Antioxidants: Current Summary

İlhami Gülçin[1,2]

[1]Department of Chemistry, Atatürk University Faculty of Sciences, Erzurum, Türkiye
[2]Rectorate of Ağrı İbrahim Çeçen University, Ağrı, Türkiye

Free radicals are highly reactive and unstable species-atoms, molecules, or ions-that contain one or more unpaired electrons, enabling them to readily interact with and damage surrounding molecules. In biological systems, they are primarily generated through normal metabolic processes, such as mitochondrial respiration, or from external sources including pollution, radiation, cigarette smoke, and certain drugs. Endogenously, free radicals are produced as a normal byproduct of mitochondrial metabolism and arise from various biological processes such as xanthine oxidase activity, peroxisomal reactions, inflammatory responses, phagocytosis, arachidonic acid metabolism, ischemic events, and physical activity. While low to moderate levels of free radicals play essential roles in cell signaling and immune defense, excessive accumulation leads to oxidative stress-a condition that damages vital biomolecules including proteins, DNA, RNA, lipids, and sugars. A clear example of reactive oxygen species (ROS)-induced damage is the oxidation of nucleosides within DNA, illustrating how these reactive species can disrupt molecular structure and function. Such damage is implicated in the development of various diseases, including cancer, cardiovascular diseases, diabetes, and neurodegenerative disorders. The living metabolism employs antioxidant defense systems, encompassing both enzymatic and non-enzymatic antioxidants, to neutralize free radicals and maintain redox balance.[1]

Oxygen is deeply integrated into a variety of oxidation-reduction and enzymatic processes within living organisms. Among the most common free radicals are ROS, which include superoxide anion $O_2^{\bullet -}$, hydroxyl radical (OH•), and hydrogen peroxide (H_2O_2). External sources of ROS include tobacco smoke, organic solvents, specific pollutants, and pesticides. An elevated steady-state level of ROS results from an imbalance between their production and removal, which can significantly influence numerous biological processes.[2] Oxidative stress occurs when ROS levels exceed the body's antioxidant capacity, disrupting cellular functions and contributing to various pathological conditions. The effects of increased ROS depend on several factors, including the intensity and location of ROS generation, the efficiency of antioxidant defense systems, the availability of energy and metabolic resources, and the specific cellular components involved. While excessive ROS can be harmful, their controlled production plays an essential role in maintaining cellular homeostasis. To preserve this balance, living organisms employ a complex antioxidant defense system that regulates the interplay between oxidative stress and protective mechanisms.[3]

Antioxidants are compounds that protect cells from damage caused by reactive molecules such as free radicals. Even at very low concentrations, they can slow, control, or prevent oxidative reactions, thereby maintaining food quality and potentially reducing the onset and progression of degenerative diseases. Their role is critical in both biological systems and food preservation, as they counteract oxidative stress and neutralize the harmful effects of ROS. In food systems, antioxidants inhibit lipid peroxidation and prevent the formation of damaging secondary products such as malondialdehyde and 4-hydroxynonenal, thereby preserving flavor, color, and texture during storage. They also safeguard biomolecules by reducing protein and amino acid oxidation and preventing harmful interactions between lipid-derived carbonyls and proteins, which can impair protein function.[4]

Antioxidants are broadly defined as molecules that delay or inhibit the oxidation of other substances. In food science, an antioxidant is "any substance that, at low concentrations relative to an oxidizable substrate, significantly delays or prevents oxidation." In biochemistry, they are defined as agents that prevent, delay, or repair oxidative damage to biomolecules. An expanded definition includes substances that directly neutralize ROS or free radicals, enhance endogenous antioxidant defenses, or suppress ROS formation.[5] A distinction exists between "antioxidant activity" and "antioxidant capacity". "Antioxidant activity" refers to the effectiveness of a single compound in a specific assay, whereas "total antioxidant capacity" describes the overall antioxidant potential of a mixture or complex sample, such as an extract, beverage, or biological fluid.[6] In this context, total antioxidant capacity is used to evaluate the antioxidant performance of complex systems.

Corresponding author: İlhami Gülçin, Department of Chemistry, Atatürk University Faculty of Sciences, Erzurum, Türkiye; Rectorate of Ağrı İbrahim Çeçen University, Ağrı, Türkiye
e-mail: igulcin@atauni.edu.tr/igulcin@yahoo.com
Available Online Date: xxxxxx • **DOI:** 10.4274/balkanmedj.galenos.2025.2025.130825
Available at www.balkanmedicaljournal.org
ORCID iD of the author: İ.G. 0000-0001-5993-1668.
Cite this article as: Gülçin İ. Antioxidants. Balkan Med J.;

Antioxidants extend shelf life by scavenging free radicals and delaying lipid oxidation-a primary factor in the degradation of foods and pharmaceuticals during processing and storage. They can be categorized in several ways.[7] Based on their mode of action, antioxidants are classified as primary (chain-breaking) or secondary (preventive). From a biological perspective, they are divided into enzymatic and non-enzymatic antioxidants. In addition, synthetic antioxidants form a distinct group widely used in the food and pharmaceutical industries.[8]

Enzymatic antioxidants are generally divided into primary and secondary defense systems. While several intracellular enzymes contribute to ROS production, the primary enzymatic defense centers on three key enzymes that prevent the formation of free radicals or neutralize them: catalase (CAT), superoxide dismutase (SOD), and glutathione peroxidase (GPx). SOD catalyzes the conversion of the $O_2^{\bullet -}$ into H_2O_2, which is subsequently broken down by CAT into water and oxygen. CAT is notable for its exceptionally high turnover rate, capable of decomposing billions of H_2O_2 molecules per second. GPx reduces H_2O_2 and organic hydroperoxides using glutathione (GSH), producing water and oxidized glutathione (GSSG) and preventing the buildup of peroxides that could initiate harmful Fenton-type reactions. The secondary enzymatic defense includes enzymes such as glutathione reductase (GR) and glucose-6-phosphate dehydrogenase (G6PD). GR regenerates reduced GSH from its oxidized form (GSSG), ensuring the continuous detoxification of ROS. G6PD supports antioxidant function by producing nicotinamide adenine dinucleotide phosphate, an essential coenzyme for maintaining redox balance. Although these secondary enzymes do not directly neutralize free radicals, they are critical for sustaining the activity of primary antioxidant enzymes. Cofactors such as copper, zinc, manganese, and iron are essential for the optimal function of these antioxidant enzymes.[9]

Non-enzymatic antioxidants play a crucial role in protecting the human body from the harmful effects of ROS and free radicals, which are implicated in chronic diseases such as cardiovascular disorders, diabetes, cancer, and arthritis. Increasing attention has been given to identifying natural, safe antioxidant sources-particularly from plants-to replace synthetic additives in food. In food systems, antioxidants are added to prevent oxidative chain reactions by disrupting the initiation and propagation phases, thereby halting oxidation. These compounds are vital for extending shelf life while preserving sensory and nutritional quality. Effective food antioxidants must meet several key criteria: they should be safe at low concentrations, stable under processing conditions, odorless, tasteless, colorless, and easily incorporated into various food matrices.[7]

Phenolic compounds, characterized by one or more aromatic rings, are abundant in plant-based foods and contribute to many of their sensory attributes, including flavor and color. They range from simple monophenols, such as 3-ethylphenol and 3,4-dimethylphenol, to more complex molecules like hydroxycinnamic acids (e.g., caffeic and ferulic acids), flavonoids (e.g., catechins, anthocyanidins, flavonols), and tannins. The average daily intake of phenolic compounds may reach approximately 1 g, although the flavonoid fraction typically amounts to only a few dozen milligrams. Derived from plant secondary metabolites, phenolic compounds contribute to oxidative stability in foods by functioning as antioxidants even at low concentrations (Figure 1). Their antioxidant activity strongly depends on the number and position of hydroxyl groups in their structure. Substitutions at *ortho*- or *para*-positions increase electron density, lowering the O-H bond dissociation energy and enhancing radical-scavenging activity, whereas substitutions at the *meta*-position exert less influence. The overall effectiveness of phenolic antioxidants is determined by both electronic and steric factors.[10] The hydrogen abstraction mechanism of phenolics during the inhibition of lipid autoxidation has been investigated using molecular orbital theory, which describes electrons in terms of orbitals extending over the entire molecule. This approach offers valuable insight into the structure, reactivity, and antioxidant properties of phenolic compounds.[11]

Butylated hydroxyanisole (BHA) and butylated hydroxytoluene (BHT) are synthetic phenolic antioxidants widely used in the food, cosmetic, and pharmaceutical industries to prevent lipid oxidation and extend product shelf life. They function by donating hydrogen atoms to free radicals, thereby interrupting oxidative chain reactions. α-tocopherol, a lipid-soluble antioxidant found in plant oils, nuts, and seeds, protects cell membranes and food lipids from oxidative damage by neutralizing lipid peroxyl radicals. While BHA and BHT are cost-effective and heat-stable, α-tocopherol also provides nutritional benefits in addition to its antioxidant activity (Figure 2a).[5,8]

Natural phenolic compounds exhibit strong antioxidant activity through multiple mechanisms, including hydrogen atom or electron donation, metal ion chelation, and free radical scavenging. Their effectiveness is largely determined by the number and position of hydroxyl groups on their aromatic rings.[12] Phenolics-such as flavonoids, phenolic acids, and tannins-protect biological molecules from oxidative damage and contribute to the prevention of chronic diseases. Abundant in fruits, vegetables, teas, and medicinal plants, these compounds play an important role in both food preservation and human health.[13] Figure 2b presents the chemical structures of selected natural phenolic antioxidant with broad biological activity.

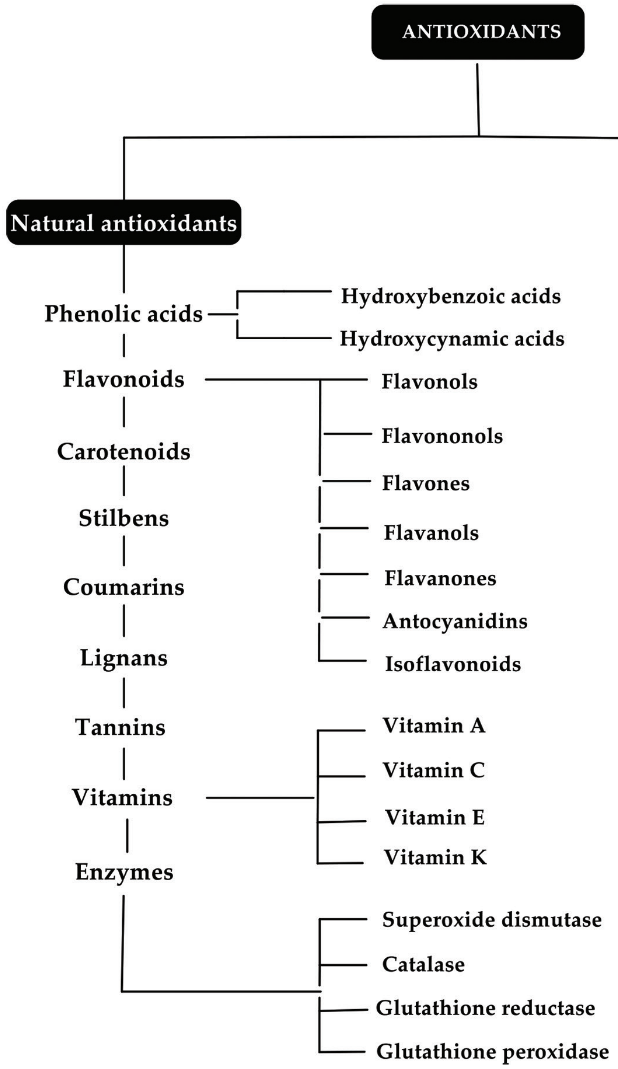

FIG. 1. Classification of antioxidant compounds (Adapted from Gülçin 2025).

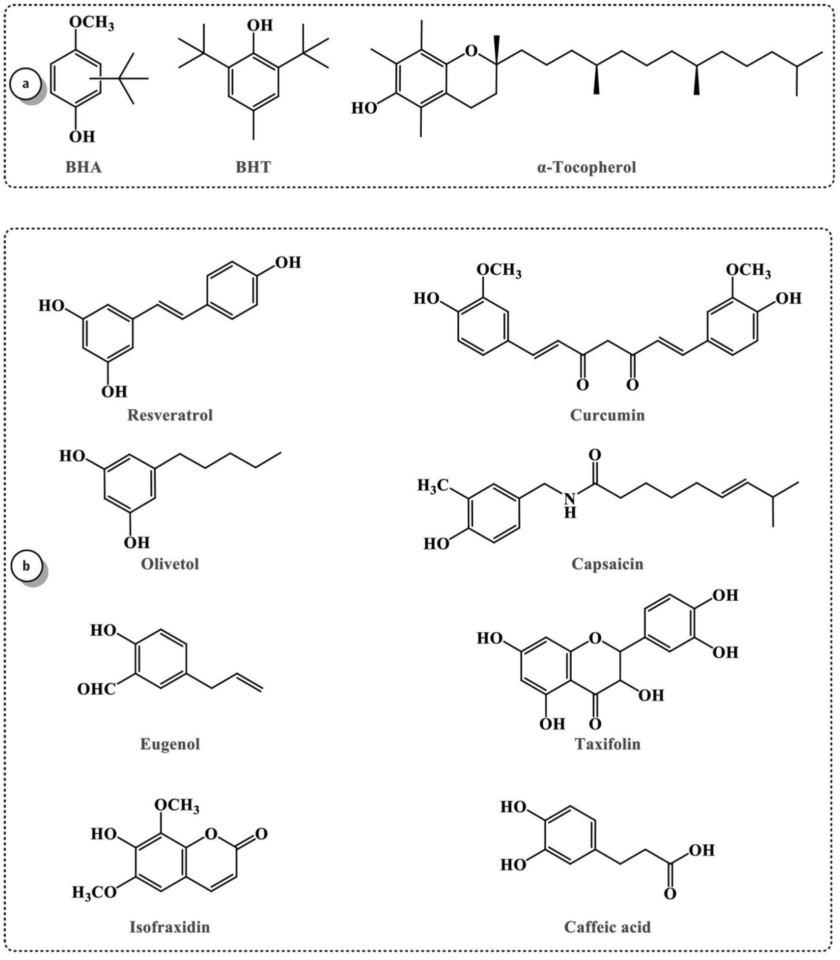

FIG. 2. Chemical structures of some standard antioxidants (a) and natural phenolic antioxidants (b).
BHA, butylated hydroxyanisole; BHT, butylated hydroxytoluene.

Among the most extensively studied natural phenolic compounds in recent years are resveratrol, a natural phytoalexin found in the fruits and leaves of several edible plants, including peanuts, mulberries, and grape[14]; capsaicin, the active component in chili peppers responsible for their pungency, widely investigated for its pain-relieving, metabolism-boosting, and anti-inflammatory effects[15]; taxifolin (dihydroquercetin), a natural flavonoid with strong antioxidant, anti-inflammatory, and cardioprotective properties, present in various fruits, vegetables, and plants[16]; caffeic acid, a naturally occurring phenolic compound found in many plants[17]; curcumin, the principal bioactive compound in turmeric, recognized for its potent anti-inflammatory, antioxidant, and potential anticancer effects[18]; rosmarinic acid, a natural polyphenolic compound found in herbs such as rosemary, basil, and sage, noted for its strong antioxidant activity[19]; and eugenol, a natural phenolic constituent of clove oil with antiseptic, analgesic, and anti-inflammatory properties.[20]

Phenolic compounds comprise a diverse group of plant-derived secondary metabolites with significant implications for human health and disease prevention. Their biological activities-including antioxidant, antimicrobial, anti-inflammatory, and anticancer effects-are largely determined by their structural features, particularly the number and arrangement of hydroxyl groups. By neutralizing ROS and modulating key biochemical pathways, phenolics help reduce the risk of chronic diseases such as cardiovascular disorders, neurodegenerative conditions, and certain cancers. Their ability to act synergistically with other bioactive molecules further enhances their therapeutic potential. Advances in analytical techniques have improved the identification and quantification of phenolic constituents in foods, herbs, and medicinal plants, enabling targeted dietary interventions and the development of functional foods. However, despite substantial *in vitro* and *in vivo* evidence, challenges remain in addressing their bioavailability, metabolism, and clinical efficacy. Future research should aim to clarify the molecular mechanisms underlying their health benefits, optimize extraction and delivery systems, and conduct well-designed clinical trials to translate laboratory findings into practical applications. Overall, phenolic compounds represent promising natural agents for promoting health and preventing disease, offering a strong foundation for the development of novel nutraceuticals and pharmaceuticals.

REFERENCES

1. Gulçin İ, Alwasel SH. DPPH radical scavenging assay. *Processes.* 2023;11:2248. [CrossRef]
2. Md Yusoff MH, Shafie MH. A review of in vitro antioxidant and antidiabetic polysaccharides: extraction methods, physicochemical and structure-activity relationships. *Int J Biol Macromol.* 2024;282:137143. [CrossRef]
3. Gulçin İ, Alwasel SH. Metal ions, metal chelators and metal chelating assay as antioxidant method. *Processes.* 2022;10:132. [CrossRef]
4. Cetinkaya Y, Göçer H, Menzek A, Gülçin I. Synthesis and antioxidant properties of (3,4-dihydroxyphenyl)(2,3,4-trihydroxyphenyl)methanone and its derivatives. *Arch Pharm (Weinheim).* 2012;345:323-334. [CrossRef]
5. Gülçin İ. Antioxidants-a comprehensive review. *Arch Toxicol.* 2025;99:1893-1997. [CrossRef]
6. Sadowska-Bartosz I, Bartosz G. Evaluation of the antioxidant capacity of food products: methods, applications and limitations. *Processes.* 2022;10:2031. [CrossRef]
7. Karagecili H, Yılmaz MA, Ertürk A, et al. Comprehensive metabolite profiling of berdav propolis using LC-MS/MS: determination of antioxidant, anticholinergic, antiglaucoma, and antidiabetic effects. *Molecules.* 2023;28:1739. [CrossRef]
8. Gülçin İ. Antioxidants and antioxidant methods-an updated overview. *Arch Toxicol.* 2020;4:651-715. [CrossRef]
9. Apak R, Calokerinos A, Gorinstein S, et al. Methods to evaluate the scavenging activity of antioxidants toward reactive oxygen and nitrogen species. *Pure Appl Chem.* 2022;94:87-144. [CrossRef]
10. Botosoa EP, Shahidi F. Phenolics and polyphenolics in mangrove plants: antioxidant activity and recent trends in food application - a review. *Crit Rev Food Sci Nutr*. 2025:1-35. [CrossRef]
11. Gülçin İ, Gören AC, Taslimi P, et al. Anticholinergic, antidiabetic and antioxidant activities of Anatolian pennyroyal (*Mentha pulegium*)-analysis of its polyphenol contents by LC-MS/MS. *Biocatal Agric Biotechnol.* 2020;23:101441. [CrossRef]
12. Aytac S, Gundogdu O, Bingol Z, Gulcin İ. Synthesis of schiff bases containing phenol rings and investigation of their antioxidant capacity, anticholinesterase, butyrylcholinesterase, and carbonic anhydrase inhibition properties. *Pharmaceutics.* 2023;15:779. [CrossRef]
13. Gülçin İ. Antioxidant activity of food constituents: an overview. *Arch Toxicol.* 2012;86:345-391. [CrossRef]
14. Gülçin İ. Antioxidant properties of resveratrol: a structure-activity insight. *Innov Food Sci Emerg.* 2010;11:210-218. [CrossRef]
15. Arabaci B, Gulcin I, Alwasel S. Capsaicin: a potent inhibitor of carbonic anhydrase isoenzymes. *Molecules.* 2014;19:10103-10114. [CrossRef]
16. Topal F, Nar M, Gocer H, et al. Antioxidant activity of taxifolin: an activity-structure relationship. *J Enzyme Inhib Med Chem.* 2016;31:674-683. [CrossRef]
17. Gülçin İ. Antioxidant activity of caffeic acid (3,4-dihydroxycinnamic acid). *Toxicology.* 2006;217:213-220. [CrossRef]
18. Ak T, Gülçin İ. Antioxidant and radical scavenging properties of curcumin. *Chem Biol Interact.* 2008;174:27-37. [CrossRef]
19. Durmaz L, Gülçin İ, Taslimi P, Tüzün B. Isofraxidin: antioxidant, anti-carbonic anhydrase, anti-cholinesterase, anti-diabetic, and in silico properties. *ChemistrySelect.* 2023;8:e202300170. [CrossRef]
20. Gülçin İ. Antioxidant activity of eugenol-a structure and activity relationship study. *J Med Food*. 2011;14:975-985. [CrossRef]