyde, protein carbonyls, 8-OHdG, total antioxidant capacity, carotenoids, and antioxidant enzyme activities may be useful for conservation risk assessment (Faraguna et al., 2025).

7 ANALYTICAL AND INTERPRETIVE CHALLENGES

The use of biomarkers is a major challenge due to their lack of specificity. Raised levels of malondialdehyde or changes in catalase activity can be caused by infection, heat stress, exposure to toxic chemicals, poor nutrition status, transport, reproductive stress, aging, or mishandling samples. Thus, the use of oxidative stress markers should be used with caution, and must be backed up with clinical, experimental, or environmental data to support one cause (Mangan & Siwek, 2024). Another important point is the type of sample analysed as plasma, serum, erythrocytes, urine, milk, saliva, liver, gill, kidney, brain, muscle, and reproductive tissues can have varying oxidation profiles. The response in one tissue type may not match the response in another, such as a marker that is sensitive in liver but not in serum and/or a gill response that does not necessarily correlate with an edible muscle response, and, therefore, tissue selection needs to be aligned with the exposure route, target organ, and research question (Perez-Montero et al., 2025).

Storage temperature, hemolysis, anticoagulants, freeze-thaw cycles, sampling time, and lipid content are factors that can affect measured values, and may cause different results depending on the type of assay (spectrophotometric assays, ELISA kits, chromatographic methods, and mass spectrometry). Therefore, it is crucial to have standardized sampling and validated assays for comparison between studies (Perez-Montero et al., 2024b). The higher the antioxidant activity, the better; the lower the activity, the worse, but only in the context of exposure timing, duration, intensity, physiological status, tissue injury and outcome endpoints (De La Riva De La Riva et al., 2022).

8 SCIENTIFIC VALUE AND FUTURE RESEARCH FRAMEWORK

The scientific relevance of oxidative stress biomarkers is enhanced by being combined in multi-layered panels. A good panel should consist of at least one marker of antioxidant defence, one marker of molecular damage, one marker of tissue damage or a clinical endpoint and one contextual variable, such as exposure concentration, diet, age at sampling, sampling time or production stage. (Jain & Shakkarpude, 2024). Studies should focus on longitudinal monitoring instead of cross-sectional measurement in the future. Transient adaptation can be distinguished from persistent injury by repeated sampling and the evidence of recovery following intervention can be obtained. The ability to see if biomarker change predicts disease, reduced productivity, reproductive failure, or ecosystem decline is provided by repeated sampling. This is especially significant in the context of One Health where animal health, food safety, environmental contamination, and human exposure are linked (Anetor et al., 2022). The next step in methodology is the integration of oxidative stress markers with omics technologies such as metabolomics, proteomics, transcriptomics, and microbiome technologies. These may help to detect changes in the pathways, confirm the epigenetic regulation of traditional biomarkers, and determine if the redox disruption is a primary effect or a secondary effect of inflammation, toxic exposure, or tissue damage (Jomova et al., 2023).

9 CONCLUSION

In general, oxidative stress biomarkers are useful instruments to gain insight in the biological effects of disease and production stress, exposure to toxins and environmental contamination in animals. They play a key role in identifying subclinical redox imbalance and molecular damage in the absence of severe pathology, productivity loss, or population decline. In animal health, these biomarkers are useful for the evaluation of metabolic disease, heat stress, inflammation, renal disease, surgery, age-related disease, nutritional intervention and reproduction. In toxicology, they show the initial impact of drugs, heavy metals, mycotoxins, pesticides and micro-plastics. Sentinel species like fish, birds, bees, turtles and mammals can be used as living indicators of ecosystem stress in environmental monitoring. The future of this field is in the form of integrated biomarker panels, longitudinal monitoring, omics based validation, and One Health surveillance systems with the link to sustainability of the environment.

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