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Natural chemicals for healthy living: plant secondary metabolic compounds

Abstract

Free radicals are produced by a variety of normal biological processes including aerobic metabolism and pathogenic defense mechanisms. They can also be a result of external exposures such as radiation, pollutants, and cigarette smoke. Reactive oxygen species, or ROS, are a subset of free radicals that contain oxygen. A diet high in antioxidants may reduce the risk of many diseases (including heart disease and certain cancers). Antioxidants scavenge free radicals from the body cells and prevent or reduce the damage caused by oxidation. The protective effect of antioxidants continues to be studied around the world. During aerobic metabolism, oxidants, or reactive oxygen species, are created in our bodies, which can cause a number of illnesses, including cancer and cardiovascular conditions. The substances that balance these oxidants called antioxidants. Natural antioxidants are recommended over synthetic antioxidants, which have been discovered to have adverse effects. Natural antioxidants are the secondary metabolites of phytochemicals. The impact of oxidants on human health and how natural antioxidants counteract them have both been reviewed. The main plant sources of natural antioxidants, several methods for recovering antioxidants from plant matrices, and the superiority of indirect supercritical fluid extraction over other methods are all discussed.

Keywords: oxidative stress, secondary metabolites, recovery processes, and process factors

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Victor Duniya Sheneni,[1] Sani Sade Muhammad,[1] Isaac Eleojo Shaibu[2]

[1]Department of Biochemistry, Faculty of Science, Federal University Lokoja, Nigeria

[2]Department of Biochemistry, Faculty of Natural Science, Kogi State University, Nigeria

Correspondence: Victor Duniya Sheneni, Department of Biochemistry, Faculty of Science, Federal University Lokoja, Lokoja, Nigeria, Tel +234-8033519009, Email victor.shenen@fulokoja.edu.ng, shenenivicto@gmail.com

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Introduction

Phytochemicals are plant-based bioactive compounds produced by plants for their protection. They can be derived from various sources such as whole grains, fruits, vegetables, nuts, and herbs, and more than a thousand phytochemicals have been discovered to date. Some of the significant phytochemicals are carotenoids, polyphenols, isoprenoids, phytosterols, saponins, dietary fibers, and certain polysaccharides. These phytochemicals possess strong antioxidant activities and exhibit antimicrobial, antidiarrheal, anthelmintic, antiallergic, antispasmodic, and antiviral activities.[1,2] In the human body and in plants, such as fruits and vegetables, antioxidants are secondary components or metabolites. Anything that inhibits or prevents the oxidation of a susceptible substrate might be referred to as an antioxidant. To stop the oxidation of the vulnerable substrate, plants create an astounding variety of antioxidant substances, such as carotenoids, flavonoids, cinnamic acids, benzoic acids, folic acid, ascorbic acid, tocopherols, and tocotrienols.[3] Vitamins A, C, and E as well as specific substances known as carotenoids (such as lutein and beta-carotene) are examples of common antioxidants.[4] Because our endogenous antioxidants fall short in protecting us from the ongoing and inescapable threat posed by reactive oxygen species (ROS; oxidants), it is thought that these plant-based dietary antioxidants play a significant role in maintaining human health.[5]

Polyphenols are a category of natural compounds with phenolic structures. This family has four major subclasses, such as flavonoids, stilbenes, phenolic acids, and lignans. Flavonoids are further classified as flavanones, flavones, flavonols, and anthocyanidins. Polyphenols are abundantly found in artichoke (*Cynara cardunculus var. scolymus* L.), spinach (*Spinacia oleracea* L.), broccoli (*Brassica oleracea var. italica* L.), chicory (*Cichorium intybus* L.), flax (*Linum usitatissimum* L.), onion (*Allium cepa* L.), apple (*Malus domestica* L.), plum (*Prunus subg. Prunus* L.), pear (*Pyrus* L.), grape (*Vitis vinifera* L.), and cherry (*Prunus avium* L.). Beverages such as olive oil, tea and red wine are considered good sources of polyphenols.[6] Flavanones have almost 350 aglycones and 100 glycosylate forms where a flavan nucleus is formed of two aromatic rings linked through a dihydropyrone ring.[6] Flavones represent a large group of flavonoids where the presence of a double bond between C-2 and C-3, as well as the attachment of the B ring to C-2, distinguishes these compound.[7] Flavonols have a double bond between C-2 and C-3 and they differ from flavanones by the hydroxyl group at the third position.[8] Anthocyanidins are mostly found in nature as their sugar-conjugated derivatives anthocyanins, which are responsible for the red, blue, and purple colors found in fruit and floral tissue.[9] Health benefits of polyphenols include action against free radicals; protective effects against cardiovascular diseases, cancers, and other age-related diseases; and prevention of inflammation and allergies.[10] Flavonoids have been also found to be beneficial in angina pectoris, cervical lesions, chronic venous insufficiency, dermatopathy, diabetes, gastrointestinal diseases, lymphocytic leukemia, menopausal symptoms, rhinitis, traumatic cerebral infarction, etc.

Oxidative stress is caused by the production of free radicals or reactive oxygen species (ROS) during metabolism and other processes that exceed the antioxidant capacity of a biological system.[11] According to Sian et al.[12] oxidative stress contributes to heart disease, malaria, neurological illnesses, cancer, AIDS, and the aging process.

Growing evidence that oxidative damage contributes to the onset of chronic, age-related degenerative diseases and that dietary antioxidants counteract this and reduce disease risk supports this idea.[13,14] Hence the need to extract these antioxidants from the plant matrices arises. In a recent study Grigonisa[15] antioxidants from the plants were isolated using a variety of extraction procedures, including dispersed-solids, percolation, Soxhlet, microwave aided extraction, and supercritical fluid extraction.[16] According to Nguyen et al.[17] supercritical fluid extraction (SFE) is a practical and sophisticated method for extracting antioxidants.

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Higher purity antioxidants are produced by the solid phase extraction (SPE) method using ultra critical fluids like CO2. The impact of oxidants on human health and how antioxidants counteract them are covered in this overview. The various types of antioxidants, their characteristics, and the methods used to extract them from plant matrices have all been covered in detail. We report recent developments in the supercritical extraction of antioxidants.

Formation of oxidants

Although oxygen is necessary for life, it can also cause tissue to be destroyed or impede its capacity to function normal.[18] Reactive oxygen species (ROS), also known as oxidants, free radicals, or oxygen-free radicals (OFR), are produced by a variety of exogenous and endogenous causes. One or more unpaired electrons are present in a free radical, which can sustain itself independently. The harmful effects of O_2 may result from the production of oxygen radicals. The catalytic elimination of the superoxide free radical, O2, is carried out by a group of enzymes known as superoxide dismutase (SODS).[19] An average person's bodily cells are attacked daily by 10,000–20,000 free radicals. While ROS are sometimes created deliberately to carry out necessary biological tasks, they are also sometimes created as a consequence of metabolic processes.[20]

Exogenous sources

Exogenous sources for the production of oxidants include environmental and artificial radiation exposure. Gamma rays and other low-wavelength electromagnetic radiation break bodily water, creating the hydroxyl radical (OH-), in the process. The resulting extremely reactive OH- starts to vigorously react with the adjacent cells.[21] Although most endogenous compounds react with OH- at rates faster than 1010 M-1 sec-1, OH- scavengers often have rates higher than this. Antioxidant systems protect against OH- damage by halting the synthesis of the compound and mending the harm it does.[22]

One to three percent of the oxygen we breathe in is thought to be converted into O2. Since humans utilize a lot of oxygen, a straightforward calculation reveals that the body produces around 2 kg of oxygen annually; those with chronic inflammation may produce even more. According to Fraga et al.[23] these oxidants cause damage to DNA, proteins, and lipids, which over time may accelerate aging and age-related illnesses.

Endogenous sources and characteristics of oxygen radicals

Free radicals are created in cells via enzymatically or non-enzymatically mediated electron transfer reactions in addition to external sources like radiation exposure. The main source of free radicals is electron leakage from electron transport chains, such as those in the mitochondria and endoplasmic reticulum, to molecular oxygen.[24] The four endogenous sources listed below are mostly where oxidants are produced in our body's cells.

a. The mitochondrial use of oxygen during regular aerobic respiration to create water. This process leaves behind oxidants like oxygen free radicals, H2O2, and hydroxyl radicals.

b. Phagogytic cells destroy bacteria and virus-infected cells, releasing nitric oxide, hydrogen peroxide, and oxygen free radicals in the process.

c. When peroxisomes break down fatty acids and other compounds, they release hydrogen peroxide as a byproduct, which catalase then breaks down. Oxidative DNA damage results from the non-degraded peroxide entering other cell compartments nearby.[25] A non-radical is created when two free radicals interact because their unpaired electrons form a covalent connection. But a free radical reacts with a non-radical to create a radical, which can start a chain reaction in the body.

d. Oxidants created during the process of degrading natural poisons on page 450.

To reduce the quantity of reactive oxidants and the harm they cause, organisms have evolved a variety of defense mechanisms.[26] The development of age-dependent diseases like cancer, arteriosclerosis, arthritis, neurodegenerative disorders, and others has been suggested to be significantly influenced by radical-related damage to DNA and proteins, despite the cell's anti-oxidant defense system to combat oxidative damage from free radicals.[27] According to Atoui et al.[13] reactive oxygen species interacts with DNA bases in cells to create broken bases or strands. Lipids or proteins are oxidized by oxygen radicals, creating intermediates that interact with DNA to generate adducts. Due to the changes in the environment, which are also caused by human activities like deforestation, an increase in atmospheric carbon dioxide, etc., it is very necessary to take antioxidants exogenously.

Antioxidant and its mechanism

According to Guteridge et al.[28] an antioxidant is a chemical that, when present in low concentrations compared to those of an oxidizable substrate, considerably slows down or stops that substance from oxidizing. Antioxidant enzymes, iron binding and transport proteins, and other substances impacting signal transduction and gene expression are included in the idea of antioxidants for the in vivo scenario. Antioxidants in meals and drinks are linked to the preservation of particular oxidation substrates or the production of particular oxidations. Other helpful concepts relating to antioxidants include synergism, antagonism, co-antioxidants, and oxidation retarders.

Synergism is the phenomena in which several chemicals, when present in the same system, have an impact that is more pronounced than if they were acting independently. Similar definitions of antagonism and co-antioxidants can be found by changing "more" to "less" and "same" to "same." Retarders of oxidation are substances that slow down the rate of oxidation without exhibiting a clear lag phase. The effectiveness of antioxidants is determined by the length of the lag phase and by a reduction in the overall rate of oxidation.

Chain-breaking antioxidants and preventative antioxidants are the two categories into which antioxidants are separated. By slowing down the rate of chain start, preventive antioxidants prevent oxidation. In most situations, the oxidation's byproduct, ROOH, hydroperoxide, is what starts the process. Antioxidants used as preventative measures change hydroperoxides into molecular compounds that do not have the ability to produce free radicals.[29] Most biologically active antioxidants that prevent oxidation also degrade peroxide. Some enzymes, including glutathione peroxidase, can convert lipid hydroperoxides to the equivalent alcohol as well as reduce H_2O_2 to water.

Antioxidants that break chains commercially are typically phenols or aromatic amines. Due to their capacity to capture peroxyl radicals, they exhibit antioxidant action. Synthetically produced antioxidants are another option. These fall under the category of artificial antioxidants. The main drawback of these antioxidants is their in vivo side effects.[30] When compared to synthetic antioxidants, it has been discovered that the majority of natural antioxidants have stronger antioxidant activity.

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According to a number of theories, fruits and vegetables' antioxidant components aid in the influence on defense. The benefits of taking antioxidant supplements have been demonstrated in epidemiological studies and intervention trials on the prevention of diseases like cancer and cardiovascular disease in individuals.[31,32]

Antioxidants made by plants for survival include carotenoids, flavonoids, cinnamic acids, benzoic acids, folic acid, ascorbic acid, tocopherols, and tocotrienols. Beta-carotene, ascorbic acid, and alpha tocopherol are a few of the well-known antioxidants.[33] It is known that beta-carotene is a precursor to vitamin A; the liver and the lining of the small intestine are where it gets transformed into vitamin A. According to Dagenais et al.[34] betacarotene is considered to be safer because it can be consumed in virtually infinite amounts without having a harmful effect on the body. Ascorbic acid has numerous beneficial effects.[35]

Ascorbic acid can function as an antioxidant, pro-oxidant, metal chelator, reducing agent, or oxygen scavenger depending on the situation. In aqueous systems with metals, ascorbic acid can operate as a pro-oxidant by lowering the metals, which become more potent oxidation catalysts in their lower valence state. Ascorbic acid is a powerful antioxidant at high concentrations in the absence of other metals.[36]

A class of substances known as vitamin E performs well-known antioxidant effects. Tocopherol and notably alphtocopherol are the vitamin E molecules with the highest biological activity. Mammalian tissue frequently contains tocopherol.[37] Se, an antioxidant that occurs naturally, prevents polyunsaturated fatty acid oxidation to maintain tissue suppleness. Se has a crucial role in glutathione peroxidase. According to Zima et al.[38] se deficiency has been linked to the onset of congestive cardiomyopathy, accelerated atherosclerosis, skeletal muscle myopathy, increased cancer risk, aging, cataracts, and dysregulated immunological function. As anticarcinogens and preventative measures against degenerative diseases, small molecule dietary antioxidants including vitamin C (ascorbate), vitamin E (tocopherol), and cartoneoids have drawn a lot of attention.[39] Table 1 provides information on various antioxidants are a few examples.[40–48]

Table 1 List of common antioxidants and their uses

Antioxidant	Plant sources	Applications
Beta-Carotene C40H56	*Elaeis oleifera, Elaeis Guineensis Momordica Cochinchinnensis Spreng Eurycoma Longifolia Zanthoxylum Myriacanthum*	Is reported to have analgesic, antidotal, aphrodisiac, diuretic, and vulnerary properties. Folk medicine uses oil palm as a liniment for slow-growing tumors and a treatment for rheumatism and headaches. Used as a male aphrodisiac, stomach discomfort remedy, and anticancer agent in steamed glutinous rice.
Alpha-Tocopherol C29H50O2	*Citrus Hystrix Calamus Sciþronum Averrhoa Belimbi*	Fruit is utilized in both savory and sweet foods as a preservative and flavour. Leaves are both a medication and a wash for hair. These canes have edible buds that are also medicinal and antibacterial in nature. They are frequently used to relieve pains and fever. The fruit's syrup can help with mild cases of internal hemorrhoids, stomach bleeding, and mild cases of bowel bleeding in addition to quenching thirst and reducing frenzied excitation.
Ascorbic Acid C6H8O6	*Apium Graveolens Sauropus Androgynous*	Rheumatism, lower back pain, anxiety, and arthritis. Resistant to illness and insects.
Palmitic Acid $CH_3(CH_2)_{14}COOH$	*Elaeis Oleifera, Elaeis Guineensis*	Aphrodisiac, anodyne, antidotal, diuretic, and vulnerable. Oil palm is a source of palmitic acid and is used as a traditional medicine for rheumatism, headaches, and cancer.
Beta Sitosterol C29H50O	*Morinda Citrifolia Alpinia Officinarum Sida Acuta*	Diabetes, hypertension, arthritis, skin disorders, and aging processes Stomach sickness, dyspepsia, vomiting, and flatulence are all prescribed as treatments for stomach cancer. Whole plant to relieve stomach pain.
Selenium	*Astragalus Membranaceus Valeriana Officinalis Achillea Millefolium*	Prevents cancer patients from experiencing severe adverse effects from chemotherapy. The development of mouse renal cell cancer is slowed. Immune system activation. Active sedation. Cardiovascular system general tonic that reduces blood pressure and slows heartbeat.
Anthraquinone C14H8O2	*Cassia Acutifolia*	Antihelminthic, antibacterial, laxative, diuretic, for treatment of snakebites and uterine disorders.
Tannic acid $C_{76}H_{52}O_{46}$	*Costus Spinosa*	Tanning of leather.
Quercetin $C_{15}H_{10}O_7$	*Blumea Balsamifera*	Treatment for the swelling of pancreas.

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Antioxidant and cancer

One human cell is thought to get around 105 oxidative impacts each day from hydroxyl radicals and other similar oxidant molecules. According to Lopaczynsk and Zeisel[49] and Dreher et al.[50] under typical metabolic circumstances, around 2-5% of the O2 absorbed by mitochondria is transformed to ROS. ROS are common oxidant by-products of aerobic metabolism. Thusly produced oxidative stress irreversibly alters the genetic code, resulting in a variety of degenerative or chronic disorders, including atherosclerosis and cancer (Ames et al., 1993). DNA damage that wasn't properly repaired could lead to mutations such base substitution and deletion, which could cause carcinogenesis.[51] It is believed that carcinogenesis and oxidative damage are both caused by two distinct pathways. The first approach involves changing the way that genes are expressed. The activation of growth signals and proliferation can result from epigenetic influences on gene expression.[52] According to Bohr et al.[53] strand breakage misrepair is hypothesized to cause chromosomal rearrangements, which then contribute to genetic amplifications, iterations in gene expression, and loss of heterozygosity, all of which may hasten the development of cancers. Signal transduction pathways can be affected by active oxygen species since they have been shown to trigger the protein kinase and poly (ADP ribosylation) pathways. Additionally, this may result in a modification of the expression of vital genes for tumor promotion and cell proliferation.[54] Free radical signal may be transmitted via ras signal transduction pathways, according to certain research.[55]

According to the second mechanism, radicals cause genetic changes in the form of mutations and chromosomal rearrangements, which can lead to the development of cancer.[56,57] Numerous chromosomal defects brought on by oxidative DNA damage result in a barrier of DNA replication and widespread cytotoxicity. Misrepair or ineffective replication can result in mutations, whereas strand breakage misrepair can cause chromosomal rearrangements. DNA damages are known to develop with age since repair mechanisms are known to deteriorate with time.[58] The frequency of mutations is influenced by the sequence specificity of DNA damage sites.[59] Therefore, research into the sequence specificity of DNA damage would be helpful in the fight against cancer. The quantity of oxidative DNA damages that are not repaired directly relates to mutagenic potential.

Antioxidant extraction processes

Although not every metabolite exists in every species, plants have a diverse range of metabolites, as many as 200,000 distinct chemicals.[60] These metabolites include derivatives of several different kinds of chemicals, including organic acids, amino acids, and fatty acids. The metabolites' physical-chemical characteristics vary greatly. Since the ideal extraction conditions vary greatly for various types of chemicals, it is necessary to select appropriate extraction techniques. Effective plant metabolite extraction requires appropriate homogenization of the plant tissue.

There are several methods, including homogenization using a metal pestle attached to an electric drill Edlund et al.[61] grinding using a mortar and pestle and liquid nitrogen[62] and milling in vibration mills with chilled holders. The level of homogenization determines how well the solvent can permeate the tissue, which has a significant impact on how long it takes to extract the solvent from the tissue. Shaking homogenized plant tissue in organic solvents or solvent mixtures at low or high temperatures is the most typical method for extracting metabolites.[63] The primary solvents employed for extracting. 101 include methanol, ethanol, and water. for non-polar ones, a solvent. Other extraction methods include supercritical fluid extraction (SFE)[64] pressurized liquid extraction (PLE), microwave-assisted extraction (MAE), subcritical water extraction (SWE), and pressurized liquid extraction (PLE). Antioxidants from the plants have been isolated using a variety of extraction methods, including Soxhlet, microwave assisted extraction (MAE), and supercritical fluid extractions (SFE) (Lopez-Sebastian et al., 1998). Each extraction has unique benefits and drawbacks. Long extraction times, thermolabile chemical degradation, and a small range of solvent options are the main drawbacks of Soxhlet extraction.[65] Other traditional liquid-solid extraction techniques take a long time, need huge volumes of solvents, which can include dangerous compounds, and as a result, further cleaning and concentration stages are needed. MAE[66] a different laboratory scale extraction process, has recently been used and has proven to be noticeably quicker. When compared to Soxhlet extraction, MAE offers greater recoveries and requires less solvent. The primary drawback of MAE is that it is frequently carried out at higher temperatures (110–150 C). The thermolabile compounds may become denaturated at this temperature range.[67]

Carbon dioxide supercritical fluid extraction (SFE) is a highly appealing extraction technique.This is owing to the fact that CO2 is an inexpensive, inert, non-flammable, non-explosive, clean, odorless, and colorless solvent that doesn't leave any solvent residue in the finished product. Additionally, carbon dioxide's 304o K threshold temperature makes it desirable for the extraction of thermo labile chemicals. Although it can be somewhat increased by applying the right modifier, carbon dioxide is constrained by its insufficient solvating power for highly polar analytes. In some applications, substantial modifier concentrations (10–50%) are also of interest. SFE modifiers, like ethanol, are injected at levels of 1–10%. The most crucial elements for successful recoveries are typically thought to be the optimization of the working parameters, including pressure, modifier %, fluid pressure and temperature, and extraction duration.[68]

Grigonisa et al.[15] compared the outcomes of various extraction techniques for the separation of the antioxidant 5,8 dihydroxycoumarin from sweet grass (*Hierochlo odorata*). For the Soxhlet extraction, a high yield of 0.58% and concentration of 40.4% were achieved. However, due to the lengthy extraction process and high solvent consumption, this type of extraction is not always suitable for industrial applications. Alternatives include SFE extraction, which yields the second-best compound yield of 0.46% and a concentration of 20.3%. Due to a reduced extraction yield of 0.30% for 5, 8-dihydroxycoumarin, MAE was less successful in 102. Comparison of extraction times reveals that MAE takes 15 min, while SFE takes 1–2 h.

The latter, however, needs some time for the extract to cool down; as a result, the overall MAE and extract cooling time lengthen. For thermolabile antioxidants, Soxhlet and microwave assisted extraction are inappropriate. By recycling, the main drawback of high modifier usage in SFE can be greatly minimized. As a result, SFE is a viable technique for isolating antioxidants. When sampling medicinal plants, methods like Soxhlet extraction, microwave assisted extraction (MAE), or pressurized liquid extraction (PLE) frequently lead to non-selective extraction of significant amounts of undesirable components (such as lipids, sterols, and chlorophylls), which can negatively impact the product's quality.[69] Prior to turning the extract into a useful product, additional clean-up procedures are typically required for the direct supercritical fluid extraction process, which uses supercritical carbon dioxide to directly extract chemicals from plant matrices.[70] Solid phase extraction is a well-liked and efficient tool for analyte extraction and/or concentration as well as for clean-up.

The solubility and functional group interactions of the sample, solvent, and adsorbent are tuned to affect the retention and elution in solid-phase extraction (SPE), a straightforward preparation technique based on the principles of liquid chromatography.[71] Analytes recovered from non-polar solutions onto polar sorbents range in polarity from moderately polar to polar. Cyano, diol, or amino groups are added to sorbents for normal phase. Analytes are removed from polar solutions onto non-polar sorbents that range in polarity from non-polar to moderately polar.[72] Silica gel and synthetic resins, two forms of adsorbent materials that have undergone chemical modification, allow for exact group separation based on various physicochemical interactions. Indirect supercritical fluid extraction, a technique that combines solid phase extraction with supercritical fluid technology Khundker et al.[73] has been used to extract phytochemicals from aqueous matrices and has been reported to produce antioxidants with a greater yield, concentration, and purity.

Future potential

The main illnesses that affect humans include diabetes, cancer, cardiovascular disease, and others. It has been discovered that antioxidants can fend off certain illnesses. Antioxidants are primarily found in plants. They can, however, be produced synthetically. When consumed in vivo, synthetic antioxidants have negative side effects. Recovery of antioxidants from plants has advanced greatly due to the most recent trend of turning to natural sources for health and medicine. Antioxidants can be extracted from plants using a variety of approaches and procedures, each of which has advantages and disadvantages of its own. Therefore, it is necessary to select an appropriate technique that can produce larger concentrations and purer antioxidants while still being economically practical on an industrial scale. This is met via indirect supercritical fluid extraction. To get the intended outcomes, the proper process parameters, such as temperature, pressure, and exact modifier, must be chosen.

Conclusion

Numerous diseases have been linked to reactive oxygen species (ROS), also known as oxidants, which are produced in our bodies as a result of external and endogenous stimuli. The potential of phytochemical antioxidants as health benefits is being discovered daily through research. This is as a result of their capacity to scavenge the free radicals, reactive oxygen species, or oxidants that initiate cell damage. Antioxidants made from synthetic materials have been found to be unhealthy. The majority of naturally occurring antioxidants derived from plants are healthier for humans and have higher antioxidant activity. Antioxidants are extracted from plant matrices using a variety of extraction techniques, including Soxhlet extraction, subcritical water extraction (SWE), pressurized liquid extraction (PLE), microwave-assisted extraction (MAE), and supercritical fluid extraction (SFE). It is discovered that solid phase extraction using indirect supercritical fluid extraction or supercritical fluid technology is an industrially viable technique.

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Conflicts of interest

The authors declares that there are no conflicts of interest.

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