

Unravelling the Role of Oxidative Stress and Reactive Oxygen Species in Biological Systems: A Review

Divya Jain ¹, Chaithanya R ², Uddappanda Bopaiah Roy ³, Sanghamitra Satapathy ⁴,
Malathi H ⁵, Shivendra Kumar ⁶, Sharangouda J Patil ^{7,*}

¹ Department of Microbiology, School of Applied and Life Sciences, Uttaranchal University, Dehradun-248007, Uttarakhand, India; divyajain@uumail.in

² School of Mathematics and Natural Sciences, Chanakya University, Bengaluru-562165, Karnataka, India; chaithanya.r@chanakyauniversity.edu.in

³ Department of Zoology and Genetics, Nrupathunga University, Bengaluru-560001, Karnataka, India; royuroyu.dce@ka.gov.in

⁴ School of Pharmacy, Centurion University of Technology and Management, Rayagada-765001, Odisha, India; sanghamitrasatapathy02@gmail.com; sanghamitra.satapathy@cutm.ac.in

⁵ Department of Genetics, School of Sciences, Jain (Deemed to be University), Bengaluru-560027, Karnataka, India; h.malathi@jainuniversity.ac.in

⁶ Department of Pharmacology, Rajiv Academy for Pharmacy, Mathura-281001, Uttar Pradesh, India; shivendra.kumar_mph19@gla.ac.in

⁷ Department of Zoology, NMKRV College for Women Autonomous, Bengaluru-560011, Karnataka, India; sharangouda.nmkrv@rvei.edu.in

* Correspondence: sharangouda.nmkrv@rvei.edu.in;

Received: 28.05.2024; Accepted: 1.01.2025; Published: 6.09.2025

Abstract: Natural cellular metabolism and external stimuli cause organisms to produce reactive oxygen species (ROS). These substances are highly reactive and possess the capacity to degrade and alter the functionality of several cell constituents, including nucleic acids, proteins, and lipids. Oxidative stress refers to a change in the ratio of oxidants to antioxidants that favors oxidants. It is essential for organ function, cell survival, activation, and proliferation, so that reducing and oxidizing states are regulated. The integrated antioxidant systems of aerobic organisms consist of both enzymatic and non-enzymatic antioxidants, which are generally successful in preventing the negative effects of ROS. Many pathological states and illnesses are linked to oxidative stress. In this review, we cover the cellular implications and processes of oxidative stress as well as an overview of the cellular oxidant and antioxidant systems.

Keywords: antioxidant; oxidative stress; reactive oxygen species; mitochondrial pathway.

© 2025 by the authors. This article is an open-access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (<https://creativecommons.org/licenses/by/4.0/>), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. The authors retain copyright of their work, and no permission is required from the authors or the publisher to reuse or distribute this article, as long as proper attribution is given to the original source.

1. Introduction

In the modern era, cancer is a global threat to people worldwide as it is increasing day by day. There is an urgent need for united efforts to find and define more effective medications in light of this staggering loss of human life. One of the essential signaling molecules that is involved in several disorders is ROS [1, 2].

These are very reactive oxygen-containing molecules that are continuously generated by organelles that are metabolizing, specifically the endoplasmic reticulum, peroxisomes, and mitochondria. ROS tends to have an extremely unusual range of effects, acting both pro- and

anti-malignant. Under typical cellular circumstances, antioxidant effectors regulate the dynamic equilibrium that produces and stores minute quantities of ROS [3].

A small threshold point in favor of oxidative stress initiates several signaling cascades that accelerate mutagenesis, damage deoxyribonucleic acid (DNA), increase cellular growth, and intensify the Warburg metabolic effect that cancer cells exploit. Various mechanisms promote carcinogenesis and encourage the transformation of cells into malignancies. It is highly believed that ROS build-up causes anticancer effects, including cell cycle arrest and cell death via necroptosis, apoptosis, and autophagy. The complexity of cell signaling in cancer sparked a hunt for therapeutics that may target and alter signaling molecules' functions to fight various ailments [4]. In this review, we summarize the oxidative stress, redox modulation, and antioxidant systems against ROS.

2. Oxidative Stress (OS)

The imbalance between the production and breakdown of ROS within the cell is known as oxidative stress. It is a potential contributor to pathogenesis that causes several diseases like cardiovascular disease, diabetes, and cancer. During the past two decades, researchers have shown that in normal cells, ROS acts as a secondary messenger for signaling in cells and is essential for several biological mechanisms [5,6]. Also, it acts as a mediator that initiates tumors by inducing various agents both in animal and human models of disease due to its oncogene activation by lack of blood supply and various other variances to the cell [7].

ROS like superoxide ($O_2\bullet$) or its metabolites hydrogen peroxide (H_2O_2) or hydroperoxyl radical ($ROO\bullet$), hydroxyl radical (OH), and nitrogen species (RNS) such as nitric oxide ($NO\bullet$) and peroxynitrite ($\bullet ONOO^-$) damages the macromolecules that include, DNA, lipids, proteins, enzymes and other cell components that lead to necrosis or apoptosis. A very highly vulnerable condition develops in the human body due to changes that are being induced in DNA through oxidative damage, and they get abnormal protein expression. The rich amount of ROS helps to reduce oxidative stress through the concurrent decline in cellular antioxidative stress. This whole process suggests a new way in the development of various diseases like diabetes, neurodegenerative diseases, mitochondrial disease, heart disease, and cancer. Therefore, a real vision for the treatment to reduce the ROS is aimed at curing the proliferative or metastatic cancerous cell [8].

3. Effects of Oxidative Stress on Genetic and Biochemical Mechanisms

Oxidative stress arises when antioxidants become depleted or ROS builds up, upsetting the equilibrium between antioxidants and ROS. Through the activation or silencing of genes that encode structural proteins, transcription factors, and defense enzymes, cells try to counteract the effects of oxidative stress and restore the redox equilibrium. The oxidized/reduced glutathione ratio (2GSH/GSSG) is a significant factor in determining the presence of oxidative stress. Increased ROS generation in the body can alter the structure of DNA and lipids, modify proteins, trigger many transcription factors generated by stress, and produce cytokines that are both pro- and anti-inflammatory [9].

4. Effect of Oxidative Stress on Biomolecular Impairment

ROS targets almost every cell's substrates and alters the function of all kinds of biomolecules. Polyunsaturated fatty acids, i.e., arachidonic acid and docosahexaenoic acid, are particularly prone to oxidation. This can result in the production of malondialdehyde and 4-hydroxynonenal, which are known indicators of lipid oxidative degradation. Both the side chains and the backbone of proteins may be oxidized by reactive oxygen species. This interaction between reactive oxygen species and amino acid side chains results in the production of carbonyl functions. ROS disrupts nucleic acids by altering the structure of purine and pyridine bases, strand breakage, and DNA-protein crosslinking, which can result in DNA mutations. Taking all of this into consideration, it is crucial to remember that the innovative idea of oxidative stress depends on recognizing alterations in the cellular redox state rather than being limited to the free radical destruction of proteins [10].

According to Dean Jones, oxidative stress is a disruption in redox signaling and control in light of new findings on antioxidant mechanisms, oxidative stress indicators, and redox signaling pathways. As a result, the role of antioxidant systems is seen as more complex than simply preventing reactive free radicals. From this point of view, the stages involved in the occurrence of ROS were explained, emphasizing how the electron transport chain (ETC) in the inner membrane of mitochondria exists where this process of production occurs [11,12].

In the ETC, four membrane-bound complexes (IeIV) transfer electrons from NADH and FADH₂ to molecular oxygen, where they eventually generate water. By releasing electrons from the inner membrane during this process, molecular oxygen can be converted to superoxide radical anions (O₂^{•-}). It also results in the production of additional ROS, including hydroxyl radicals (OH[•]), hydrogen peroxide (H₂O₂), and hydroxyl ions (OH). O₂ releases reactive nitrogen species (RNS) when it reacts with nitric oxide (NO) to form peroxynitrite (ONOO). They can also produce nitrogen dioxide (NO₂) and other nitrogenated species, including nitroso-peroxycarbonate (ONOOCCO). Both in reaction to outside stimuli and during activities supported by transition metal cations (iron and copper ions), microglia and brain astrocytes generate RNS and ROS. ROS/RNS may alter lipids, proteins, DNA, and RNA, which may result in the formation of more reactive molecules (Figure 1) [13].

4.1. Effects of oxidative stress on DNA.

ROS can modify DNA in several ways, including base degradation, breaks in single- or double-stranded DNA, and alterations involving purines, pyrimidines, or sugars. This results in translocations, deletions, cross-linking with proteins, and mutations. The majority of these DNA alterations have a significant impact on aging, neurological, cardiovascular, and immunological illnesses, as well as carcinogenesis by producing free radicals or interacting with thiol groups, tobacco smoke, and heavy metals. The most well-known oxidative stress-induced DNA damage is the formation of 8-OH-G, which may serve as a biomarker for the development of cancer. Gene promoter regions include the standard transcription factor sequences. The GC-rich sequences found in these transcription factor-binding sites are at risk of oxidative stress. Formation of 8-OH-G DNA at transcription factor binding sites can change transcription factor binding and, consequently, the expression of related genes, as has been proven for AP-1 and Sp-1 target sequences. In addition to 8-OH-G, it has also been demonstrated that when present in a TATA box, 8, 59 -cyclo-29 -

deoxyadenosine (cyclo-dA) inhibits transcription from a reporter gene in a cell culture. The TATA-binding protein modifies the DNA's bending to start transcription. The presence of cyclo-dA may hinder TATA-binding protein binding [14].

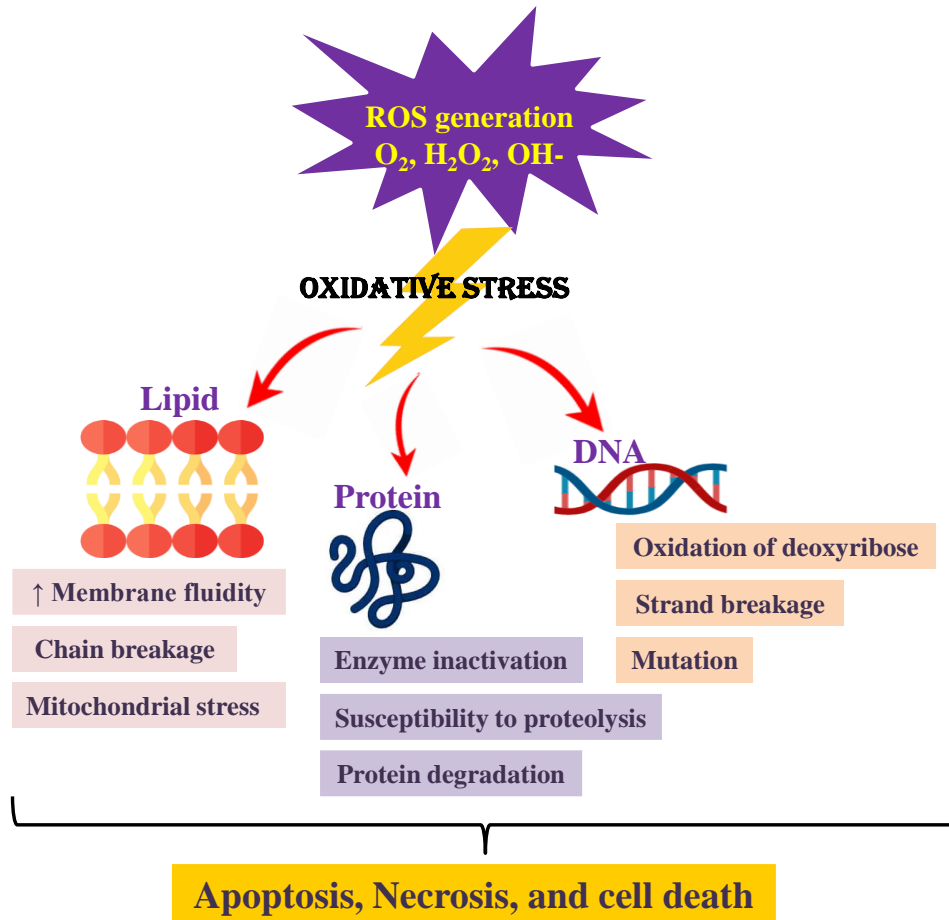


Figure 1. ROS induced oxidative damage to lipids, proteins, and DNA.

4.2. Effects of oxidative stress on lipids.

ROS can cause lipid peroxidation and disturb the lipid bilayer structure of the membrane, which can inactivate enzymes and receptors that are linked to the membrane and improve tissue permeability. Lipid peroxidation products such as thiobarbituric acid, MDA, isoprostanes, and unsaturated aldehydes can inactivate a vast majority of cellular proteins by forming cross-links in the proteins. These products have been indirectly employed as oxidative stress indicators. 4-Hydroxy-2-nonenal releases peroxide, fibronectin, depletes intracellular GSH, and activates the epidermal growth factor receptor [15].

Lipid oxidation possesses the ability to cause harm to cell membranes. The unsaturated fatty acids had a high sensitivity to oxidation and were easily peroxidized by OH attack. Isoprostanes are produced when polyunsaturated fatty acids undergo peroxidation, and their concentrations are thought to correctly represent oxidative stress. Reactive aldehydes like malondialdehyde and 4-hydroxynonenal are also produced by the oxidative damage to lipids. The latter may attach themselves to proteins and interfere with their biological activities [16].

4.3. *Effects of oxidative stress on proteins.*

ROS can lead to the oxidation of several amino acids, cross-linking of proteins, peptide chain fragmentation, and changes in the electrical charge of proteins, and therefore enhance the risk of proteolysis due to the breakdown by certain proteases. Cysteine and methionine-containing protein residues are especially prone to oxidation. Conformational alterations, unfolding, and degradation of proteins are caused by the oxidation of sulfhydryl groups or methionine residues [17]. Metal-catalyzed oxidation particularly affects enzymes with metals on or around their active sites. It has been proven that oxidative alteration of enzymes inhibits their activity. Protein oxidation can lead to unfolding, misfolding, side-chain oxidation, and backbone fragmentation, all of which can cause a reduction in activity. In addition to carbonylation, which produces advanced glycation end products, oxidation of plasma thiol groups causes oxidative damage to proteins [18,19].

Every amino acid is susceptible to oxidation; cysteines and methionines oxidize easily, although disulfide reductases help to reverse many of these oxidations. Numerous irreversible changes may transpire *in vivo*, including the synthesis of S-carboxymethylcysteine and S-(2-Succinyl) cysteine. This indicates that fumarate and dicarbonyl groups are formed and covalently attached to cysteine residues. Carbonyl derivatives are also produced by the oxidation of lysine, proline, arginine, and threonine. These indicators are based on the oxidative species-mediated oxidation of proteins [20,21].

5. **Generation of ROS by Oxidative Phosphorylation**

Mitochondria play a major role in ROS generation while most of the electrons provided to the mitochondrial respiratory chain (MRC) are migrated to cytochrome c oxidase where protons and oxygen react to generate water, and some of them directly react to oxygen, they form superoxide anion ($O_2^{\cdot-}$) and H_2O_2 [22]. The process of stimulation and activation of the extracellular signal-regulated kinase with the help of mitochondrial ROS controls the mass of mitochondria by mitophagy and biogenesis, as well as the regulation process of gluconeogenesis and mitochondrial metabolic pathways. Through regulating the synthesis of amino acid 5-like 1 (GCN5L1), which further regulates FOXO1 at the post-transcriptional level, the extracellular signal-regulated kinase supervised all of these processes. During the process of prolonged fasting, FOXO1 controls the process of mitochondrial content, hepatic gluconeogenesis, and mitochondrial protein acetylation [23].

Moreover, the oxidative stress in the livers of infected patients is induced by Hepatitis C virus (HCV), and this process specifically requires HCV core, NS5A, E1, E2, NS3/4A, and NS4B proteins. Interaction of some proteins occurs with MRC after being localized to the mitochondria. Studies of the liver biopsies obtained from patients suffering from chronic HCV infection showed DNA damage and an increase in 4-hydroxynonenal and malondialdehyde levels, therefore, manifesting the severity of inflammation and fibrosis. HCV disrupts mitochondrial function by causing a disruption of calcium (Ca^{2+}) homeostasis and relocation in mitochondria. Thus, the role of Ca^{2+} chelators in the prevention of oxidative stress induction in HCV-infected cells is well established. Apart from causing mitochondrial oxidative stress and dysfunction, HCV also brings about the inactivation of the innate immune antiviral signaling and inflammatory pathways that are dependent on mitochondrion [24, 25].

6. Enzymatic Cycle of Human Cytochrome P450s

Various studies showed that human CYP 450s are directly involved in the production of H₂O₂ and superoxide. It is now that human CYP 450 enzymes produce ROS during their catalytic cycle despite completing the normal substrate oxidation reaction, and this process is called 'reaction uncoupling'. First, ROS species superoxide radical is produced as a result of the loss of reduced oxygen (O₂⁻), which then further converts to H₂O₂ formation. The entry of the proton into the active site to reduce the oxygen complex, which leads to hydrogen peroxide, avoiding the release of the water molecule, is the second possible step in the formation of ROS [26, 27].

Various CYP isoforms have varying rates for undergoing uncoupling, and substrate concentration is also another important factor that they rely on. Some studies have also shown that CYP1D1 in zebrafish and CYP1B1 had high rates of uncoupling reactions as compared to other isoforms, which have a higher capability for producing ROS due to uncoupling reactions. Structural variations among the different CYP isoforms are responsible for the above-mentioned differences in coupling reaction [28].

7. How ROS Plays its Role in Different Diseases

During pulmonary insufficiency in infants and respiratory distress syndrome in adults, supplementary oxygen is often provided. A hyperoxic condition can be created by this oxygen supply. Studies have shown that this can cause pulmonary injury in animal models, leading to increased oxidative stress and ROS levels. It was previously known that H₂O₂ increase could be due to an increase in CYP activity and reaction uncoupling. Related to this, one previous study showed that a known CYP inhibitor, cimetidine, reduced pulmonary injury in treated lambs, which was due to CYP inhibition. But later on, many studies have shown that instead of CYP inhibition, CYP induction, for instance, 3-methylcholanthrene (3-MC) and beta-naphthoflavone (BNF) have ameliorating effects against hyperoxic lung injury and liver injury [29, 30].

CYP1A1/1A2 is directly involved in hyperoxic lung and liver injury in rats after exposure to a hyperoxic environment. Studies on newborn and adult mice have also shown that CYP1A induction by various inducers like BNF ameliorates hyperoxic lung injury. Among the transcription factors regulating the CYP1 gene expression, aryl hydrocarbon receptors (AHR) are among the ones that are induced by 3-MC [31, 32].

Studies have also shown that in a hyperoxic (more than 95% O₂) environment in adult mice, Cyp1a1^{-/-}, Cyp1a2^{-/-}, and Ahr^{-/-} result in elevated pulmonary injury, indicating that CYP1A enzymes show protective effects against hyperoxia-induced lung injury. In this way, ROS is increased, leading to elevated lipid peroxidation levels and oxidative DNA damage in Cyp1a1^{-/-} and Cyp1a2^{-/-} mice's lungs. Previous studies have also shown that the metabolism of PGF₂-alpha and F₂-isoprostane to a minor metabolite actually lessens lung damage, but there is still no authentic mechanism reported so far on how CYP1A protects against lung injury [33-35].

Among other members of the CYP1 family, CYP1B1 has also been reported to play a role in modulating oxidative stress by decreasing ROS in specific experimental conditions. CYP1B1 is also involved in hyperoxic toxicity in cells that show over-expression of CYP1B1 in the MTT assay. The cells in which the CYP1B1 gene is knocked out show a decrease in caspase pathways, which are responsible for apoptosis [36, 37]. Another important factor that

is responsible for ROS increase is ethanol consumption, which leads to liver cirrhosis and cell death of the liver cells. Three enzymes mainly metabolize alcohol: alcohol dehydrogenase, catalase, and CYP2E. CYP2E1 not only metabolizes ethanol but also induces CYP2E. ROS, through uncoupling, are produced when ethanol is metabolized, which results in alcoholic liver diseases mainly due to elevated hepatic oxidative stress [38]. In cases of chronic ethanol consumption, CYP2E1 CYP2E1-mediated ROS generation results in DNA adducts, which have carcinogenic effects in the liver. Another role of CYP2E1-generated ROS is the cause of non-alcoholic fatty acid liver disease (NAFLD). CYP2E1 can cause various liver abnormalities as ROS produced by it, even in substrate absence due to reaction uncoupling [39, 40].

Among the CYP3 family, CYP3A4 is known as a major enzyme as it metabolizes 50% of the drugs available in the market. It has an active site that can accommodate more than one substrate at a time, which is responsible for an increased rate of uncoupling reaction as CYP2A4 has a tendency to produce ROS as well. CYP3A4-mediated ROS counts for altering protein secretion that is mainly implicated in liver carcinogenesis, which involves two types of signaling, autocrine and paracrine [41, 42].

CYP3A4-mediated ROS generation causes further damage in tumor cells. Methyl 3-(4-nitrophenyl) propionate (NPP), metabolized by CYP3A4, results in ROS generation to induce cytotoxicity. Arachidonic acid has two major metabolic pathways, the first one is cyclooxygenase (COX), and the second one is lipoxygenase (LOX), which is responsible for its metabolism. CYP enzymes are also responsible for arachidonic acid metabolism, including these two pathways. As a result of the above arachidonic acid metabolism, the metabolites that are formed are called eicosanoids, and these eicosanoids play important roles in respiratory, cardiac, and cardiovascular functions and are also known to be involved in cancer-related problems. Among the major metabolites that are formed by the arachidonic acid metabolism are hydroxy eicosatetraenoic acids (HETEs), prostaglandins, leukotrienes, and major metabolites formed by CYP metabolic pathways are epoxyeicosatrienoic acids (EETs) [43, 44].

Among CYP 4 family, CYP4A and CYP4F metabolise arachidonic acid to HETEs, and among CYP 2 family CYP2C and CYP2J families metabolise AA to EETs. Both of these metabolites, EETs and HETEs, are known to play important roles in cardiovascular diseases. More specifically, HETEs, especially 20-HETE, which are produced by CYP4A and CYP4F enzymes, are known to cause inflammation and vasoconstriction, which can further cause cardiac-related problems. The other metabolites, EETs that are generated by CYP2C and CYP2J families are responsible for vasodilation and, in some cases, for angiogenesis and also cause cardiovascular-related problems [45, 46].

8. ROS Molecules Generated by Mitochondrial Pathways

The aging process results in some non-functional organelles and the accumulation of damaged macromolecules, which are sometimes not removed, and these aggregations result in severe interruptions in cellular functions. Studies on nematodes in rodents showed that some specific genes have a critical role in the aging process. Caloric restriction is a non-genetic mechanism that can increase lifespan by some critical mechanisms relating to the aging process. ROS, which are produced by the process of aging, mainly result in the process of senescence. Antioxidant agents in the diet can increase lifespan and cause less cellular damage to cellular events that control the aging process. Some experimental studies on

different mouse strains have supported these results, showing that feeding mouse strains with antioxidant agents. Tremendous research work has been done to show the role of ROS in aging [47].

Elevated ROS levels cause interruptions in defense mechanisms, mainly the autophagy process. It is mediated by ROS molecules and leads to cell death by either autophagy or apoptotic pathways by acting as signaling molecules. No authentic studies have yet proved that cell death may be caused by autophagy. In some cases, the induction of the autophagy process by some ROS-mediated signaling pathways has also been reported. Furthermore, in starvation conditions, ROS production by mitochondria results in an increase in the autophagy process. During the process of starvation, when cells are treated with antioxidants, they completely stop the formation of autophagosomes. This is how ROS helps in the autophagy process. At4 enzymes are mainly responsible for de-lipidation, and if these enzymes are oxidized or inhibited, then the maturation and control of autophagosomes get affected by ROS in starvation. Many dual membrane structures, such as autophagosomes, which protect most of the cytosolic contents, are degraded by autophagy. Then, in turn, these autophagosomes are degraded by lysosomes. Moreover, the autophagy process also discards the non-functional and misfolded proteins and organelles. These non-functional proteins and misfolded proteins can pile up in the nervous and skeletal tissues and cells during the post-mitotic cell division process. In this way, the autophagy process delays aging in the above-mentioned tissues and cells. In starvation and fasting processes, autophagy is induced, which helps in the utilization of amino acids by gluconeogenesis which occurs in the liver. Autophagy is also an active process in tumor suppression and immune response events. ROS molecules are not only generated by mitochondria, but they also target mitochondria. Mitophagy is a special form of autophagy in which damaged and non-functional mitochondria are degraded. In this way, the high proportion of functional mitochondria is maintained [48, 49].

9. Antioxidants and Their System Against ROS

Antioxidants are useful in fighting against many diseases and processes that lead to certain diseases, like Alzheimer's, atherosclerosis, heart disease, cancer, Parkinson's, and diabetes. It is a group of substances that act to prevent the carcinogenic onset, which is useful for cells as a cytotoxic agent that destroys the tumor-causing cells. Antioxidants have the properties of reducing behavior, quenching of singlet oxygen, or donation of hydrogen that can protect these processes, which cause different types of diseases. Living species produce enzymatic (catalase, superoxide dismutase, and peroxidase) and non-enzymatic molecules (flavonoids, ascorbic acid, cysteine, glutathione, and vitamin K) for the safety to fight against these free radicals. Although the natural mechanism of antioxidants is not sufficient, it is important to take antioxidant compounds in addition to our diet [50, 51].

These compounds include antioxidant phytonutrients present in plant foods, antioxidant enzymes, nutrient-derived antioxidants, and metal-binding proteins. The plant's strong antioxidant capacity to scavenge damaging ROS has been linked to its tolerance to diverse environmental stresses. Enhancing plant tolerance has been associated with a strong antioxidant ability to scavenge harmful ROS. Therefore, high antioxidant ROS levels are maintained in both stressed and unstressed cells in various cells [52].

Catalase (CAT), superoxide dismutase (SOD), and glutathione redox cycle enzymes such as glutathione peroxidase and glutathione reductase are antioxidant defense mechanisms

in cells and extracellular fluid to intent the reactive oxygen radicals into a less toxic state. SOD turns superoxide anion enzymatically into H_2O_2 and molecular oxygen. H_2O_2 is eliminated by H_2O and O_2 with two key intracellular enzymes, glutathione peroxidase and catalase. In addition, transferrin, zinc, ceruloplasmin, selenium, ferritin, cytochrome oxidase, and lactoferrin prevent the formation of ROS. Zinc inhibits DNA damage and prevents lipid peroxidation [7].

9.1. Superoxide dismutase (SOD).

SOD is the first enzyme engaged in detoxification and the most powerful antioxidant in the cell. It functions as a first-line defense against ROS. It reduces the toxicity of the potentially harmful superoxide anion by catalyzing the conversion of two superoxide anion molecules into molecular oxygen (O_2) and H_2O_2 (Figure 2). Since a metalloenzyme, SOD requires a metal cofactor to operate. Different versions of the enzyme exist based on the type of metal ion that SOD requires as a cofactor. SOD usually binds zinc (Zn), iron (Fe), manganese (Mn) copper (Cu), and metal ions. Accordingly, SODs are divided into three categories, which are as follows: (i) Fe-SOD, which is frequently present in prokaryotes and some plant chloroplasts, (ii) Mn-SOD, found in prokaryotes and eukaryotic mitochondria, and (iii) In eukaryotes, Cu/Zn-SOD is more common and mostly located in the cytosol but also present in peroxisomes and chloroplasts [55].

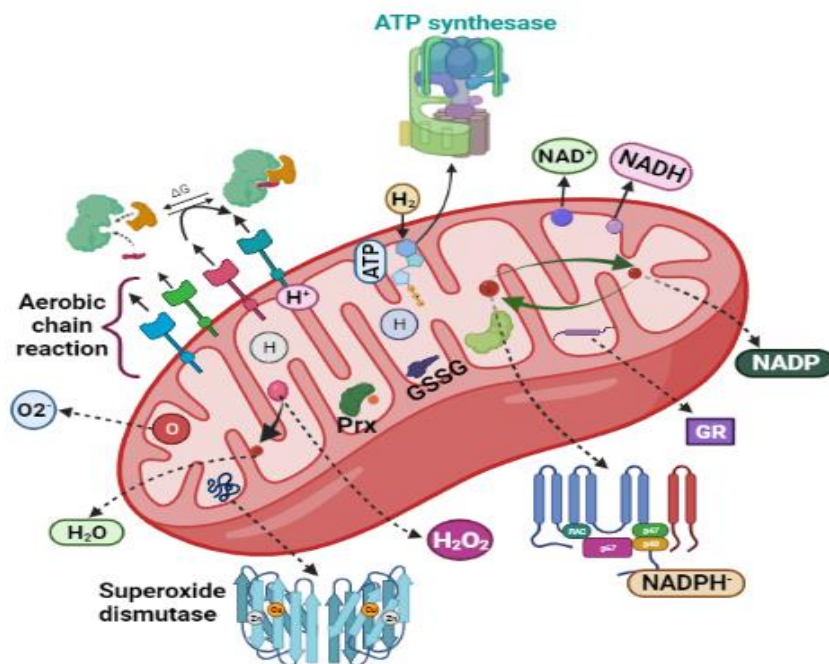


Figure 2. Superoxide dismutase defense mechanism against oxygen-free radicals.

Deficiency of the SOD enzyme is frequent as it shields body cells from free radicals, oxygen radicals, and other dangerous substances that accelerate aging or cell death, the enzyme is therefore essential to cellular health. While free radical production rises with aging, SOD levels decrease. According to some research, taking the recommended daily dosage of SOD supplements can strengthen one's immune system, drastically lower the risk of contracting illnesses, and eventually slow down the aging process [56].

9.2. Catalase (CAT).

The common antioxidant enzyme, catalase, is present in almost all oxygen-consuming biological tissues. CAT is a tetrameric protein (240 kDa) that has four components that are comparable to each other. The *ctl1* gene, which maps to chromosome 11, encodes the CAT protein. Every polypeptide subunit weighs 60 kDa and has one ferriprotoporphyrin. Using iron or manganese as a cofactor, the enzyme catalyzes the breakdown or reduction of H_2O_2 to water and molecular oxygen, completing the detoxification process that SOD had started (Figure 3) [57].

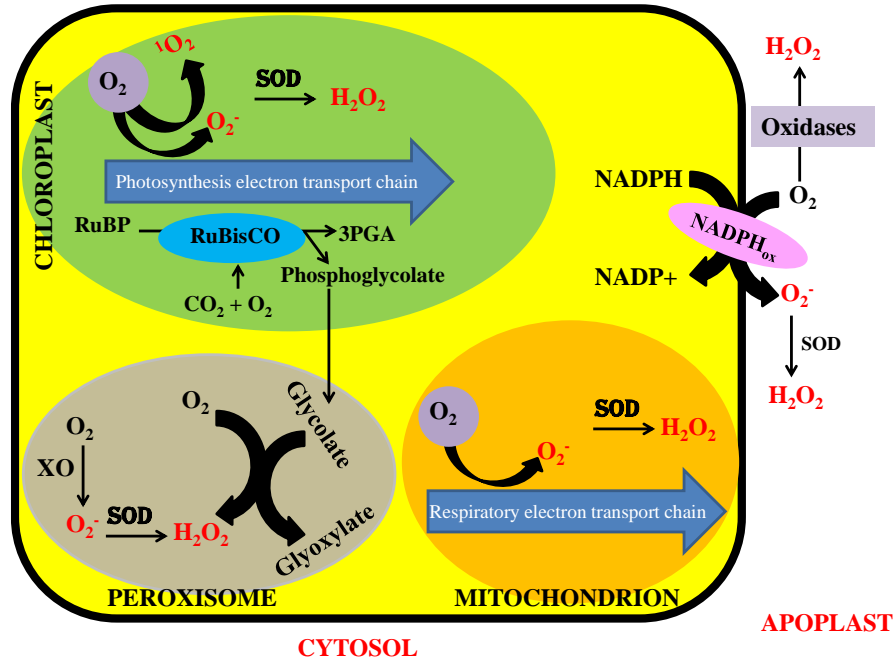


Figure 3. Molecular mechanisms of CAT-induced oxidative stress.

It is widely distributed throughout cells, where it is constantly searching for molecules of H_2O_2 . Millions of molecules of H_2O_2 may be broken down by CAT in one second due to its great efficiency. The enzyme is not present in the mitochondria but is mostly located in the peroxisomes of mammalian cells. The rat heart's mitochondria are the only exception. This indicates that another enzyme called glutathione peroxidase in mammalian cell mitochondria is accountable for breaking down H_2O_2 into H_2O and O_2 . Additionally, CAT has an effective reaction with hydrogen donors that have peroxidase activity. There are two processes involved in the CAT activity. A H_2O_2 molecule converts the heme into an oxyferryl species. When one oxidation equivalent is taken out of the porphyrin ring and one from iron, a porphyrin cation radical is produced. To renew the enzyme in the resting state and produce an oxygen and water molecule, a second H_2O_2 molecule serves as a reducing agent. Although H_2O_2 appears to control various physiological functions, including platelet activation, mitochondrial function, cell death, and proliferation signaling, at low concentrations, it is known to be highly harmful to cells. At high concentrations, on the other hand, it is beneficial to mitochondrial function, cell death, and glucose metabolism. Therefore, CAT's capacity to efficiently restrict the amount of H_2O_2 in cells highlights its significance in the physiological processes, in addition to serving as a first-line antioxidant defense enzyme. Numerous illnesses and abnormalities have been related to the enzyme's absence or mutation [57, 58].

9.3. Glutathione peroxidases (GPx).

This vital intracellular enzyme converts H_2O_2 to H_2O and lipid peroxides to their corresponding alcohols, primarily in the mitochondria and sometimes in the cytoplasm (Figure 4). Selenium is a micronutrient cofactor that is often required for its function. Because of this, GPx is frequently called a selenocysteine peroxidase. The enzyme has a more significant role in protecting cells from oxidative damage by preventing the lipid peroxidation process [58]. Morón and Cortázar state that humans possess GPx1-GPx8, or at least eight GPx enzymes. The corresponding chromosomes for the GPx 1-8 genes (i.e., 3, 14, 5, 19, 6, 6, 1, and 5). GPx1 is the glutathione peroxidase that is most prevalent among selenoperoxidases and is found in almost all cells [59].

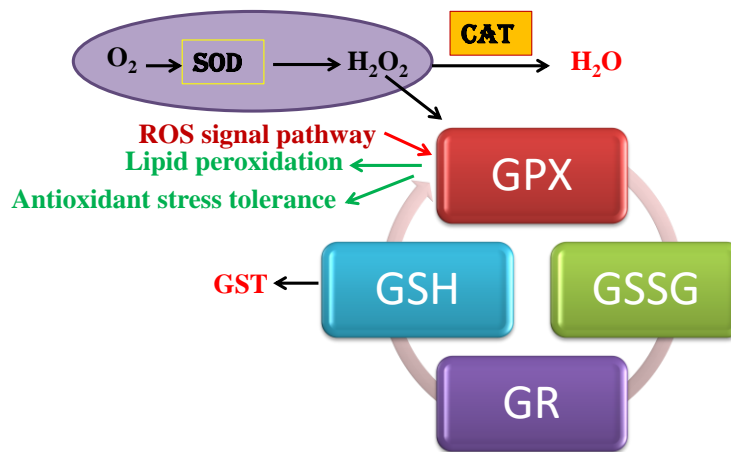


Figure 4. Role of Glutathione peroxidase (GPx) in the ROS signaling pathway.

The gastrointestinal system, particularly the intestine, contains a lot more GPx2. When compared to other organs, the kidney contains the majority of GPx3, but it is also present in extracellular fluids as a glycoprotein [60-62]. When compared to other organs, the kidney contains the majority of GPx3, but it is also present in extracellular fluids as a glycoprotein. This is because phospholipid hydroperoxides can only be broken down by the GPx enzyme Gpx4. The enzyme also has a structural function in sperm maturation that is independent of peroxidase and facilitates the apoptotic response to oxidative stress in a mitochondrial isoform. GPx5 from humans and GPx6 from rodents are not dependent on selenium for their activities, which sets them apart from other glutathione peroxidases. This indicates that these GPx forms might not be able to scavenge H_2O_2 as efficiently, which is an attribute of glutathione peroxidases that are dependent on selenium [63-65].

10. Antioxidants that Neutralize the Radicals

Ascorbic acid (vitamin C) and α -Tocopherol (vitamin E) play a key role as antioxidants. Vitamin C has antioxidant activity in the cytoplasm and extracellular fluids, which prevents the elimination of anti-proteases with oxidants. Vitamin E and flavonoids prohibit lipid peroxidation in the cell membrane. Protection from oxidative stress is the most documented activity of flavonoids. Moreover, glutathione is a versatile and intracellular sulfur-containing antioxidant with the potential for metal-chelating and anti-glycation. N-acetyl-L-cysteine is a thiol that contains an antioxidant utilized to reduce oxidative stress conditions [66].

11. Conclusion

Oxidative stress can result from an excess of ROS produced by oxygen-consuming metabolic activities that shift the antioxidant-oxidant status balance in favor of the oxidants. Both internal metabolic processes and external elements like cigarette smoke or air pollution create ROS. Because of the unpaired electrons in their structure, ROS are very reactive molecules that react with a variety of biological macromolecules in cells, including proteins, lipids, carbohydrates, and nucleic acids, changing their activities. ROS also modifies chromatin remodeling through changes in histone acetylation/deacetylation and up-regulates redox-sensitive transcription factors, which in turn impact the expression of many genes. For organ function, cell survival, activation, and proliferation, redox state regulation is essential. The activities of glutathione peroxidase (GPX), CAT, and SOD comprise the first-line antioxidant defense system, which is fundamental to the overall defensive mechanisms, strategies, and techniques in biological systems.

Author Contributions

Conceptualization, D.J.; writing—original draft preparation, D.J.; validation, C.K. and U.B.R.; data curation, S.S. and M.H.; writing—review and editing, S.K.; supervision, S.J.P. All authors have read and agreed to the published version of the manuscript.

Institutional Review Board Statement

Not applicable

Informed Consent Statement

Not applicable

Data Availability Statement

No new data were created or analyzed in this study. Data sharing is not applicable.

Funding

The authors further declare that no funds, grants, or other support were received from any government. Or another private funding agency that was received during the preparation of this manuscript.

Acknowledgments

The author would like to express gratitude to their University for being a consistent source of support and for establishing the environment for research.

Conflicts of Interest

The authors declare no conflict of interest.

References

1. Wehbe, N.; Slika, H.; Mesmar, J.; Nasser, S.A.; Pintus, G.; Baydoun, S.; Badran, A.; Kobeissy, F.; Eid, A.H.; Baydoun, E. The Role of Epac in Cancer Progression. *Int J Mol Sci* **2020**, *5*, 6489, <https://doi.org/10.3390/ijms21186489>.
2. El-Hachem, N.; Fardoun, M.M.; Slika, H.; Baydoun, E.; Eid, A.H. Repurposing Cilostazol for Raynaud's Phenomenon. *Curr Med Chem* **2021**, *28*, 2409-2417, <https://doi.org/10.2174/0929867327666200903114154>.
3. Moloney, J.N.; Cotter, T.G. ROS signalling in the biology of cancer. *Semin Cell Dev Biol* **2018**, *80*, 50-64, <https://doi.org/10.1016/j.semcdb.2017.05.023>.
4. Helfinger, V.; Schröder, K. Redox control in cancer development and progression. *Mol Aspects Med* **2018**, *63*, 88-98, <https://doi.org/10.1016/j.mam.2018.02.003>.
5. Clerkin, J.S.; Naughton, R.; Quiney, C.; Cotter, T.G. Mechanisms of ROS modulated cell survival during carcinogenesis. *Cancer Lett* **2008**, *266*, 30-6, <https://doi.org/10.1016/j.canlet.2008.02.029>.
6. Ushio-Fukai, M.; Nakamura, Y. Reactive oxygen species and angiogenesis: NADPH oxidase as target for cancer therapy. *Cancer Lett* **2008**, *266*, 37-52, <https://doi.org/10.1016/j.canlet.2008.02.044>.
7. Jain, D.; Murti, Y.; Khan, W.U.; Hossain, R.; Hossain, M.N.; Agrawal, K.K.; Ashraf, R.A.; Islam, M.T.; Janmeda, P.; Taheri, Y.; Alshehri, M.M.; Daştan, S.D.; Yeskaliyeva, B.; Kipchakbayeva, A.; Sharifi-Rad, J.; Cho, W.C. Role of therapeutic bioactive compounds in hepatocellular carcinoma. *Oxid Med Cell Longev* **2021**, 9068850, <https://doi.org/10.1155/2021/9068850>.
8. Janmeda, P.; Jain, D.; Chaudhary, P.; Meena, M.; Singh, D. A review on N-nitrosodiethylamine (a multipotent carcinogenic agent), its major risk assessment and precautions. *J. Appl. Toxicol.* **2024**, *4*, <https://doi.org/10.1002/jat.4574>.
9. D'Autréaux, B.; Toledano, M.B. ROS as signalling molecules: mechanisms that generate specificity in ROS homeostasis. *Nat Rev Mol Cell Biol* **2007**, *8*, 813-24, <https://doi.org/10.1038/nrm2256>.
10. Gandhi, S.; Abramov, A.Y. Mechanism of oxidative stress in neurodegeneration. *Oxid. Med. Cell. Longev* **2012**, *2012*, 428010, <http://dx.doi.org/10.1155/2012/428010>.
11. Finley, J.W.; Kong, A.N.; Hintze, K.J.; Jeffery, E.H.; Ji, L.L.; Lei, X.G. Antioxidants in foods: state of the science important to the food industry. *J Agric Food Chem* **2011**, *13*, 6837-6846, <http://dx.doi.org/10.1021/jf2013875>.
12. Persson, T.; Popescu, B.O.; Cedazo-Minguez, A. Oxidative stress in Alzheimer's disease: why did antioxidant therapy fail. *Oxid. Med. Cell. Longev* **2014**, *2014*, 427318, <http://dx.doi.org/10.1155/2014/427318>.
13. Doorn, J.A.; Petersen, D.R. Covalent adduction of nucleophilic amino acids by 4-hydroxynonenal and 4-oxononenal. *Chem Biol Interact* **2003**, *1*, 143-144, [http://dx.doi.org/10.1016/s0009-2797\(02\)00178-3](http://dx.doi.org/10.1016/s0009-2797(02)00178-3).
14. Marietta, C.; Gulam, H.; Brooks, P.J. A single 8,5'-cyclo-2'-deoxyadenosine lesion in a TATA box prevents binding of the TATA binding protein and strongly reduces transcription *in vivo*. *DNA Repair* **2002**, *1*, 967-75, [http://dx.doi.org/10.1016/s1568-7864\(02\)00148-9](http://dx.doi.org/10.1016/s1568-7864(02)00148-9).
15. Tsukagoshi, H.; Kawata, T.; Shimizu, Y.; Ishizuka, T.; Dobashi, K.; Mori M. 4-Hydroxy-2-nonenal enhances fibronectin production by IMR-90 human lung fibroblasts partly via activation of epidermal growth factor receptor-linked extracellular signal-regulated kinase p44/42 pathway. *Toxicol Appl Pharmacol* **2002**, *184*, 127-135, <https://doi.org/10.1006/taap.2002.9514>.
16. Liu, T.; Stern, A.; Roberts, L.J.; Morrow, J.D. The isoprostanes: novel prostaglandin-like products of the free radical-catalyzed peroxidation of arachidonic acid. *J Biomed Sci* **1999**, *6*, 226-35, <http://dx.doi.org/10.1007/BF02253564>.
17. Kelly, F.J.; Mudway, I.S. Protein oxidation at the air-lung interface. *Amino Acids* **2003**, *25*, 375-396, <http://dx.doi.org/10.1007/s00726-003-0024-x>.
18. Headlam, H.A.; Davies, M.J. Markers of protein oxidation: different oxidants give rise to variable yields of bound and released carbonyl products. *Free Radic Biol Med* **2004**, *36*, 1175-1184, <http://dx.doi.org/10.1016/j.freeradbiomed.2004.02.017>.
19. Sung, Ch-Ch.; Hsu, Y.Ch.; Chen, Ch-Ch.; Lin, Y.F.; Wu, Ch-Ch. Oxidative stress and nucleic acid oxidation in patients with chronic kidney disease. *Oxid. Med. Cell. Longev* **2013**, *2013*, 301982, <http://dx.doi.org/10.1155/2013/301982>.
20. Alderson, N.L.; Wang, Y.; Blatnik, M.; Frizzell, N.; Walla, M.D.; Lyons, T.J.; Alt, N.; Carson, J.A.; Nagai, R.; Thorpe, S.R.; Baynes, J.W. S-(2-Succinyl)cysteine: a novel chemical modification of tissue

- proteins by a Krebs cycle intermediate. *Arch Biochem Biophys* **2006**, *450*, 1-8, <http://dx.doi.org/10.1016/j.abb.2006.03.005>.
21. Zeng, J.; Davies, M.J. Evidence for the formation of adducts and S-(carboxymethyl) cysteine on reaction of alpha-dicarbonyl compounds with thiol groups on amino acids, peptides, and proteins. *Chem Res Toxicol* **2005**, *18*, 1232-41, <http://dx.doi.org/10.1021/tx050074u>.
 22. Jain, D.; Janmeda, P. Exposure, formation, various available treatments to combat Hepatocellular carcinoma: A Comprehensive review. *Appl. Biol. Chem. J* **2023**, *4*, 69-83, <https://doi.org/10.52679/tabcj.2023.0007>.
 23. Wang, L.; Scott, I.; Zhu, L.; Wu, K.; Han, K.; Chen, Y.; Gucek, M.; Sack, M.N. GCN5L1 modulates cross-talk between mitochondria and cell signaling to regulate FoxO1 stability and gluconeogenesis. *Nat Commun* **2017**, *8*, 523, <https://doi.org/10.1038/s41467-017-00521-8>.
 24. Paracha, U.Z.; Fatima, K.; Alqahtani, M.; Chaudhary, A.; Abuzenadah, A.; Damanhour, G.; Qadri, I. Oxidative stress and hepatitis C virus. *Virology* **2013**, *J10*, 251, <https://doi.org/10.1186/1743-422X-10-251>.
 25. Presser, L.D.; Haskett, A.; Waris, G. Hepatitis C virus-induced furin and thrombospondin-1 activate TGF- β 1: role of TGF- β 1 in HCV replication. *Virology* **2011**, *10*, 284-96, <https://doi.org/10.1016/j.virol.2010.12.051>.
 26. Denisov, I.G.; Makris, T.M.; Sligar, S.G.; Schlichting, I. Structure and chemistry of cytochrome P450. *Chem Rev* **2005**, *105*, 2253-2277, <https://doi.org/10.1021/cr0307143>.
 27. Bae, Y.S.; Oh, H.; Rhee, S.G.; Yoo, Y.D. Regulation of reactive oxygen species generation in cell signaling. *Mol Cells* **2011**, *32*, 491-509, <https://doi.org/10.1007/s10059-011-0276-3>.
 28. Harskamp, J.; Britz-McKibbin, P.; Wilson, J.Y. Functional screening of cytochrome P450 activity and uncoupling by capillary electrophoresis. *Anal Chem* **2012**, *84*, 862-866, <https://doi.org/10.1021/ac202787n>.
 29. Sweeney, R.M.; McAuley, D.F. Acute respiratory distress syndrome. *Lancet* **2016**, *388*, 2416-2430, [https://doi.org/10.1016/S0140-6736\(16\)00578-X](https://doi.org/10.1016/S0140-6736(16)00578-X).
 30. Buczynski, B.W.; Maduekwe, E.T.; O'Reilly, M.A. The role of hyperoxia in the pathogenesis of experimental BPD. *Semin Perinatol* **2013**, *37*, 69-78, <https://doi.org/10.1053/j.semperi.2013.01.002>.
 31. Couroucli, X.I.; Liang, Y.H.; Jiang, W.; Wang, L.; Barrios, R.; Yang, P.; Moorthy, B. Prenatal administration of the cytochrome P4501A inducer, B-naphthoflavone (BNF), attenuates hyperoxic lung injury in newborn mice: implications for bronchopulmonary dysplasia (BPD) in premature infants. *Toxicol Appl Pharmacol* **2011**, *15*, 83-94, <https://doi.org/10.1016/j.taap.2011.06.018>.
 32. Maturu, P.; Wei-Liang, Y.; Jiang, W.; Wang, L.; Lingappan, K.; Barrios, R.; Liang, Y.; Moorthy, B.; Couroucli, X.I. Correction to: Newborn Mice Lacking the Gene for Cyp1a1 Are More Susceptible to Oxygen-Mediated Lung Injury, and Are Rescued by Postnatal β -Naphthoflavone Administration: Implications for Bronchopulmonary Dysplasia in Premature Infants. *Toxicol Sci* **2022**, *187*, 187, <https://doi.org/10.1093/toxsci/kfac031>.
 33. Lingappan, K.; Jiang, W.; Wang, L.; Wang, G.; Couroucli, X.I.; Shivanna, B.; Welty, S.E.; Barrios, R.; Khan, M.F.; Nebert, D.W.; Roberts, L.J.; Moorthy, B. Mice deficient in the gene for cytochrome P450 (CYP)1A1 are more susceptible than wild-type to hyperoxic lung injury: evidence for protective role of CYP1A1 against oxidative stress. *Toxicol Sci* **2014**, *41*, 68-77, <https://doi.org/10.1093/toxsci/kfu106>.
 34. Wang, L.; Lingappan, K.; Jiang, W.; Couroucli, X.I.; Welty, S.E.; Shivanna, B.; Barrios, R.; Wang, G.; Firoze, K.M.; Gonzalez, F.J.; Jackson, R.L.; Moorthy, B. Disruption of cytochrome P4501A2 in mice leads to increased susceptibility to hyperoxic lung injury. *Free Radic Biol Med* **2015**, *82*, 47-59, <https://doi.org/10.1016/j.freeradbiomed.2015.01.019>.
 35. Lingappan, K.; Maity, S.; Jiang, W.; Wang, L.; Couroucli, X.; Veith, A.; Zhou, G.; Coarfa, C.; Moorthy, B. Role of Cytochrome P450 (CYP)1A in Hyperoxic Lung Injury: Analysis of the Transcriptome and Proteome. *Sci Rep* **2017**, *4*, 642, <https://doi.org/10.1038/s41598-017-00516-x>.
 36. Jennings, B.L.; George, L.W.; Pingili, A.K.; Khan, N.S.; Estes, A.M.; Fang, X.R.; Gonzalez, F.J.; Malik KU. Estrogen metabolism by cytochrome P450 1B1 modulates the hypertensive effect of angiotensin II in female mice. *Hypertension* **2014**, *64*, 134-40, <https://doi.org/10.1161/HYPERTENSIONAHA.114.03275>.
 37. Dinu, D.; Chu, C.; Veith, A.; Lingappan, K.; Couroucli, X.; Jefcoate, C.R.; Sheibani, N.; Moorthy, B. Mechanistic role of cytochrome P450 (CYP)1B1 in oxygen-mediated toxicity in pulmonary cells: A novel target for prevention of hyperoxic lung injury. *Biochem Biophys Res Commun* **2016**, *476*, 346-351, <https://doi.org/10.1016/j.bbrc.2016.05.125>.

38. Louvet, A.; Mathurin P. Alcoholic liver disease: mechanisms of injury and targeted treatment. *Nat Rev Gastroenterol Hepatol* **2015**, *12*, 231-242, <https://doi.org/10.1038/nrgastro.2015.35>.
39. Linhart, K.; Bartsch, H.; Seitz, H.K. The role of reactive oxygen species (ROS) and cytochrome P-450 2E1 in the generation of carcinogenic etheno-DNA adducts. *Redox Biol* **2014**, *3*, 56-62, <https://doi.org/10.1016/j.redox.2014.08.009>.
40. Leung, T.M.; Nieto, N. CYP2E1 and oxidant stress in alcoholic and non-alcoholic fatty liver disease. *J Hepatol* **2013**, *58*, 395-398, <https://doi.org/10.1016/j.jhep.2012.08.018>.
41. Grinkova, Y.V.; Denisov, I.G.; McLean, M.A.; Sligar, S.G. Oxidase uncoupling in heme monooxygenases: human cytochrome P450 CYP3A4 in Nanodiscs. *Biochem Biophys Res Commun* **2013**, *25*, 1223-1227, <https://doi.org/10.1016/j.bbrc.2012.12.072>.
42. Zangar, R.C.; Bollinger, N.; Weber, T.J.; Tan, R.M.; Markillie, L.M.; Karin, N.J. Reactive oxygen species alter autocrine and paracrine signaling. *Free Radic Biol Med* **2011**, *51*, 2041-2047, <https://doi.org/10.1016/j.freeradbiomed.2011.09.001>.
43. Reczek, C. R.; Chandel, N.S. The two faces of reactive oxygen species in cancer. *Annu Rev Cancer Biol* **2017**, *1*, 79-98, <https://doi.org/10.1146/annurev-cancerbio-041916-065808>.
44. Sun, X.; Ai, M.; Wang, Y.; Shen, S.; Gu, Y.; Jin, Y.; Zhou, Z.; Long, Y.; Yu, Q. Selective induction of tumor cell apoptosis by a novel P450-mediated reactive oxygen species (ROS) inducer methyl 3-(4-nitrophenyl) propiolate. *J Biol Chem* **2013**, *288*, 8826-8837, <https://doi.org/10.1074/jbc.M112.429316>.
45. Johnson, A.L.; Edson, K.Z.; Totah, R.A.; Rettie, A.E. Cytochrome P450 ω -Hydroxylases in Inflammation and Cancer. *Adv Pharmacol* **2015**, *74*, 223-62, <https://doi.org/10.1016/bs.apha.2015.05.002>.
46. Jamieson, K.L.; Endo, T.; Darwesh, A.M.; Samokhvalov, V.; Seubert, J.M. Cytochrome P450-derived eicosanoids and heart function. *Pharmacol Ther* **2017**, *179*, 47-83, <https://doi.org/10.1016/j.pharmthera.2017.05.005>.
47. Kroemer, G.; Levine, B. Autophagic cell death: the story of a misnomer. *Nat Rev Mol Cell Biol* **2008**, *9*, 1004-10, <https://doi.org/10.1038/nrm2529>.
48. Scherz-Shouval, R.; Shvets, E.; Fass, E.; Shorer, H.; Gil, L.; Elazar, Z. Reactive oxygen species are essential for autophagy and specifically regulate the activity of Atg4. *EMBO J* **2007**, *4*, 1749-60, <https://doi.org/10.1038/sj.emboj.7601623>.
49. Furge, L.L.; Guengerich, F.P. Cytochrome P450 enzymes in drug metabolism and chemical toxicology: An introduction. *Biochem Mol Biol Educ* **2006**, *34*, 66-74, <https://doi.org/10.1002/bmb.2006.49403402066>.
50. Valko, M.; Leibfritz, D.; Moncol, J.; Cronin, M.T.; Mazur, M.; Telser, J. Free radicals and antioxidants in normal physiological functions and human disease. *Int J Biochem Cell Biol* **2007**, *39*, 44-84, <https://doi.org/10.1016/j.biocel.2006.07.001>.
51. Kähkönen, M.P.; Hopia, A.I.; Vuorela, H.J.; Rauha, J.P.; Pihlaja, K.; Kujala, T.S.; Heinonen, M. Antioxidant activity of plant extracts containing phenolic compounds. *J Agric Food Chem* **1999**, *47*, 3954-62, <https://doi.org/10.1021/jf990146l>.
52. Mohammad, Z.; Alakbar, Q.R.; Mashalla, B.S.; Ali, A.M. The Effect of the Interaction between Genotypes and Drought Stress on the Superoxide Dismutase and Chlorophyll Content in Durum Wheat Landraces. *Turk J Biol* **2009**, *33*, 1-7, <https://doi.org/10.3906/biy-0801-12>.
53. Fridovich, I. Superoxide radical and superoxide dismutases. *Annu Rev Biochem* **1995**, *64*, 97-112, <https://doi.org/10.1146/annurev.bi.64.070195.000525>.
54. Dringen, R.; Pawlowski, P.G.; Hirrlinger, J. Peroxide detoxification by brain cells. *J Neurosci Res* **2005**, *79*, 157-65, <https://doi.org/10.1002/jnr.20280>.
55. Gill, S.S.; Tuteja, N. Reactive oxygen species and antioxidant machinery in abiotic stress tolerance in crop plants. *Plant Physiol Biochem* **2010**, *48*, 909-30, <https://doi.org/10.1016/j.plaphy.2010.08.016>.
56. Krishnamurthy, P.; Wadhvani, A. Antioxidant Enzymes and Human Health. In Antioxidant Enzyme, El-Missiry, M.A.A., Ed.; IntechOpen: Rijeka, **2012**; <https://doi.org/10.5772/48109>.
57. Chelikani, P.; Fita, I.; Loewen, P.C. Diversity of structures and properties among catalases. *Cell Mol Life Sci* **2004**, *61*, 192-208, <https://doi.org/10.1007/s00018-003-3206-5>.
58. Góth, L.; Rass, P.; Páy, A. Catalase enzyme mutations and their association with diseases. *Mol Diagn* **2004**, *8*, 141-149, <https://doi.org/10.1007/BF03260057>.
59. Castilla-Cortázar, I.; Morón, Ú.M. Protection Against Oxidative Stress and “IGF-I Deficiency Conditions”. In Antioxidant Enzyme, El-Missiry, M.A.A., Ed.; IntechOpen: Rijeka, **2012**; <https://doi.org/10.5772/51047>.

60. Drevet, J.R. The antioxidant glutathione peroxidase family and spermatozoa: a complex story. *Mol Cell Endocrinol* **2006**, *250*, 70-9, <https://doi.org/10.1016/j.mce.2005.12.027>.
61. Baek, I.J.; Seo, D.S.; Yon, J.M.; Lee, S.R.; Jin, Y.; Nahm, S.S.; Jeong, J.H.; Choo, Y.K.; Kang, J.K.; Lee, B.J.; Yun, Y.W.; Nam, S.Y. Tissue expression and cellular localization of phospholipid hydroperoxide glutathione peroxidase (PHGPx) mRNA in male mice. *J Mol Histol* **2007**, *38*, 237-44, <https://doi.org/10.1007/s10735-007-9092-7>.
62. Burk, R.F.; Olson, G.E.; Winfrey, V.P.; Hill, K.E.; Yin, D. Glutathione peroxidase-3 produced by the kidney binds to a population of basement membranes in the gastrointestinal tract and in other tissues. *Am J Physiol Gastrointest Liver Physiol* **2011**, *301*, G32-8, <https://doi.org/10.1152/ajpgi.00064.2011>.
63. Liang, H.; Ran, Q.; Jang, Y.C.; Holstein, D.; Lechleiter, J.; McDonald-Marsh, T.; Musatov, A.; Song, W.; Van, R.H.; Richardson, A. Glutathione peroxidase 4 differentially regulates the release of apoptogenic proteins from mitochondria. *Free Radic Biol Med* **2009**, *47*, 312-20, <https://doi.org/10.1016/j.freeradbiomed.2009.05.012>.
64. Noblanc, A.; Kocer, A.; Chabory, E.; Vernet, P.; Saez, F.; Cadet, R.; Conrad, M.; Drevet, J.R. Glutathione peroxidases at work on epididymal spermatozoa: an example of the dual effect of reactive oxygen species on mammalian male fertilizing ability. *J Androl* **2011**, *32*, 641-50, <https://doi.org/10.2164/jandrol.110.012823>.
65. Kryukov, G.V.; Castellano, S.; Novoselov, S.V.; Lobanov, A.V.; Zehtab, O.; Guigó, R.; Gladyshev, V.N. Characterization of mammalian selenoproteomes. *Science* **2003**, *300*, 1439-43, <https://doi.org/10.1126/science.1083516>.
66. Kurutas, E.B. The importance of antioxidants which play the role in cellular response against oxidative/nitrosative stress: current state. *Nutri J* **2016**, *15*, 71, <https://doi.org/10.1186/s12937-016-0186-5>.

Publisher's Note & Disclaimer

The statements, opinions, and data presented in this publication are solely those of the individual author(s) and contributor(s) and do not necessarily reflect the views of the publisher and/or the editor(s). The publisher and/or the editor(s) disclaim any responsibility for the accuracy, completeness, or reliability of the content. Neither the publisher nor the editor(s) assume any legal liability for any errors, omissions, or consequences arising from the use of the information presented in this publication. Furthermore, the publisher and/or the editor(s) disclaim any liability for any injury, damage, or loss to persons or property that may result from the use of any ideas, methods, instructions, or products mentioned in the content. Readers are encouraged to independently verify any information before relying on it, and the publisher assumes no responsibility for any consequences arising from the use of materials contained in this publication.