

Review Article

Antioxidants and Oxidative Stress: Biochemical Mechanisms, Medicinal Applications and Bioavailability of Natural Products

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ABSTRACT

Oxidative stress is an imbalance between the generation of reactive oxygen and nitrogen species (ROS/RNS) and the capacity of cellular antioxidant systems to control them; persistent imbalance damages lipids, proteins and DNA and contributes to cardiovascular disease, neurodegeneration, cancer and metabolic disorders. This review integrates the chemistry and cellular sources of reactive species with the architecture of the antioxidant defence network and the pharmacology of natural antioxidants. Endogenous defence comprises enzymatic catalysts (superoxide dismutases, catalase, the glutathione peroxidase/reductase cycle and the peroxiredoxin/thioredoxin system) and non-enzymatic reductants (glutathione, ascorbate, tocopherols and carotenoids) supported by trace-element cofactors. Exogenous phenolic antioxidants scavenge radicals through hydrogen atom transfer (HAT), single electron transfer–proton transfer (SET-PT) and sequential proton-loss electron transfer (SPLET), chelate transition metals and, more importantly in vivo, activate the KEAP1–Nrf2–ARE axis and modulate NF-κB and MAPK signalling. Large randomised trials (ATBC, CARET, SELECT, HOPE) and Cochrane meta-analysis show that high-dose synthetic supplementation is ineffective or harmful—the "antioxidant paradox"—which is explained by pro-oxidant conversion, suppression of mitohormesis and blunting of adaptive redox signalling. In parallel, the clinical translation of curcumin, quercetin, resveratrol and epigallocatechin gallate is limited by poor solubility, chemical instability, extensive first-pass metabolism and efflux transport. Nano-encapsulation platforms—liposomes and phytosomes, solid lipid and polymeric nanoparticles, nanoemulsions and cyclodextrin complexes—increase oral bioavailability by one to two orders of magnitude and protect the payload during gastrointestinal transit. Effective antioxidant therapy should therefore shift from stoichiometric scavenging by unregulated high-dose supplements toward phytochemical-rich diets, targeted Nrf2 modulators and rationally designed delivery systems evaluated in adequately powered clinical trials.

Keywords: Antioxidant paradox; Bioavailability; Nanoencapsulation; Nrf2; Oxidative stress; Polyphenols; Reactive oxygen species

1. INTRODUCTION

Redox homeostasis is a dynamic steady state in which the rate of formation of reactive oxygen and nitrogen species (ROS/RNS) is matched by the capacity of intrinsic and extrinsic antioxidant systems to remove them [1,2]. Molecular oxygen is indispensable for aerobic energy metabolism, yet its stepwise, one-electron reduction in the mitochondrial respiratory chain unavoidably generates partially reduced intermediates that are simultaneously useful signalling molecules and potentially destructive oxidants [2,3]. The concept of oxidative stress, originally formulated by Sies as "a disturbance in the pro-oxidant–antioxidant balance in favour of the former", has since been refined to include the loss of redox signalling and control, and is now recognised as a common mechanistic thread linking ageing, cardiovascular disease, neurodegeneration, cancer and metabolic disease [1].

Interest in antioxidants as therapeutic agents grew from strong laboratory and epidemiological evidence, but expectations were tempered by large randomised trials in which high-dose vitamin supplements failed to reduce, and sometimes increased, disease incidence and mortality [4–6]. At the same time, thousands of naturally occurring phenolic compounds with impressive activity in vitro show weak effects in humans because they are poorly absorbed, rapidly metabolised and quickly eliminated [7]. These two problems—the "antioxidant

paradox" and low bioavailability—have reshaped the field and are the main reasons why modern antioxidant pharmacology emphasises redox signalling, hormesis and advanced delivery systems rather than simple radical scavenging [8,9].

The aim of this review is to provide an integrated account of (i) the chemistry and cellular sources of reactive species, (ii) the molecular damage they inflict, (iii) the enzymatic and non-enzymatic defence network, (iv) the chemical and signalling mechanisms by which natural antioxidants act, (v) the clinical evidence for antioxidant intervention in chronic disease, including the antioxidant paradox, and (vi) the pharmaceutical barriers to bioavailability and the nano-delivery strategies developed to overcome them. Literature was retrieved from PubMed, Scopus and Web of Science up to 2025 using combinations of the terms "oxidative stress", "reactive oxygen species", "antioxidant enzymes", "polyphenol", "Nrf2", "antioxidant paradox", "bioavailability" and "nanoencapsulation"; priority was given to primary mechanistic studies, randomised controlled trials, systematic reviews and authoritative reviews.

2. CHEMISTRY AND CELLULAR SOURCES OF REACTIVE SPECIES

2.1. Free radicals and non-radical oxidants

A free radical is any atom or molecule that contains one or more unpaired electrons in its outer orbital, a configuration that confers high reactivity and a strong tendency to abstract electrons or hydrogen atoms from neighbouring molecules [2]. The principal ROS in biology are the superoxide anion radical ($O_2^{\cdot-}$), the hydroxyl radical ($\cdot OH$), the hydroperoxyl radical ($HO_2^{\cdot}$), lipid peroxyl ($LOO^{\cdot}$) and alkoxyl ($LO^{\cdot}$) radicals, together with the non-radical oxidants hydrogen peroxide (H_2O_2), singlet oxygen (1O_2), hypochlorous acid ($HOCl$) and organic hydroperoxides ($LOOH$). The RNS comprise nitric oxide ($\cdot NO$), nitrogen dioxide ($\cdot NO_2$) and peroxynitrite ($ONOO^{\cdot}$), the product of the diffusion-limited reaction between $\cdot NO$ and $O_2^{\cdot-}$ [2]. Superoxide is a relatively weak oxidant with a short diffusion radius, whereas H_2O_2 is uncharged, comparatively stable and membrane-permeant, properties that make it the principal second messenger of redox signalling [2]. The hydroxyl radical is the most reactive species known in biology; it is generated from H_2O_2 by Fenton chemistry in the presence of Fe^{2+} or Cu^+ ($H_2O_2 + Fe^{2+} \rightarrow \cdot OH + OH^- + Fe^{3+}$) and reacts at diffusion-controlled rates with virtually every biomolecule at its site of formation [2].

2.2. Physiological roles of reactive species

At low to moderate concentrations, reactive species are not merely toxic by-products but act as second messengers that regulate proliferation, differentiation, migration and survival [2]. Nitric oxide generated by endothelial nitric oxide synthase controls vascular tone through activation of soluble guanylate cyclase [2], while the localised burst of $O_2^{\cdot-}$ and H_2O_2 generated by phagocyte NADPH oxidase (NOX2) is essential for microbial killing, as demonstrated by the recurrent infections of patients with chronic granulomatous disease [10]. Moderate shifts in cellular redox state also serve as sensors that mediate adaptation to hypoxia through stabilisation of hypoxia-inducible factors [2]. The term "oxidative eustress" has been proposed for this physiological, signal-generating range of ROS, in contrast to "oxidative distress", the supraphysiological range that causes indiscriminate damage [2].

2.3. Endogenous sources

Mitochondrial electron transport chain. Mitochondria are generally regarded as the main intracellular source of ROS. Electrons leak from the flavin site of Complex I (NADH:ubiquinone oxidoreductase) and from the ubiquinol-oxidising site (Q_o) of Complex III to reduce O_2 to $O_2^{\cdot-}$, which is released into the matrix (Complexes I and III) and the intermembrane space (Complex III) [3,11]. The rate of superoxide production is highly dependent on the redox state of the ubiquinone pool and on the proton-motive force, being highest during reverse electron transport when the ubiquinone pool is reduced and the membrane potential is high [11].

NADPH oxidases. The NOX family (NOX1–5, DUOX1–2) comprises transmembrane flavocytochromes whose only known physiological function is the deliberate transfer of electrons from NADPH across the membrane to molecular oxygen, generating $O_2^{\cdot-}$ or, for NOX4 and the DUOX enzymes, H_2O_2 directly [10]. In contrast to mitochondrial ROS, NOX-derived oxidants are produced on demand in response to growth factors, cytokines and mechanical stimuli, and are central to host defence, vascular signalling and hormone biosynthesis [10].

Xanthine oxidoreductase. Xanthine oxidase, the oxidase form of xanthine oxidoreductase, catalyses the terminal steps of purine catabolism (hypoxanthine $\rightarrow$ xanthine $\rightarrow$ uric acid) with concomitant reduction of O_2 to

$O_2^{\cdot-}$ and H_2O_2 . Conversion of the dehydrogenase to the oxidase form during ischaemia, followed by reoxygenation, makes this enzyme a major contributor to ischaemia–reperfusion injury [2].

Peroxisomes and endoplasmic reticulum. Peroxisomal flavin oxidases, especially acyl-CoA oxidase during fatty-acid β -oxidation, transfer electrons directly to O_2 to produce H_2O_2 ; the high catalase content of this organelle normally prevents its escape [12]. In the endoplasmic reticulum, oxidative protein folding by endoplasmic reticulum oxidoreductin 1 (ERO1) working in concert with protein disulfide isomerase (PDI) releases one molecule of H_2O_2 for every disulfide bond formed, so that secretory cells with a high folding load are exposed to a substantial oxidative burden [12].

2.4. Exogenous sources

Ultraviolet and ionising radiation, cigarette smoke, air pollutants, heavy metals, organic solvents, some drugs (e.g. anthracyclines, paracetamol overdose) and excessive alcohol intake increase the cellular ROS load either by direct radical formation, by redox cycling or by depleting glutathione and other reductants [1,2]. Dietary factors such as high intake of pro-oxidant iron or a diet poor in antioxidant micronutrients further lower the threshold at which endogenous production becomes damaging [1].

3. OXIDATIVE DAMAGE TO CELLULAR MACROMOLECULES

3.1. Lipid peroxidation

Polyunsaturated fatty acids (PUFAs) in membrane phospholipids and lipoproteins are the primary targets of radical attack because the bis-allylic methylene hydrogens between two double bonds have unusually low bond-dissociation energies. Lipid peroxidation proceeds as a free-radical chain reaction in three phases [13]. In **initiation**, a reactive species (typically $\cdot OH$, $LO\cdot$ or $LOO\cdot$) abstracts a bis-allylic hydrogen to form a carbon-centred lipid radical ($L\cdot$), which rearranges to a conjugated diene. In **propagation**, $L\cdot$ reacts at a near diffusion-controlled rate with O_2 to form a lipid peroxy radical ($LOO\cdot$) that abstracts a hydrogen atom from an adjacent PUFA, yielding a lipid hydroperoxide ($LOOH$) and a new $L\cdot$; a single initiating event can therefore oxidise many fatty-acid molecules. In **termination**, two radicals combine to form non-radical products, or a chain-breaking antioxidant such as α -tocopherol donates a hydrogen atom to $LOO\cdot$ and arrests the cascade.

Accumulation of $LOOH$ disorders the bilayer, increases membrane permeability and inactivates membrane-bound enzymes and receptors. Decomposition of $LOOH$ generates reactive aldehydes, principally malondialdehyde (MDA) and 4-hydroxy-2-nonenal (4-HNE), which diffuse away from the site of origin and form covalent adducts with proteins and DNA; these products are the most widely used biomarkers of oxidative damage in clinical studies [13]. In addition, iron-dependent, uncontrolled lipid peroxidation is the executioner event of ferroptosis, a regulated form of cell death that is normally restrained by glutathione peroxidase 4 [14].

3.2. Protein oxidation

Because of their abundance and rapid reaction kinetics, proteins absorb a large proportion of the total oxidant flux. The sulfur-containing residues cysteine and methionine are the most readily oxidised: cysteine thiols form sulfenic ($-SOH$), sulfinic ($-SO_2H$) and sulfonic ($-SO_3H$) acids and intra- or intermolecular disulfides, while methionine is oxidised to methionine sulfoxide [13]. Metal-catalysed oxidation of lysine, arginine, proline and threonine side chains introduces carbonyl groups (protein carbonylation), an irreversible modification that serves as a general biomarker of protein oxidation [13]. Consequences include loss of catalytic activity, misfolding, aggregation, abnormal cross-linking, backbone fragmentation and accelerated proteasomal degradation; oxidised, cross-linked proteins that resist degradation accumulate as lipofuscin in ageing tissues [13].

3.3. DNA damage

The hydroxyl radical attacks both the sugar–phosphate backbone and the nitrogenous bases of nuclear and mitochondrial DNA. Guanine, having the lowest oxidation potential of the four bases, is the preferred target, and its oxidation to 8-oxo-7,8-dihydro-2'-deoxyguanosine (8-oxodG, also termed 8-OHdG) is the most abundant and best characterised lesion [15]. If not removed by base-excision repair (OGG1 and MUTYH), 8-oxodG mispairs with adenine and generates G:C $\rightarrow$ T:A transversions; together with single- and double-strand breaks and abasic sites, these lesions disrupt cell-cycle control and contribute to mutagenesis and carcinogenesis [15]. Mitochondrial DNA is particularly vulnerable because of its proximity to the respiratory chain, its lack of protective histones and its limited repair capacity [15].

4. THE ENDOGENOUS ANTIOXIDANT DEFENCE NETWORK

Aerobic cells maintain a multilayered defence in which enzymatic catalysts and low-molecular-weight reductants work in a compartment-specific and interdependent manner (Figure (1); Table (1)). The enzymatic system removes the primary oxidants $O_2^{\cdot-}$ and H_2O_2 before they can generate $\cdot OH$, whereas non-enzymatic antioxidants intercept radicals that escape enzymatic control and regenerate one another in coupled redox cycles [2].

4.1. Enzymatic antioxidants

4.1.1. Superoxide dismutases (SOD; EC 1.15.1.1)

The SODs, first described by McCord and Fridovich in 1969 [16], catalyse the dismutation of superoxide ($2 O_2^{\cdot-} + 2 H^+ \rightarrow H_2O_2 + O_2$) by cyclic reduction and re-oxidation of a transition-metal ion at the active site, accelerating a reaction that is already fast by about four orders of magnitude. Three isoforms exist in mammals, distinguished by metal cofactor and localisation [16]: **SOD1 (Cu/Zn-SOD)** is a homodimer located in the cytosol, nucleus and mitochondrial intermembrane space; copper is the catalytic ion, while zinc stabilises the protein structure. **SOD2 (Mn-SOD)** is a homotetramer confined to the mitochondrial matrix, where it detoxifies superoxide generated by Complexes I and III; homozygous deletion of *Sod2* in mice is neonatally lethal, underscoring its importance. **SOD3 (EC-SOD)** is a secreted Cu/Zn tetrameric glycoprotein that binds to heparan sulfate proteoglycans of the extracellular matrix and vascular wall, where it preserves the bioavailability of nitric oxide by preventing its reaction with $O_2^{\cdot-}$ [16].

4.1.2. Catalase (CAT; EC 1.11.1.6)

Catalase is a haem-containing homotetramer located mainly in peroxisomes (and in erythrocyte cytosol). It decomposes H_2O_2 in a two-step reaction ($2 H_2O_2 \rightarrow 2 H_2O + O_2$) via a high-valent iron–oxo intermediate known as Compound I, without the need for a reducing substrate [17]. Catalase has an exceptionally high turnover number but a relatively high Michaelis constant for H_2O_2 , so it is most effective at the high peroxide concentrations encountered in peroxisomes during fatty-acid oxidation or under severe oxidative stress, whereas low, signalling-range H_2O_2 is handled mainly by peroxidases [17].

4.1.3. Glutathione peroxidase/glutathione reductase system

Glutathione peroxidases (GPx1–8) are a family of enzymes, five of which in humans (GPx1–4 and GPx6) contain a catalytic selenocysteine residue. They reduce H_2O_2 and organic hydroperoxides to water or the corresponding alcohols at the expense of reduced glutathione ($H_2O_2 + 2 GSH \rightarrow 2 H_2O + GSSG$; $LOOH + 2 GSH \rightarrow LOH + H_2O + GSSG$) [14]. GPx1 is the ubiquitous cytosolic and mitochondrial isoform, whereas GPx4 uniquely reduces phospholipid hydroperoxides within membranes and is the key gatekeeper against ferroptosis [14]. The oxidised glutathione (GSSG) produced is recycled to GSH by the FAD-dependent glutathione reductase ($GSSG + NADPH + H^+ \rightarrow 2 GSH + NADP^+$); the NADPH required is supplied chiefly by the pentose phosphate pathway, which links antioxidant capacity directly to glucose metabolism [18].

4.1.4. Peroxiredoxin/thioredoxin system

Peroxiredoxins (Prx1–6) are abundant, non-selenium, cysteine-dependent peroxidases that reduce H_2O_2 , peroxynitrite and organic hydroperoxides using a highly reactive "peroxidatic" cysteine. Because of their abundance and high rate constants (10^7 – $10^8 M^{-1} s^{-1}$), peroxiredoxins remove the majority of cellular H_2O_2 and act as sensors and relays in H_2O_2 signalling [19]. The peroxidatic cysteine is oxidised to a sulfenic acid, which condenses with a "resolving" cysteine to form a disulfide; this is reduced by thioredoxin (Trx), which in turn is regenerated by the selenoprotein thioredoxin reductase (TrxR) at the expense of NADPH [19]. Hyperoxidation of the peroxidatic cysteine to sulfinic acid inactivates the enzyme and is reversed by the ATP-dependent enzyme sulfiredoxin, providing a "floodgate" mechanism that allows local H_2O_2 accumulation for signalling purposes [19].

Table (1): Principal enzymatic antioxidants in mammalian cells.

Enzyme (EC number)	Cofactor	Main localisation	Reaction catalysed	Physiological role
SOD1 (Cu/Zn-SOD) (1.15.1.1)	Cu (catalytic), Zn (structural)	Cytosol, nucleus, mitochondrial intermembrane space	$2 \text{O}_2^{\bullet-} + 2 \text{H}^+ \rightarrow \text{H}_2\text{O}_2 + \text{O}_2$	Removal of cytosolic superoxide; mutations cause familial ALS
SOD2 (Mn-SOD) (1.15.1.1)	Mn	Mitochondrial matrix	$2 \text{O}_2^{\bullet-} + 2 \text{H}^+ \rightarrow \text{H}_2\text{O}_2 + \text{O}_2$	Detoxification of respiratory-chain superoxide; essential for survival
SOD3 (EC-SOD) (1.15.1.1)	Cu, Zn	Extracellular matrix, vascular wall	$2 \text{O}_2^{\bullet-} + 2 \text{H}^+ \rightarrow \text{H}_2\text{O}_2 + \text{O}_2$	Preservation of nitric oxide bioavailability in vessels
Catalase (1.11.1.6)	Haem iron (Fe^{3+})	Peroxisomes; erythrocyte cytosol	$2 \text{H}_2\text{O}_2 \rightarrow 2 \text{H}_2\text{O} + \text{O}_2$	Removal of high H_2O_2 concentrations; no reductant required
Glutathione peroxidases GPx1–4, 6 (1.11.1.9)	Selenocysteine	Cytosol, mitochondria (GPx1); membranes (GPx4); plasma (GPx3)	H_2O_2 (or LOOH) + 2 GSH $\rightarrow$ 2 H_2O (or $\text{LOH} + \text{H}_2\text{O}$) + GSSG	Reduction of low H_2O_2 and lipid hydroperoxides; GPx4 prevents ferroptosis
Glutathione reductase (1.8.1.7)	FAD; NADPH	Cytosol, mitochondria	$\text{GSSG} + \text{NADPH} + \text{H}^+ \rightarrow 2 \text{GSH}$ + NADP^+	Regeneration of GSH; links antioxidant capacity to pentose phosphate pathway
Peroxiredoxins Prx1–6 (1.11.1.24)	Peroxidatic cysteine	Cytosol, mitochondria, ER, peroxisomes, plasma	$\text{H}_2\text{O}_2 + \text{Prx}(\text{SH})_2 \rightarrow 2 \text{H}_2\text{O} +$ $\text{Prx}(\text{S}-\text{S})$	Removal of most cellular H_2O_2 ; sensor/relay in redox signalling
Thioredoxin reductase (1.8.1.9)	Selenocysteine; FAD; NADPH	Cytosol (TrxR1), mitochondria (TrxR2)	$\text{Trx}(\text{S}-\text{S}) + \text{NADPH} + \text{H}^+ \rightarrow$ $\text{Trx}(\text{SH})_2 + \text{NADP}^+$	Regeneration of thioredoxin, which recycles peroxiredoxins

4.2. Non-enzymatic antioxidants

4.2.1. Lipid-soluble antioxidants

Vitamin E. The tocopherols and tocotrienols, of which RRR- α -tocopherol is preferentially retained in humans, are the principal chain-breaking antioxidants of biological membranes and lipoproteins. α -Tocopherol donates the phenolic hydrogen of its chromanol ring to $\text{LOO}^\bullet$ (rate constant $\approx 3 \times 10^6 \text{ M}^{-1} \text{ s}^{-1}$), producing LOOH and the resonance-stabilised α -tocopheroxyl radical, which is too unreactive to propagate the chain and is reduced back to tocopherol by ascorbate or ubiquinol at the membrane–water interface [20]. Each tocopherol molecule can terminate two peroxyl radicals and protects roughly a thousand phospholipid molecules.

Carotenoids. β -Carotene, lycopene, lutein and zeaxanthin are polyene pigments whose extended conjugated double-bond systems allow them to quench singlet oxygen physically by energy transfer (lycopene being the most efficient) and to scavenge peroxyl radicals, particularly at the low oxygen partial pressures found in most tissues; at high oxygen tension carotenoid-derived peroxyl radicals can behave as pro-oxidants [20].

4.2.2. Water-soluble antioxidants

Vitamin C (ascorbic acid). Ascorbate is the major water-soluble antioxidant of plasma and cytosol. Its enediol group makes it an excellent one-electron donor: it reacts with $\text{O}_2^{\bullet-}$, $\bullet\text{OH}$, HOCl , peroxyl radicals and singlet oxygen, and it regenerates α -tocopherol from the tocopheroxyl radical, thereby linking the aqueous and lipid antioxidant systems. The resulting ascorbyl radical is comparatively unreactive and is recycled to ascorbate by NADH- and glutathione-dependent reductases [21]. Ascorbate is also a cofactor for prolyl and lysyl hydroxylases and for the HIF prolyl hydroxylases, functions that are independent of radical scavenging [21].

Glutathione. The tripeptide γ -L-glutamyl-L-cysteinylglycine (GSH) is the most abundant low-molecular-weight thiol in eukaryotic cells (1–10 mM). Its nucleophilic thiol group scavenges radicals directly, conjugates electrophiles (including 4-HNE) through glutathione S-transferases, serves as the obligate electron donor for the glutathione peroxidases and glutaredoxins, and buffers protein thiols through reversible S-glutathionylation. The GSH/GSSG ratio, normally above 100:1, is a widely used index of cellular redox status [18]. GSH synthesis is rate-limited by glutamate–cysteine ligase, a transcriptional target of Nrf2, and by cysteine availability, which is the rationale for the clinical use of N-acetylcysteine [18].

Other endogenous reductants. Uric acid accounts for a large share of plasma radical-scavenging capacity, ubiquinol (reduced coenzyme Q₁₀) recycles tocopherol in mitochondrial and plasma-membrane lipids, and lipoic acid/dihydrolipoate, melatonin and bilirubin contribute in specific compartments [2].

4.2.3. Trace-element cofactors

Several micronutrients act not as radical scavengers but as essential catalytic or structural components of antioxidant metalloenzymes. **Selenium** is incorporated as selenocysteine into the GPx and TrxR families, so that dietary selenium status limits peroxide-removing capacity [14]. **Copper** is the catalytic ion and **zinc** the structural ion of SOD1 and SOD3, and zinc additionally stabilises protein thiols and competes with redox-active metals at binding sites [16]. **Manganese** cycles between Mn³⁺ and Mn²⁺ at the active site of SOD2, and **iron** in the haem prosthetic group of catalase forms the Compound I intermediate that decomposes H₂O₂ [16,17]. Deficiency of any of these elements lowers enzymatic antioxidant capacity, whereas excess free iron and copper are dangerous because they catalyse Fenton chemistry; their sequestration by ferritin, transferrin and ceruloplasmin is itself a preventive antioxidant mechanism [2].

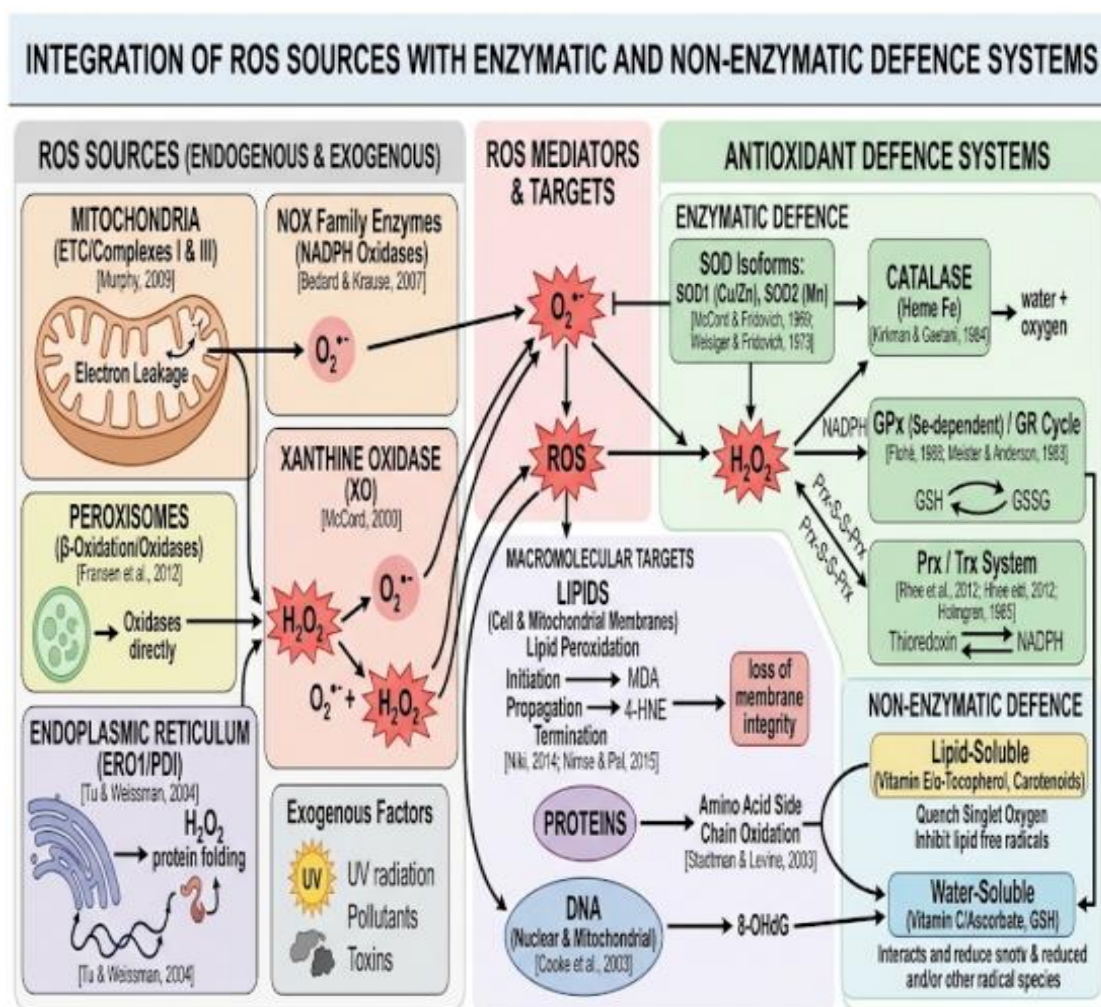


Figure (1): Integration of endogenous and exogenous sources of reactive oxygen species with the enzymatic and non-enzymatic antioxidant defence systems and their macromolecular targets. [Authors: this figure must be either an original drawing or reproduced with permission and full citation of its source. Please confirm before submission.]

5. NATURAL EXOGENOUS ANTIOXIDANTS: CHEMISTRY AND MECHANISMS OF ACTION

5.1. Chemical classes

Dietary plants contain more than 8000 phenolic compounds that share one or more aromatic rings bearing hydroxyl groups. The major classes are the phenolic acids (hydroxybenzoic acids such as gallic acid; hydroxycinnamic acids such as caffeic, ferulic and chlorogenic acids), the flavonoids (flavonols such as quercetin and kaempferol; flavan-3-ols such as catechin and epigallocatechin gallate, EGCG; flavones, flavanones, isoflavones and anthocyanidins), stilbenes (resveratrol), lignans and curcuminoids (curcumin) [7,22]. Their

antioxidant potency depends on structure: an ortho-dihydroxy (catechol) arrangement on the B-ring, a 2,3-double bond conjugated with a 4-oxo function on the C-ring, and hydroxyl groups at positions 3 and 5 confer the highest radical-scavenging activity, as exemplified by quercetin [22]. Glycosylation generally reduces activity in vitro but affects absorption in vivo (Section 8). The main food sources, mechanisms and bioavailability limitations of representative compounds are summarised in Table (2).

5.2. Direct radical-scavenging mechanisms

Phenolic antioxidants (ArOH) neutralise free radicals ($R^\bullet$) through three principal, thermodynamically distinct pathways whose relative importance depends on the solvent, pH and the radical involved [23]. In **hydrogen atom transfer (HAT)**, the phenol donates the hydroxyl hydrogen atom in a single step ($\text{ArOH} + R^\bullet \rightarrow \text{ArO}^\bullet + \text{RH}$); this pathway dominates in non-polar media and is governed by the O–H bond-dissociation enthalpy. In **single electron transfer followed by proton transfer (SET-PT)**, the phenol first transfers an electron to form a radical cation ($\text{ArOH} + R^\bullet \rightarrow \text{ArOH}^{+\bullet} + R^-$), which then deprotonates; this route is controlled by the ionisation potential and is favoured in polar solvents that stabilise charged intermediates. In **sequential proton-loss electron transfer (SPLET)**, the phenol first ionises to the phenolate anion ($\text{ArOH} \rightarrow \text{ArO}^- + \text{H}^+$), which then donates an electron to the radical ($\text{ArO}^- + R^\bullet \rightarrow \text{ArO}^\bullet + R^-$); SPLET becomes important in aqueous media at physiological pH and explains the strong pH dependence of many polyphenol activities [23]. In all three cases the resulting aryloxy radical ($\text{ArO}^\bullet$) is stabilised by resonance delocalisation over the aromatic system and by intramolecular hydrogen bonding in catechol moieties, and is too unreactive to initiate new chains. Common in vitro assays reflect these mechanisms differently: ORAC measures predominantly HAT, whereas DPPH, ABTS and FRAP assays are dominated by electron transfer [23].

5.3. Transition-metal chelation

Many polyphenols possess catechol, galloyl or 3-hydroxy-4-keto groups that chelate $\text{Fe}^{2+}/\text{Fe}^{3+}$ and $\text{Cu}^+/\text{Cu}^{2+}$. Chelation lowers the redox potential of the metal, keeps it in a form that cannot participate in Fenton and Haber–Weiss reactions, and thereby prevents the generation of $^\bullet\text{OH}$ at its source; this "preventive" antioxidant action is often more relevant in vivo than direct scavenging, given the low plasma concentrations that most polyphenols reach [23]. Quercetin, EGCG, curcumin and the anthocyanins are effective chelators, and the same property underlies the well-documented inhibition of non-haem iron absorption by tea and coffee polyphenols [23].

5.4. Indirect antioxidant action through redox-sensitive signalling

The concentrations of polyphenols and their metabolites in human plasma (usually $< 1 \mu\text{M}$) are far below those of endogenous reductants such as ascorbate ($50\text{--}100 \mu\text{M}$) and GSH (millimolar), which makes it unlikely that direct scavenging explains their effects in vivo. Instead, current evidence indicates that these compounds act mainly as electrophilic or pro-electrophilic signalling molecules that upregulate the cell's own cytoprotective machinery [8,9]. Three interconnected pathways are central (Section 6): activation of the KEAP1–Nrf2–ARE system, inhibition of NF- κ B-dependent inflammatory gene expression, and modulation of the MAPK and PI3K/Akt cascades. This indirect, "hormetic" mode of action reconciles the low tissue concentrations of polyphenols with their measurable biological effects and explains why the benefit of dietary phytochemicals is not reproduced by high doses of simple scavengers such as vitamin E [8,9].

Table (2): Representative natural antioxidants: sources, mechanisms and principal bioavailability limitations.

Compound (class)	Main dietary sources	Principal antioxidant mechanisms	Oral bioavailability limitation	Ref.
Quercetin (flavonol)	Onion, apple, capers, tea, berries	HAT/SPLET scavenging (catechol B-ring, 3-OH, 4-oxo); Fe/Cu chelation; Nrf2 activation; NF-κB inhibition	Aglycone insoluble; glycosides need hydrolysis; extensive glucuronidation/sulfation/methylation; ~2% as free aglycone	[7,22]
Epigallocatechin gallate (flavan-3-ol)	Green tea	Galloyl and pyrogallol groups: SET/HAT; metal chelation; Nrf2 activation; at high dose pro-oxidant H ₂ O ₂ formation	Unstable at intestinal pH; efflux by MRP2; bioavailability < 1%; hepatotoxicity reported at high doses	[7]
Curcumin (curcuminoid)	Turmeric (<i>Curcuma longa</i>)	Phenolic OH and β-diketone: HAT; metal chelation; KEAP1 C151 modification → Nrf2; IKK/NF-κB inhibition	Water solubility ~11 ng/mL; degrades at neutral pH; rapid glucuronidation; plasma < 50 ng/mL after 8 g oral dose	[24]
Resveratrol (stilbene)	Grapes, red wine, peanuts	SET/HAT (4'-OH most active); SIRT1 and Nrf2 activation; anti-inflammatory	≥70% absorbed but sulfated/glucuronidated almost completely; free resveratrol barely detectable in plasma	[25]
Anthocyanins (flavonoid)	Berries, red cabbage, black rice	SET/HAT (catechol B-ring in cyanidin/delphinidin); metal chelation	pH-dependent instability; < 1% recovered in urine; extensive colonic catabolism to phenolic acids	[7]
Lycopene / β-carotene (carotenoid)	Tomato, watermelon / carrot, sweet potato	Singlet-oxygen quenching; peroxy scavenging at low pO ₂	Requires fat and micellar solubilisation; absorption 10–30%; matrix-dependent (heat processing increases lycopene release)	[20]
α-Tocopherol (vitamin E)	Vegetable oils, nuts, seeds	Chain-breaking HAT to LOO•; regenerated by ascorbate/ubiquinol	Requires bile and chylomicron transport; hepatic α-TTP selects RRR-isomer; synthetic all-rac forms have lower bioactivity	[20]
Ascorbic acid (vitamin C)	Citrus, peppers, kiwi, broccoli	SET to O ₂ • ⁻ , •OH, LOO•; tocopherol regeneration; enzyme cofactor	Saturable transport (SVCT1); plasma plateau ~70–80 μM at ≥ 200 mg/day; excess excreted	[21]

6. REDOX-SENSITIVE SIGNALLING PATHWAYS

Reactive species, particularly H₂O₂, function as molecular switches through reversible, site-specific oxidation of low-pK_a cysteine residues in sensor proteins [2]. Three pathways are especially relevant to antioxidant pharmacology.

KEAP1–Nrf2–ARE. Under basal conditions the transcription factor Nrf2 (nuclear factor erythroid 2-related factor 2) is bound in the cytosol by Kelch-like ECH-associated protein 1 (KEAP1), an adaptor for the Cullin-3 E3 ubiquitin ligase that targets Nrf2 for continuous proteasomal degradation (half-life ≈ 20 min). Oxidation or electrophilic modification of specific KEAP1 cysteines—C151 by many electrophiles including sulforaphane and curcumin, and C273/C288 by lipid-derived aldehydes and prostaglandins—alters the conformation of the complex, halts ubiquitination and allows newly synthesised Nrf2 to accumulate and translocate to the nucleus [26]. There, Nrf2 heterodimerises with small Maf proteins and binds antioxidant response elements (ARE; 5'-TGACnnnGC-3') in the promoters of more than 200 genes, including those encoding glutamate–cysteine ligase, glutathione reductase, GPx, thioredoxin, peroxiredoxins, NAD(P)H:quinone oxidoreductase 1 (NQO1), haem oxygenase-1 (HO-1) and the enzymes of NADPH generation [26]. Nrf2 activation is thus the principal mechanism by which sulforaphane, curcumin, resveratrol, EGCG, quercetin and many other phytochemicals raise the antioxidant capacity of cells in a coordinated manner [9,26].

NF-κB. Nuclear factor-κB is retained in the cytosol by inhibitory IκB proteins. Pro-inflammatory stimuli and ROS activate the IκB kinase (IKK) complex, which phosphorylates IκBα and marks it for ubiquitin-dependent degradation, freeing NF-κB (p65/p50) to enter the nucleus and drive the expression of cytokines, chemokines, adhesion molecules, cyclooxygenase-2 and inducible nitric oxide synthase [9]. ROS both activate and inhibit this pathway depending on context and cell type; oxidation of cysteine 62 in the p50 DNA-binding domain, for example, inhibits DNA binding, whereas ROS-mediated activation of IKK is pro-inflammatory [9]. Polyphenols such as curcumin and EGCG suppress NF-κB activity by inhibiting IKK and by reducing the ROS that sustain its activation, providing a mechanistic link between antioxidant and anti-inflammatory effects [9].

MAPK and phosphatase regulation. H_2O_2 reversibly oxidises the catalytic cysteine of protein tyrosine phosphatases (PTP1B, SHP-2) and of the lipid phosphatase PTEN to sulfenic acid, transiently inactivating them and thereby sustaining the phosphorylation-dependent activity of receptor tyrosine kinases, the Ras–Raf–MEK–ERK cascade and the PI3K/Akt survival pathway [9]. Sustained or excessive ROS instead activate the stress-responsive JNK and p38 MAPKs, partly through oxidation of thioredoxin and release of apoptosis signal-regulating kinase 1 (ASK1), leading to apoptosis [9]. The balance between these outputs determines whether a given ROS flux is mitogenic, adaptive or lethal, and it is this balance that non-selective, high-dose antioxidants disturb.

7. OXIDATIVE STRESS IN CHRONIC DISEASE AND THE ANTIOXIDANT PARADOX

7.1. Cardiovascular disease

Oxidative modification of low-density lipoprotein (LDL) in the subendothelial space is a central event in atherogenesis. Oxidised LDL (oxLDL) activates endothelial cells to express adhesion molecules (VCAM-1, ICAM-1) and monocyte chemoattractant protein-1, recruits monocytes, and is taken up through scavenger receptors (CD36, SR-A) by the macrophages into which they differentiate, producing the lipid-laden foam cells of the fatty streak [27]. Superoxide generated by NOX enzymes and uncoupled endothelial nitric oxide synthase reacts with $\cdot\text{NO}$ to form peroxynitrite, reducing NO bioavailability and causing endothelial dysfunction, vasoconstriction and a pro-thrombotic state [27]. Under physiological conditions, vascular EC-SOD and glutathione peroxidase limit LDL oxidation and preserve NO signalling [27]. Yet, despite this compelling mechanistic and observational rationale, high-dose vitamin E supplementation did not reduce cardiovascular events in large randomised trials such as HOPE [28], a discrepancy that highlights the difference between specific endogenous redox regulation and non-specific scavenging by exogenous compounds.

7.2. Neurodegenerative disorders

The brain is exceptionally vulnerable to oxidative damage: it accounts for about 2% of body mass but consumes around 20% of the body's oxygen, contains high concentrations of readily peroxidisable PUFAs (docosahexaenoic and arachidonic acids) and redox-active iron and copper, has relatively low catalase and GPx activity compared with peripheral organs, and consists largely of post-mitotic neurons that cannot be replaced [29]. In Alzheimer's disease, amyloid- β oligomers bound to Cu^{2+} generate H_2O_2 , lipid peroxidation products (4-HNE, acrolein) accumulate in affected cortex, and oxidative damage to glucose-metabolising enzymes contributes to the characteristic cerebral hypometabolism [29]. In Parkinson's disease, dopaminergic neurons of the substantia nigra are exposed to ROS from dopamine auto-oxidation and monoamine oxidase activity, mitochondrial Complex I deficiency and high iron content, and show reduced GSH; oxidative modification promotes α -synuclein aggregation, which further impairs mitochondrial function [29]. Nrf2 activation and iron chelation are currently more promising therapeutic strategies in these diseases than direct scavengers [29].

7.3. Cancer

Oxidative DNA damage contributes to carcinogenesis in a stage-dependent manner: 8-oxodG-driven transversions in proto-oncogenes (e.g. *KRAS*) and tumour-suppressor genes (e.g. *TP53*) promote genomic instability, whereas sublethal ROS activate MAPK/ERK, PI3K/Akt and HIF-1 α signalling that support proliferation, angiogenesis and metastasis [30]. Cancer cells characteristically maintain higher basal ROS than normal cells and compensate by upregulating Nrf2, GSH and thioredoxin systems, which makes them dependent on their antioxidant machinery and, paradoxically, sensitive to agents that push ROS beyond the lethal threshold [30]. Many effective therapies—anthracyclines, platinum compounds, arsenic trioxide, ionising radiation and inducers of ferroptosis—exploit this vulnerability [30]. Consequently, antioxidant supplementation during cancer treatment can be counterproductive: in genetically engineered mouse models, N-acetylcysteine and vitamin E accelerated the progression of lung tumours by reducing ROS, decreasing p53 activation and allowing damaged cells to proliferate [30], and similar effects promoted metastasis of melanoma [30].

7.4. Metabolic disease

Hyperglycaemia increases mitochondrial superoxide production, which activates the polyol, hexosamine, protein kinase C and advanced glycation end-product pathways that underlie diabetic microvascular complications [8]. Insulin resistance and non-alcoholic fatty liver disease are likewise associated with mitochondrial dysfunction and lipid peroxidation, and dietary polyphenols improve markers of glycaemic control and hepatic steatosis in some trials, although effects are modest and heterogeneous [7].

7.5. The antioxidant paradox: clinical evidence

Large randomised controlled trials and meta-analyses have consistently failed to confirm the benefit predicted for high-dose exogenous antioxidants, and several have revealed harm (Table (3)). In the Alpha-Tocopherol, Beta-Carotene Cancer Prevention (ATBC) study of 29,133 male smokers, 20 mg/day β -carotene for 5–8 years increased lung-cancer incidence by 18% and total mortality by 8% [4]. The Beta-Carotene and Retinol Efficacy Trial (CARET) in 18,314 smokers and asbestos-exposed workers was stopped early because the combination of 30 mg β -carotene and 25,000 IU retinyl palmitate increased lung-cancer incidence by 28% and all-cause mortality by 17% [5]. The Selenium and Vitamin E Cancer Prevention Trial (SELECT) in 35,533 healthy men found that 400 IU/day of all-rac- α -tocopheryl acetate significantly increased the risk of prostate cancer by 17% [6], and the HOPE trial found no cardiovascular benefit from 400 IU/day of natural-source vitamin E over 4.5 years [28]. Synthesising these and other studies, the Cochrane systematic review by Bjelakovic et al. (78 trials, 296,707 participants) concluded that supplementation with β -carotene and vitamin E, and possibly high-dose vitamin A, is associated with a small but significant increase in all-cause mortality, whereas vitamin C and selenium showed no effect [31].

Table (3): Key randomised trials and meta-analyses underlying the antioxidant paradox.

Trial / analysis (year)	Population	Intervention and duration	Main outcome	Ref.
ATBC (1994)	29,133 Finnish male smokers, 50–69 y	α -Tocopherol 50 mg/day and/or β -carotene 20 mg/day; 5–8 y	β -Carotene: lung-cancer incidence +18%, total mortality +8%; α -tocopherol: no effect on lung cancer	[4]
CARET (1996)	18,314 smokers, former smokers and asbestos-exposed workers	β -Carotene 30 mg + retinyl palmitate 25,000 IU/day; median 4 y (stopped early)	Lung-cancer incidence +28%; all-cause mortality +17%; cardiovascular mortality +26%	[5]
HOPE (2000)	9,541 patients $\geq$ 55 y at high cardiovascular risk	Natural-source vitamin E 400 IU/day; 4.5 y	No reduction in myocardial infarction, stroke or cardiovascular death	[28]
SELECT (2011 update)	35,533 healthy men $\geq$ 50 y (USA, Canada, Puerto Rico)	Vitamin E 400 IU/day and/or selenium 200 μ g/day; median 7 y follow-up	Vitamin E alone: prostate-cancer risk +17% (HR 1.17, P = 0.008); selenium: no effect	[6]
Cochrane meta-analysis (2012)	78 RCTs, 296,707 participants (healthy and diseased)	β -Carotene, vitamin A, vitamin C, vitamin E, selenium; single or combined	All-cause mortality increased by β -carotene and vitamin E (and possibly high-dose vitamin A); no effect of vitamin C or selenium	[31]
Ristow et al. (2009)	40 healthy young men, 4-week exercise programme	Vitamin C 1000 mg + vitamin E 400 IU/day versus none	Supplementation abolished exercise-induced improvement in insulin sensitivity and induction of SOD1, SOD2, GPx1 and PGC-1 α	[32]

7.6. Mechanistic explanations for the paradox

Several mechanisms, not mutually exclusive, account for these findings [8]. **Pro-oxidant conversion.** Chain-breaking antioxidants become pro-oxidants when the reductive network that recycles them is insufficient. The α -tocopheroxyl radical formed when tocopherol scavenges LOO $\cdot$ is normally reduced by ascorbate or ubiquinol; in their absence—within the core of an LDL particle, for example—it abstracts hydrogen from neighbouring PUFAs and propagates rather than terminates peroxidation, a phenomenon known as tocopherol-mediated peroxidation [20]. Carotenoid-derived radicals behave similarly at the high oxygen tension of the lung, which may explain the specific lung-cancer signal in smokers [20]. Similarly, ascorbate and polyphenols reduce Fe $^{3+}$ to Fe $^{2+}$ and thereby fuel Fenton chemistry when free iron is available [2].

Suppression of mitohormesis and adaptive signalling. Transient increases in mitochondrial ROS induced by exercise, caloric restriction or mild metabolic stress trigger Nrf2-, PGC-1 α - and FOXO-dependent programmes that raise endogenous antioxidant capacity, improve insulin sensitivity and extend lifespan in model organisms; this adaptive response has been termed mitohormesis [32]. In a landmark human study, 1000 mg vitamin C plus 400 IU vitamin E daily abolished the exercise-induced improvement in insulin sensitivity and the upregulation of SOD1, SOD2 and GPx1 in skeletal muscle [32]. Non-selective scavenging therefore removes the very signal that makes cells more resistant, and may blunt immune surveillance and the ROS-dependent apoptosis of pre-malignant cells [8,30].

Pharmacological and trial-design factors. Synthetic all-rac- α -tocopherol and single isolated compounds do not reproduce the mixture of isomers and co-factors present in food; supplements were often given to populations with adequate baseline status, at doses far above nutritional requirements, and for durations too short to influence diseases that develop over decades [8]. Most importantly, the biomarkers used to select patients rarely confirmed that oxidative stress was present, so many participants could not have benefited from its correction [8]. Taken together, the paradox is less paradoxical than it first appeared: it reflects a misunderstanding of ROS as purely harmful and of antioxidants as inert scavengers [8].

8. BIOAVAILABILITY OF NATURAL ANTIOXIDANTS

8.1. Determinants and barriers

Bioavailability—the fraction of an ingested compound that reaches the systemic circulation in an active form—is the main obstacle to the clinical use of natural antioxidants. Most polyphenols are bioavailable to only 1–10%, and their metabolites rather than the parent compounds are what circulates [7]. The barriers are sequential (Figure (2)). **Release and solubility.** Compounds bound to the plant matrix must be liberated during digestion and must dissolve in intestinal fluid; curcumin, resveratrol and quercetin aglycone are practically insoluble in water (curcumin ≈ 11 ng/mL) and dissolve slowly [24]. **Chemical instability.** Curcumin degrades within minutes at neutral–alkaline pH, anthocyanins are unstable above pH 4, and EGCG oxidises readily in the small intestine [7,24]. **Absorption.** Only aglycones and some glucosides cross the enterocyte membrane; most flavonoid glycosides must first be hydrolysed by lactase-phlorizin hydrolase at the brush border or by cytosolic β -glucosidase, whereas rutinoides and larger conjugates are not absorbed in the small intestine and reach the colon [7]. **First-pass metabolism.** Absorbed aglycones are rapidly conjugated by uridine 5'-diphosphoglucuronosyltransferases, sulfotransferases and catechol-O-methyltransferase in the enterocyte and liver; in a human study, oral ^{14}C -resveratrol was absorbed to at least 70% but the free compound was almost undetectable in plasma because of extensive sulfation and glucuronidation [25]. **Efflux transport.** P-glycoprotein, multidrug-resistance-associated protein 2 and breast-cancer resistance protein pump conjugated metabolites back into the gut lumen [7]. **Colonic metabolism.** Unabsorbed polyphenols are catabolised by the gut microbiota to small phenolic acids (e.g. protocatechuic, phenylacetic and hydroxyphenylpropionic acids, urolithins from ellagitannins, equol from daidzein) that are well absorbed and may account for a large part of the biological activity attributed to the parent compounds [7]. **Elimination.** Conjugates are cleared rapidly by renal and biliary excretion, so that plasma half-lives are generally short (1–3 h) [7].

8.2. Consequences for interpretation of in vitro data

These pharmacokinetic realities imply that concentrations used in most cell-culture studies (10–100 μM of parent aglycone) are not achievable in vivo, that the relevant bioactive species are usually glucuronidated, sulfated or methylated metabolites and microbial catabolites with different properties, and that inter-individual variation in microbiota composition ("metabotypes") contributes to the heterogeneity of clinical responses [7]. Improving bioavailability is therefore a prerequisite for translating in vitro efficacy into clinical benefit.

8.3. Conventional strategies

Simple approaches include co-administration with bioenhancers—piperine, an inhibitor of glucuronidation, increased the bioavailability of curcumin in humans by about 2000% [24]—consumption with dietary fat to promote micellar solubilisation, use of glycoside or ester prodrugs, and structural modification such as methylation to reduce conjugation [24]. These measures offer moderate gains and are limited by the intrinsic solubility and stability of the compounds, which has stimulated the development of nano-scale delivery systems.

9. NANO-DELIVERY SYSTEMS FOR NATURAL ANTIOXIDANTS

Nanocarriers (typically 10–500 nm) address several barriers simultaneously: they solubilise lipophilic payloads, protect them from pH-, enzyme- and oxygen-dependent degradation, present a large surface area for dissolution, promote uptake through the lymphatic route (which bypasses hepatic first-pass metabolism) and can be surface-modified for mucoadhesion, prolonged circulation or targeting [33]. The principal platforms are compared in Table (4).

Liposomes and phospholipid complexes. Liposomes are phospholipid bilayer vesicles that accommodate lipophilic compounds within the bilayer and hydrophilic compounds in the aqueous core. They are biocompatible and biodegradable but are prone to bile-salt-induced disruption and lipase digestion in the gut, which limits oral use unless the bilayer is stabilised with cholesterol, surface-coated with chitosan or polyethylene glycol, or

reformulated as phytosomes [34]. Phytosomes (phospholipid complexes) formed by stoichiometric complexation of polyphenols with phosphatidylcholine markedly increase lipophilicity and membrane permeation; a curcumin–phosphatidylcholine complex (Meriva) yielded 29-fold higher total curcuminoid absorption in human volunteers than unformulated curcumin [34].

Solid lipid nanoparticles (SLN) and nanostructured lipid carriers (NLC). SLN consist of a solid lipid matrix (triglycerides, fatty acids, waxes) stabilised by surfactants; NLC blend solid and liquid lipids to create imperfections that increase loading capacity and reduce drug expulsion during storage [33]. Both protect polyphenols from oxidation and gastric degradation, undergo lipase-mediated digestion to form mixed micelles that facilitate absorption, and enhance lymphatic uptake. Curcumin-SLN and resveratrol-NLC formulations have increased oral bioavailability in rodents by 10- to 40-fold [33].

Polymeric nanoparticles. Biodegradable polymers such as poly(lactic-co-glycolic acid) (PLGA), poly(ϵ -caprolactone), chitosan and food-grade proteins (zein, casein, whey) encapsulate antioxidants by nanoprecipitation, emulsification–evaporation or electrospraying, and release them in a controlled or pH-triggered manner [33]. Chitosan nanoparticles are mucoadhesive and open tight junctions transiently, favouring paracellular transport; PLGA-curcumin nanoparticles increased oral bioavailability up to 22-fold in rats relative to curcumin suspension [33].

Nanoemulsions and self-emulsifying systems. Oil-in-water nanoemulsions (droplets 20–200 nm) and self-nanoemulsifying drug delivery systems (SNEDDS) that form spontaneously on contact with gastrointestinal fluids increase the dissolution rate and micellar solubilisation of lipophilic compounds. Theracurmin, a colloidal submicron dispersion of curcumin, achieved a 27-fold higher area under the plasma concentration curve than curcumin powder in humans [35].

Cyclodextrin inclusion complexes. Cyclodextrins are cyclic oligosaccharides with a hydrophilic exterior and a hydrophobic cavity that accommodates poorly soluble guest molecules, increasing their apparent solubility and stability without covalent modification. Hydroxypropyl- β -cyclodextrin and sulfobutylether- β -cyclodextrin are pharmaceutically approved and have been applied to curcumin, quercetin, resveratrol and carotenoids [33].

Emerging platforms. Nanocrystals (pure drug nanoparticles stabilised by surfactants), dendrimers, mesoporous silica, metal–organic frameworks, exosome-based carriers and inorganic nanoparticles with intrinsic catalytic ("nanozyme") antioxidant activity such as cerium oxide are under investigation, as are stimulus-responsive systems that release their cargo in response to the elevated ROS of inflamed or tumour tissue [33].

Table (4): Nano-delivery platforms for natural antioxidants.

Platform (typical size)	Composition	Advantages	Limitations	Representative examples	Ref.
Liposomes / phytosomes (50–500 nm)	Phospholipid bilayer $\pm$ cholesterol; polyphenol–phosphatidylcholine complex	Biocompatible; carry both lipophilic and hydrophilic cargo; phytosomes enhance membrane permeation	Bile-salt and lipase instability; leakage; cost; limited shelf life	Curcumin–phosphatidylcholine (Meriva): 29-fold $\uparrow$ absorption in humans; quercetin and silybin phytosomes	[34]
Solid lipid nanoparticles / NLC (50–1000 nm)	Solid ($\pm$ liquid) lipid core, surfactant shell	Protection from degradation; lymphatic uptake bypasses first pass; scalable (high-pressure homogenisation)	Limited loading (SLN); drug expulsion on storage; polymorphic transitions	Curcumin-SLN, resveratrol-NLC: 10–40-fold $\uparrow$ oral bioavailability in rodents	[33]
Polymeric nanoparticles (100–300 nm)	PLGA, PCL, chitosan, zein, casein, alginate	Controlled/pH-triggered release; mucoadhesion (chitosan); surface functionalisation for targeting	Organic solvents in preparation; burst release; polymer cost	PLGA-curcumin: up to 22-fold $\uparrow$ bioavailability in rats; chitosan-EGCG	[33]
Nanoemulsions / SNEDDS (20–200 nm)	Oil, surfactant, co-surfactant	Rapid dissolution; high solubilisation; easy oral dosing	Surfactant toxicity at high load; thermodynamic instability (nanoemulsions)	Theracurmin (colloidal curcumin): 27-fold $\uparrow$ AUC in humans	[35]
Cyclodextrin inclusion complexes (1–2 nm cavity)	α -, β -, γ -cyclodextrins and derivatives (HP- β -CD, SBE- β -CD)	Solubility and stability $\uparrow$ without covalent change; regulatory acceptance; masks taste	Limited to guests fitting the cavity; 1:1 stoichiometry restricts loading	Curcumin-, quercetin-, resveratrol-HP- β -CD complexes	[33]
Nanocrystals, nanozymes, ROS-responsive carriers	Pure drug nanocrystals; CeO ₂ or Mn ₃ O ₄ nanoparticles; thioketal/boronate polymers	High drug loading; intrinsic catalytic SOD/CAT-like activity; release at inflamed/tumour sites	Long-term toxicity of inorganic particles unclear; complex synthesis; limited clinical data	Cerium oxide nanozymes; ROS-triggered curcumin release in colitis models	[33]

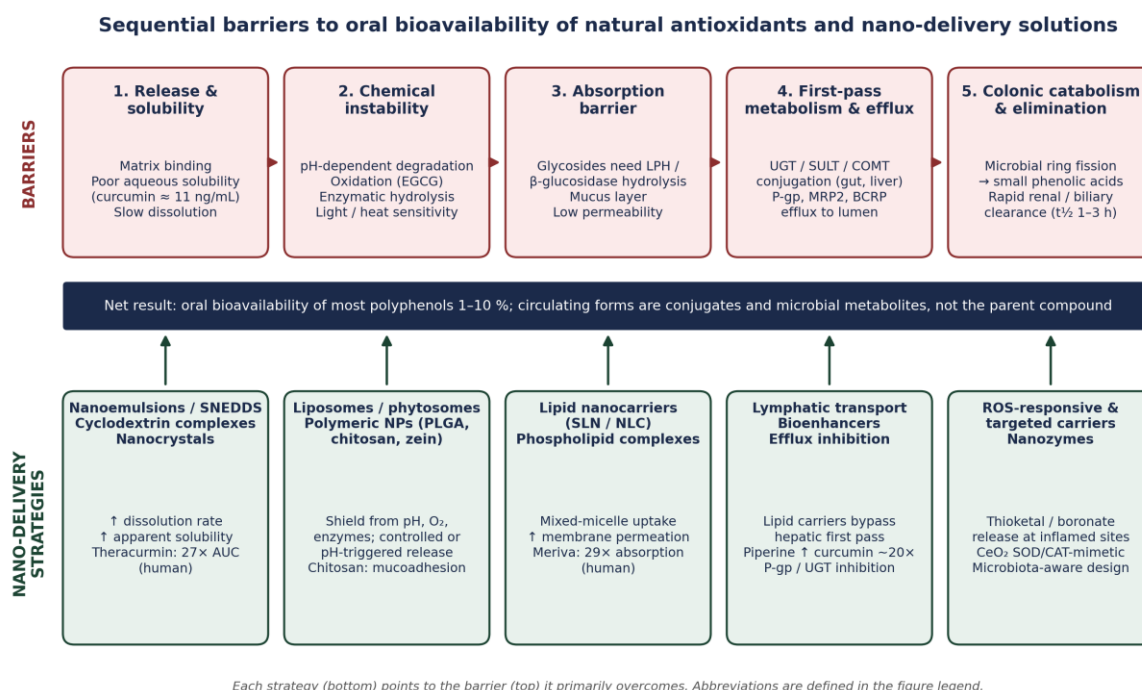


Figure (2): Sequential barriers to the oral bioavailability of natural antioxidants and the corresponding nano-delivery strategies developed to overcome them. Abbreviations: LPH, lactase-phlorizin hydrolase; UGT, UDP-glucuronosyltransferase; SULT, sulfotransferase; COMT, catechol-O-methyltransferase; P-gp, P-glycoprotein; MRP2, multidrug-resistance-associated protein 2; SLN, solid lipid nanoparticles; NLC, nanostructured lipid carriers; SNEDDS, self-nanoemulsifying drug delivery systems.

9.1. Safety, standardisation and regulatory considerations

Nano-formulation changes the pharmacokinetics and tissue distribution of a compound, so the safety profile of the encapsulated product cannot be assumed from that of the free compound; polyphenols delivered at concentrations that were previously unattainable may exert pro-oxidant, enzyme-inhibitory or drug-interaction effects (for example, inhibition of cytochrome P450 3A4 and P-glycoprotein by curcumin and quercetin) [8,24]. Carrier materials must be biodegradable and non-immunogenic, and the long-term fate of inorganic and non-degradable nanoparticles requires evaluation [8]. Batch-to-batch reproducibility, scalability, storage stability, validated methods for measuring encapsulation efficiency and release, and clear regulatory definitions of "nano" ingredients in foods and supplements remain practical obstacles. Most importantly, the majority of bioavailability data come from rodent studies; randomised, adequately powered human trials with clinical rather than surrogate end-points are still scarce [33].

10. FUTURE PERSPECTIVES

Several directions are likely to shape the next decade of antioxidant research. First, the field is moving from "antioxidant" to "redox-modulating" therapy: pharmacological Nrf2 activators (dimethyl fumarate is approved for multiple sclerosis; sulforaphane, bardoxolone methyl and omaveloxolone are in advanced clinical evaluation), mitochondria-targeted antioxidants (MitoQ, SkQ1), NOX inhibitors and ferroptosis inhibitors target specific sources or sensors of ROS rather than scavenging all of them [8,26]. Second, patient selection using validated biomarkers of oxidative damage (F₂-isoprostanes, 8-oxodG, protein carbonyls, GSH/GSSG) should identify individuals in whom oxidative stress is actually present. Third, the gut microbiota is emerging as a co-determinant of polyphenol efficacy, suggesting that prebiotic or probiotic co-administration and metabolite-based personalisation could improve outcomes [7]. Fourth, combining nano-encapsulation with targeting ligands and ROS-responsive release may deliver antioxidants preferentially to diseased tissue while sparing physiological signalling elsewhere [33]. Finally, artificial intelligence and molecular modelling are increasingly used to predict the antioxidant mechanism (HAT versus SET-PT versus SPLET), metabolism and optimal carrier design of natural compounds, accelerating rational selection for clinical testing [23].

11. CONCLUSIONS

Oxidative stress is best understood not as an excess of harmful radicals to be eliminated, but as a disturbance of a finely regulated redox signalling network. Endogenous antioxidant defence operates as a coordinated, compartment-specific system in which enzymatic catalysts, low-molecular-weight reductants and trace-element cofactors regenerate one another and are transcriptionally tuned by the KEAP1–Nrf2 axis. Natural phenolic antioxidants scavenge radicals through HAT, SET-PT and SPLET mechanisms and chelate transition metals *in vitro*, but at the low concentrations reached *in vivo* they act chiefly by hormetic activation of endogenous cytoprotective programmes. The failure of high-dose synthetic supplements in ATBC, CARET, SELECT and HOPE, and the excess mortality identified in meta-analyses, are explained by pro-oxidant conversion, disruption of mitohormesis and suppression of adaptive ROS signalling. The clinical promise of natural compounds is constrained by poor solubility, instability, extensive first-pass metabolism and efflux, obstacles that nano-encapsulation platforms—liposomes, phytosomes, lipid and polymeric nanoparticles, nanoemulsions and cyclodextrin complexes—can substantially reduce. Translation of antioxidant therapy should therefore rely on balanced, phytochemical-rich dietary patterns, biomarker-guided selection of patients, targeted redox modulators and rationally designed, well-characterised delivery systems, evaluated in adequately powered clinical trials rather than on unregulated high-dose supplementation.

DECLARATIONS

Author contributions. Z.T.O. conceived the review, drafted the sections on reactive species and antioxidant defence and prepared the figures; B.A.S. drafted the sections on cellular damage and chronic disease; Z.A.H. drafted the sections on bioavailability and nano-delivery; N.A.-I. drafted the sections on signalling pathways and the antioxidant paradox and compiled the tables. All authors revised the manuscript critically and approved the final version.

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