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Secondary Metabolites as a Nonenzymatic Antioxidant System in Plants: A Brief Review

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Abstract:

Plants are constantly challenged by abiotic and biotic stresses including drought, salinity, heavy metals, UV radiation, and pathogen infections that result in excessive production of reactive oxygen species (ROS). Excessive ROS can cause oxidization of proteins, lipids, carbohydrates and nucleic acids which are detrimental to cellular components and thus impact plant growth and development. To avoid oxidative stress, plants require a finely tuned collaborative antioxidant defense system which includes enzymatic (peroxidases, catalases) and non-enzymatic (ascorbate, glutathione, alkaloids, flavonoids, and phenolic chemicals) antioxidants. Phenolic metabolites are derived from shikimate and mevalonate pathways with various biological actions. They are accumulated differentially across a variety of tissues, such as, roots, leaves, stems, flowers, fruits and seeds. The structural characteristics of phenolics aromatic rings with hydroxyl functional groups render them electro-donors or hydrogen donors to initiate ROS scavenging and ultimately stabilize the free radicals. Thus, we review herein that phenolics act as non-enzymatic antioxidants whose upregulation under stress is important for adaptive mechanism to postpone lipid peroxidation of membranes and nucleotides injury leading to premature programmed cell death. Therefore, this study seeks to collate the literature to date, to elucidate the biochemical truth about phenolics and their ecological significance in stress response with suggestions for further investigation.

Keywords: antioxidants, phenolic chemicals, secondary metabolites, plants, ROS.

مركبات الايض الثانوي كنظام مضاد للأكسدة غير انزيمي في النباتات

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الخلاصة:

تواجه النباتات باستمرار ضغوطًا حيوية وغير حيوية مثل الجفاف والملوحة والمعادن الثقيلة والإشعاع فوق البنفسجي والإصابات الممراضية، والتي تؤدي إلى الإنتاج المفرط لـ reactive oxygen species ROS. إن الزيادة في ROS تسبب أكسدة البروتينات والدهون والكربوهيدرات والأحماض النووية، مما يضر بالمكونات الخلوية وينعكس سلبًا على نمو النبات وتطوره. ولتجنب الإجهاد التأكسدي، تحتاج النباتات إلى منظومة دفاعية من مضادات الأكسدة تعمل بتنسيق دقيق وتشمل مضادات الأكسدة الإنزيمية (peroxidases, catalases) وغير الإنزيمية (ascorbate, glutathione, alkaloids, flavonoids, phenolic chemicals). وتشتق المركبات الفينولية من shikimate و mevalonate pathways، وتمارس أدوارًا بيولوجية متعددة. تتراكم هذه المركبات في أنسجة مختلفة مثل الجذور والأوراق والسيقان والأزهار والثمار والبذور.

إن الخصائص التركيبية للفينولات، والمتمثلة في الحلقات العطرية مع مجموعات هيدروكسيل، تجعلها مانحة للإلكترونات أو للهيدروجين لبدء إزالة ROS واستقرار الجذور الحرة في النهاية. ومن هنا نستعرض أن الفينولات تعمل كمضادات أكسدة غير إنزيمية، حيث يُعَدّ تحفيز إنتاجها تحت ظروف الإجهاد آلية تكيفية مهمة لتأخير بيروكسدة الدهون في الأغشية ومنع تلف النيوكليوتيدات التي تؤدي إلى الموت الخلوي المبرمج المبكر. وبناءً على ذلك، يهدف هذا البحث إلى تجميع الأدبيات الحديثة لتوضيح الحقيقة البيوكيميائية حول الفينولات وأهميتها البيئية في الاستجابة للإجهاد مع تقديم مقترحات لمزيد من الدراسات المستقبلية.

الكلمات المفتاحية: مضادات الاكسدة، المركبات الفينولية، مركبات الايض الثانوي، النباتات، انواع الاوكسجين التفاعلي.

1. Introduction:

The plants produce primary and secondary metabolites which both exhibit specific functions. Proteins, lipids, nucleic acids, carbohydrates and fatty acids act in plant primary metabolism. Also, plants produce secondary metabolites that act as a natural supply of antioxidants to protect the plants [1]. Different plant components, including leaves, stems, flowers, fruits, and seeds, produce these substances. There are several ways to release these chemicals into the atmosphere or into the rhizosphere, including Leaching, root exudation, volatilization, and soil residue breakdown. It is divided into several types depending on its composition and characteristics. The study by [2] divided secondary metabolites into organic acids such as ketones, aliphatic

aldehydes, and straight-chain alcohols, Long-chain fatty acids, unsaturated lactones, simple, quinines, derivatives of cinamic acid, Coumarins chemicals, phenols, Tannins, Flavonoids and Terpenoid. Certain large groups of secondary metabolites are synthesized under environmental stress and act as antioxidants in plants. Both enzymatic and non-enzymatic antioxidants make up a plant's antioxidant system; the most significant non-enzymatic antioxidants are alkaloids, glutathione, phenolic chemicals, ascorbic acid, flavonoids, and non-protein amino acids. Additionally, plants produce enzymatic antioxidants like peroxidase, catalase, superoxide dismutase and monodehydroascorbate reductase [3].

Phenolic chemicals:

Plants synthesize large numbers of phenol derivatives collectively named as phenolic chemicals. The chemical structure of these compounds is aromatic with one or more hydroxyl groups and also have at least one benzene ring with one or more hydroxyl groups, which is able to donate hydrogen atoms or electrons in order to equation radicals or any environmental stress and reactive oxygen species [1]. Biosynthetic pathway of principal phenolic and flavonoid secondary metabolites in plants. Shikimic acid and phenylpropanoid pathways. Involved in glycolytic and pentose phosphate pathways—phosphoenolpyruvate and erythrose-4-phosphate—contribute to shikimate pathway; derive aromatic amino acids (phenylalanine, tyrosine, tryptophan) and gallic acid. Phenylalanine is converted to cinnamic acid via the phenylpropanoid pathway and to p-coumaric acid and p-coumaroyl-CoA, central and common intermediates for an extensive variety of phenolic chemicals (i.e. tannins, lignins, coumarins) as well as precursors to flavonoids via chalcone. Differing branches then proceed to create flavonols, anthocyanidins, catechins, isoflavonoids, flavones [4] Figure (1).

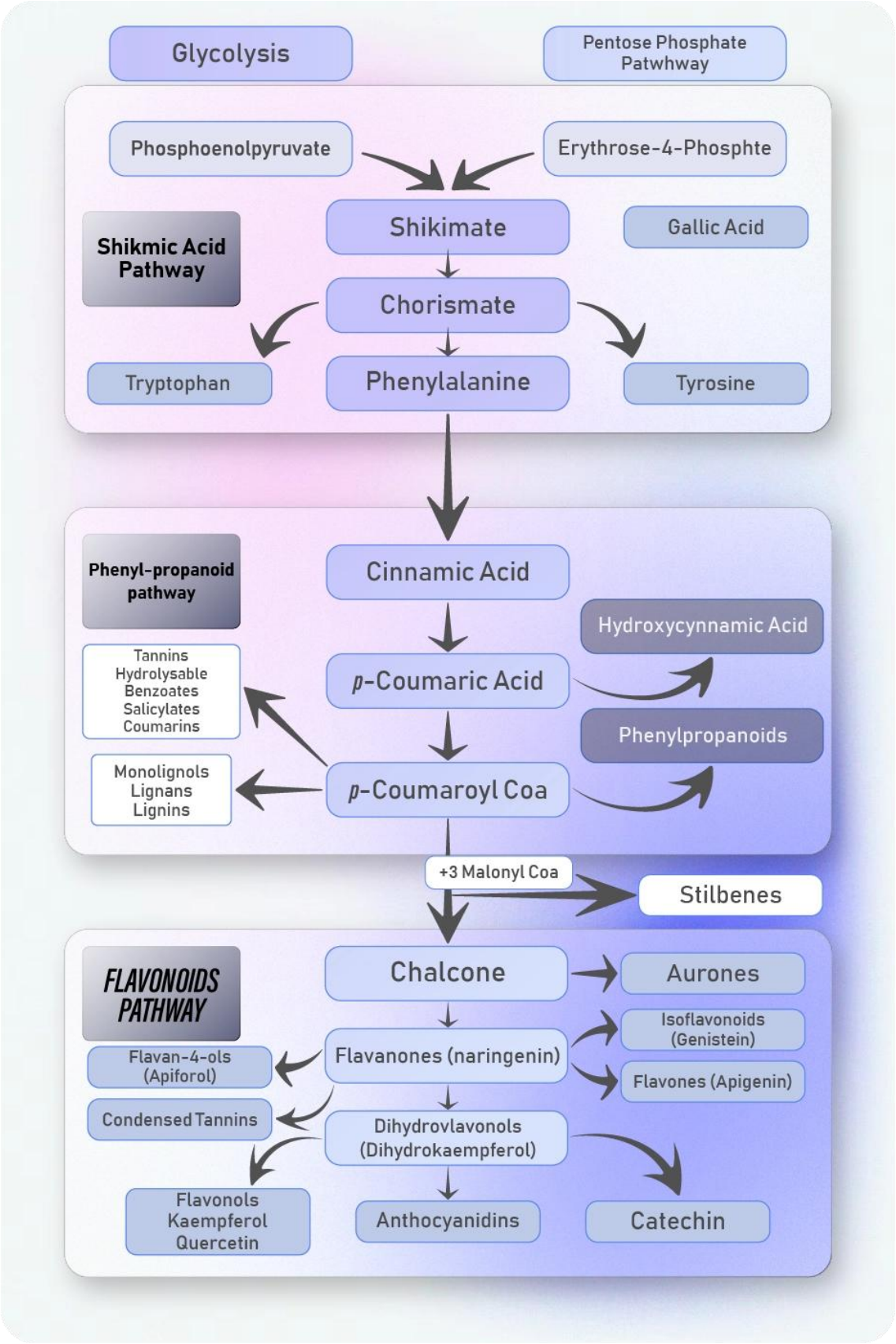


Fig. 1: Biosynthesis pathway of phenolic chemicals in plants.

Certain phenolic chemicals are categorized based on the degree of oxidation or unsaturation or the number of carbon atoms in their structure [5]. Plants photosynthesize approximately 2% of all organic molecules, subsequently transforming them into phenolic chemicals. Most phenolic groups are conjugated naturally with mono or polysaccharides. Plant phenolics are the most widely produced and distributed in plants, bacteria, algae, and fungi. Plant leaves contain hydroxycinnamic acids, flavonoids, lignin and glycosylated flavonoids [6,7]. Phenolic chemicals are classified into simple and polyphenolic chemicals according to the number of phenol units contained. Simple phenolics such as catechol, resorcinol, and hydroquinone (Figure 2) contain one phenol unit.

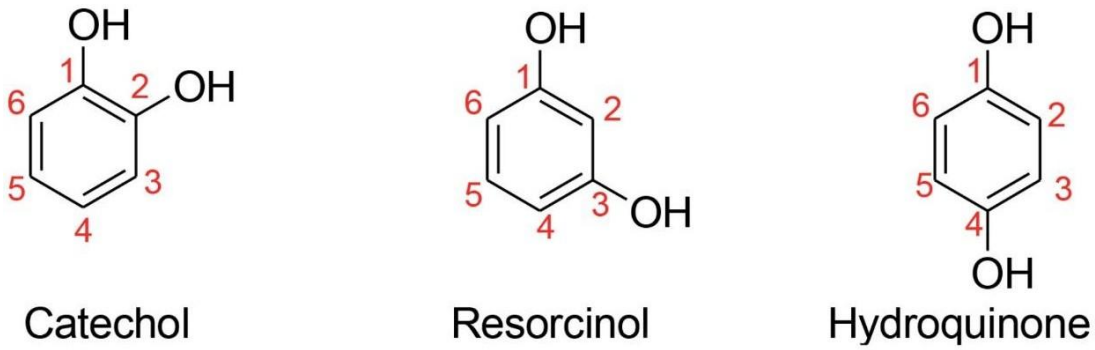


Fig. 2: Simple phenols.

Polyphenols contain more than one phenol group, such as coumarin, ferulic acid, sinapic acid, cinnamic acid, caffeine, and hydrocoumarin (Figure 3).

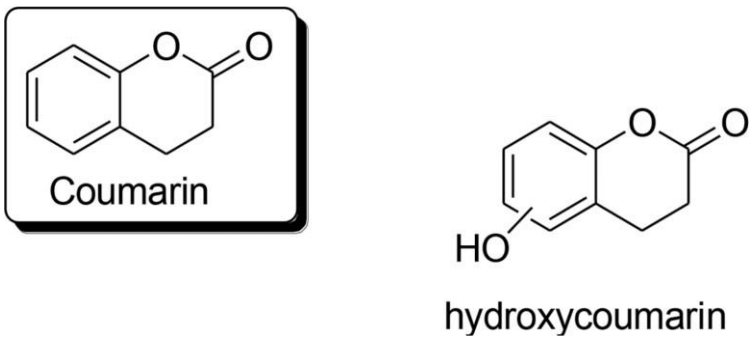


Fig. 3: Polyphenols.

Biosynthesis of phenolic chemicals is often affected by plant environments, like plant pathogens, moisture, UV light, pH, and water stress. When plants are exposed to environmental stressors like drought stress, phenolic chemicals play a significant role as their non-enzymatic antioxidant system. UV light, pH, and numerous additional biotic and abiotic elements cause plants to create an unbalanced rate of reactive oxygen. Synthesis of the plant antioxidant system and reactive oxygen species (ROS) result in the plant's elevated oxidative stress [8]. The aim of this review is to summarize the role of secondary metabolites, especially phenolic chemicals, as non-enzymatic antioxidants in plants exposed to various stresses. The review also identifies gaps in current knowledge and suggests directions for future research to improve our understanding of plant stress tolerance and its applications in crop production.

Phenolic chemicals' Function in Abiotic Stress: Under abiotic stress, phenolic acids interact with their environment and mediate adaptive responses by producing protective secondary metabolites[9]. Phenolic chemicals build up in plant tissue in response to negative stress, which is thought to be a physiological adaptation for survival in the face of environmental disruptions [10]. The antioxidant properties of flavonoids, hydroxycinnamate esters, tannins, and lignin aid in the defense against biotic and abiotic stress. Phenolic chemicals also promote lipid peroxidation and break free radical chains [11]. Phenolic chemicals have been classified into five subcategories (flavonoids, coumarins, phenolic acids, lignin and tannins.), It has been improved that increased gene expression was the cause of the rise in phenolic chemicals in several stressed plants [12, 13, 14]. They conclude that the accumulation of phenolic chemicals in *Rehmannia glutinosa* occurs under stress because of increasing enzyme activities such as phenylalanine ammonia lyase (PAL). Abiotic stress enhances phenolic acid levels through cinnamic acid metabolism, including methylation and hydration/dehydration reactions, supporting plant survival under environmental challenges. Implication of plants to different stresses by scavenging of ROS, boosting the production of secondary metabolites, like phenolic chemicals, as a way to protect plants, [14] It is notedthat when plants are exposed to injury, such as a wound or cut in tissues, they may use sustainable methods as a defense by boosting antioxidant activity and phenolic compound production. When plants grow in areas of adverse conditions face water stress, heavy metals stress, extreme temperatures, salinity, and other abiotic stresses, which affect plant development, growth and productivity. [15]. Different reactive oxygen species (ROS) were created by plants in response to stressors. Overproduction of ROS caused damage to the cells of plants, such as carbohydrates, proteins, and DNA, finally causing disturbance of the balance between normal (ROS) production and antioxidants, leading

to oxidative stress in plants [16]. The antioxidant system helps plants adapt to oxidative stressors. It consists of enzymatic antioxidants like peroxidase, catalase, glutathione reductase, superoxide dismutase, hydroascorbate reductase, glutathione peroxidase, and ascorbate reductase, as well as nonenzymatic antioxidants like ascorbic acid, glutathione, flavonoids, phenolic chemicals, and alkaloids [17]. Under severe situations, plants' antioxidant systems are crucial because they scavenge reactive oxygen species (ROS), prevent lipid peroxidation, DNA damage, and protein breakdown; inhibit enzymes, and ultimately postpone cell death [18].Phenolic chemicals' function in heavy metal stress: One significant source of stress is brought on by the toxicity of heavy metals which causes harmful effects in physiological processes [19,20], It proved that nanoparticles such as Ag, CuO, CeO2 and Cu can be used for the induction of phenolic chemicals; increased production of ROS led to an increase in phenolic chemicals. Biosynthesis of Phenolic substances in Zea mays L. in contaminated soil with aluminum ion, rootexudates of the plant have high levels of catechin and quercetin [21,22] was reported that high levels of flavonoids in cells of Digitalis lanata L. are usuallyassociated with high concentrations of CuSo4.Increasing the supply of Molybdenum (Mo) caused improved antioxidant system in plant enzymatic and nonenzymatic processes and increased metabolism and growth [23].When plants take up heavy metals (Cr, Cd, Hg, Co, Mn, Ni, Pb and Zn) this inhibits major physiologicalactivities in plant cells such as photosynthesis, cell elongation, and respiration. The harm of heavy metals takes place through the increase of ROS by oxidation and inhibition of polynucleotide and protein functional groups, inactive enzymes responsible for detoxification of free radicals [24].

Role of phenolic chemicals under drought stress:

Drought is the primary abiotic stressor that affects plant development and reduces plant yield, plant phenolic chemicals content increases under water stress. [25] reported that quercetin concentration increased significantly in white clover when the plant was exposed to drought. [26] has reported that different types of plants such as cereals, fruits and vegetables accumulate phenolic chemicals when grown under drought conditions.Plants antioxidants that accumulate under water stress include phenolic chemicals and flavonoids. *Amaranthus tricolor* is vegetable accumulate flavonoids in response to drought conditions [27]. Under water stress phenolic chemicals act as a nonenzymatic antioxidant system to equation the harmful effects of (ROS), even plants resistant to drought conditions (arid or semiarid habitats) may produce high concentrations of phenolic chemicals and flavonoids in contrast to the same plants that thrived in water with moderate levels of moisture [28]. In response to drought stress, plants

physiologically constrict their stomata, reducing transpiration and water loss,CO2 concentration and fixation via the Calvin cycle reduced, increase NADPH+H production during photosynthesis (light reactions), causing reduced growth of the plant. As a response, the plant increases antioxidants [29]. An experiment studied the response of 21 barley cultivars to water stress, tested the impact of water deficit on phenolic compound content in the studied cultivars, antioxidant enzyme activity and oxidative stress. It was found that water stress increased content of phenolics and flavonoids. [30]

Phenolic chemicals' function in nutrient stress:

The development of plants depends on nutrients from their environment which are regulated by several factors, including microorganisms, climate, substrate quality, and nutrient leachates into the soil [31]. Plant phenolic affect the activity of soil microorganisms and nutrient cycling. When a plant is exposed to nutrient deficits, phenolic chemicals are crucial. such as (N, P, K) and S, rapidly increasing the concentration of phenolic chemicals [6]. Tomato plants increased levels of Anthocyanin and Quercetin when grown under nutrient stress conditions [32, 33], demonstrating that limitation of (N) in *Lactuca sativa* L. caused an increase in phenolic chemicals and antiradical activity, while reduction in (P) nutrition did not affect the composition of Lettuce phytochemicals.

5. Conclusions:

Plants subjected to general stress effects on cellular components like carbohydrates, proteins, lipids, and DNA were omitted with a focus on the general effect of various biological and environmental stresses leading to oxidation. Secondary metabolites and the plant's antioxidant system, which consists of both enzymatic and nonenzymatic antioxidant systems in plants act as a nonenzymatic antioxidant system divided into organic acids, ketones, simple unsaturated lactones, quinones, cinnamic acids, phenolics, tannins, flavonoids, and terpenoids. Phenolic chemicals are a certain group of secondary metabolites synthesized under environmental stress that act as a nonenzymatic antioxidant in plants, equation ROS, and delay cell death, released to the environment by various ways such as volatilization, leaching, root exudation, and decomposition.

This review focuses on the key mechanisms used by plants when subjected to various biotic and abiotic challenges, with secondary metabolites being among the most important of these plant-derived chemicals. Heat stress, salt, drought, and acidity all cause a significant increase

in the accumulation of such secondary metabolites, which act as protective agents for the plant, reducing environmental damage.

Despite the overwhelming evidence of secondary metabolites' protective impact, there are significant limitations and information gaps. For example, the particular regulatory networks that regulate their production under combined stress circumstances are not yet fully understood, and the heterogeneity among plant species needs more comparative research. Furthermore, most recent findings are based on controlled laboratory circumstances, and their relevance to real-world settings requires confirmation. Future study should integrate molecular, physiological, and ecological techniques to better understand these chemicals' dynamic activities, with a focus on genetic engineering, breeding programs, and biotechnological applications to improve crop stress tolerance.

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