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Comprehensive Insights into Three Key Classes of Grape Phenolics: Anthocyanins, Stilbenes, and Flavonols

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Abstract:

Polyphenols are abundant in grapes both qualitatively and quantitatively because of their unique properties, anthocyanins, flavonols, and stilbene derivatives in particular play crucial roles in plant metabolism. Red grapes and wines get their color from anthocyanins, which also give the wine its organoleptic qualities. They are employed for chemotaxonomic research and to assess the grape's polyphenolic ripening stage. They are utilized in the food and pharmaceutical industries, are natural colorants, and have antibacterial, antioxidant, and anticarcinogenic properties. They also have preventive effects on the human cardiovascular system. Stilbenes are vine phytoalexins found in grape berries that are linked to the health benefits of wine consumption. Resveratrol, the main stilbene, has cardioprotective, anti-inflammatory, antioxidant, and anticancer properties. Grapes also contain resveratrol dimers and oligomers, which are either created by extracellular enzymes released from pathogens in an effort to get rid of unwanted harmful substances or synthesized by the vine as active defenses against external attack. A common class of flavonoids, flavonols has the ability to co-pigment (with anthocyanins) and provide photoprotection. White grapes only contain derivatives of quercetin, kaempferol, and isorhamnetin due to the lack of expression of the enzyme flavonoid 3',5'-hydroxylase, but red grapes typically also include derivatives of myricetin, laricitrin, and syringetin. The chemistry of these substances has been better understood and greatly improved over the past 10 years due to the technical advancements in analytical instruments, especially mass spectrometry. Together with the most recent information on their chemistry, this article provides a brief overview of the biosynthesis and biological function of these grape polyphenols.

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Introduction:

Grapes have a wide variety of secondary metabolite classes, with polyphenols having a particularly rich qualitative and quantitative makeup. The primary polyphenols are derivatives of anthocyanin, flavonol, and stilbene, three groups of chemicals with distinctive properties and significant functions in plant metabolism. Cultural customs, environmental factors, and variations in grape types all have a significant impact on phenolic content. Berry phenolics improve numerous facets of human health and add to the quality of wine. Grape extracts are utilized as sources of natural chemicals in the culinary, pharmaceutical, and nutraceutical industries, while anthocyanins, flavonols, and stilbenes are important components of wine quality due to their biological and organoleptic properties. As a result, research is being done to better understand their chemistry, their functions in grape physiology, and the qualities of their products. The technological advancement of analytical apparatus over the past decade has significantly enhanced and broadened our understanding of these chemicals. The invention of multiple mass spectrometry (MS/MS and MSn) and liquid chromatography mass spectrometry (LC/MS) in the 1990s provided extremely helpful methods for researching polyphenol structures. We now have a better understanding of the methods by which anthocyanins in grape and wine contribute to winemaking and wine aging, thanks to numerous studies that have experimentally supported structures that were only postulated until recently. 18 stilbene derivatives, including monomers, glucoside derivatives, dimers (viniferins), trimers, and tetramers, were found in a recent comprehensive qualitative and quantitative analysis of stilbene compounds in grapes using precise mass spectrometry. In addition to myricetin, quercetin, and kaempferol, which are typically found in wines, LC/MS analysis of grape flavonols has also revealed the existence of isorhamnetin, laricitrin, and syringetin derivatives. The production and biological function of various polyphenol groups in grapes, together with the most recent findings regarding their chemistry, are briefly discussed in this review.

Polyphenol Biosynthesis in Grapes

Flavonoids

The phenylpropanoid pathway is used to create all phenolic compounds from the amino acid phenylalanine. The shikimate pathway, which connects the manufacture of aromatic amino acids and secondary metabolites with the metabolism of carbohydrates, is the source of phenylalanine. Figure 1 depicts the general phenylpropanoid route. Chalcone synthase can make flavonoids, while stilbene synthase can produce stilbenes, two major families of chemicals. Numerous kinds of metabolites, including flavonols, flavan-3-ols, proanthocyanidins, and anthocyanins, are produced by the flavonoid pathway.

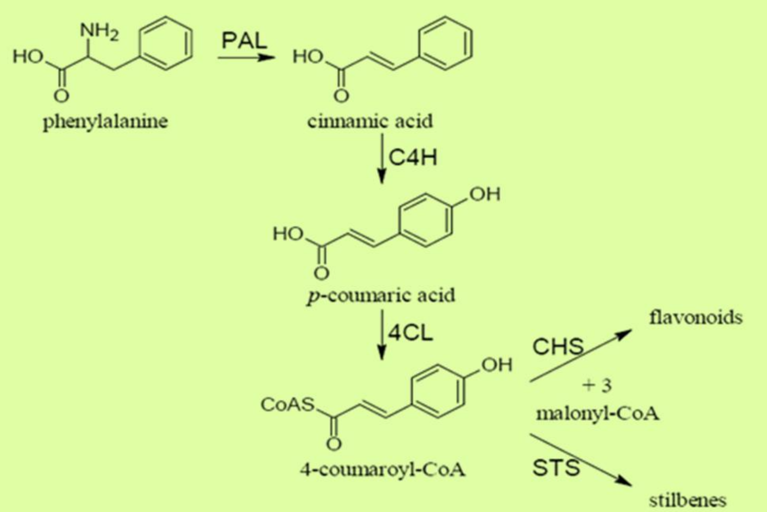

Figure 1. General phenylpropanoid pathway. PAL, phenylalanine ammonia lyase; C4H, cinnamate-4-hydroxylase; 4CL, 4-coumaroyl:CoA-ligase; CHS, chalcone synthase; STS, stilbene synthase.

Anthocyanins and Flavonols

Normally, flavonols have glycosylation at the C ring's C-3 position. Glycosylation at positions 3 and 5 of the C ring creates anthocyanins from anthocyanidins, which are then stored in berry skins and the flesh of some "teinturier" kinds from veraison until full maturity, at which point synthesis ceases. Because glycosylation causes intramolecular H-binding inside the anthocyanin molecule, anthocyanidin glycosides are typically more stable than their corresponding aglycones. Both anthocyanidins and flavonols can be glucosylated by the enzyme UDP-glucose:flavonoid 3-O-glucosyl transferase (UFGT), although the former is catalyzed with a significantly higher efficiency. The majority of red grapes contain acylated anthocyanins, most likely as a result of the anthocyanin acyl-transferase enzyme. The cytosol produces anthocyanins, which are then transported into the vacuole and stored there as anthocyanic vacuolar inclusions, which are colorful coalescences. Vacuolar uptake may rely on vesicular trafficking-based or tonoplast transporter-mediated pathways. There are two potential processes for transporter-mediated uptake. MATE-type proteins are found in the tonoplast and act as acylated anthocyanin transporters that are dependent on vacuolar H+. Glutathione S conjugate pumps called ATP-binding cassette proteins are involved in the uptake of glycosylated flavonoids even when there isn't an H+ gradient present. Therefore, it is anticipated that glutathione S-transferases (GSTs) will take role in vacuolar trafficking. The GST gene family member that shares transcriptional patterns with anthocyanin accumulation is one of them. Grape berries have yielded another putative anthocyanin carrier that is quite comparable to mammalian bilitranslocase. Even though there is evidence for many transporters, it is still unclear whether anthocyanins enter the vacuole as single molecules before aggregating or whether the tonoplast interacts with cytoplasmic vesicles that contain aggregated anthocyanins.

Stilbene Derivatives

All higher plants contain flavonoids, but only a small number of species generate stilbenes. Two closely related enzymes that specifically regulate the manufacture of flavonoids or stilbenes are stilbene synthase (STS) and chalcone synthase (CHS). Higher plants have evolved STS from CHS multiple times, and switching from chalcone to stilbene synthesis just requires a few amino acid exchanges. Following its development in the grapevine lineage, the STS gene has greatly expanded across the grapevine genome, primarily by local duplications, giving rise to a 43-member family. It is currently uncertain how redundant each gene copy is or whether specialization has taken place. On the other hand, the grapevine genome contains only three copies of CHS, and the grape fruit exhibits a variety of expression patterns. Other enzymatic processes that transform resveratrol into downstream derivatives include a glucosyltransferase, which can generate glucosides of cis- and trans-resveratrol and has residual glucosyl activity on hydroxycinnamic acids and certain flavonoids, and a resveratrol O-methyltransferase (ROMT), which catalyzes the conversion of resveratrol into the highly fungitoxic pterostilbene. Three molecules of malonyl-CoA and four coumaroyl-CoA are accepted as substrates by STS. Similar to CHS, STS produces resveratrol through three condensation processes. Resveratrol exhibits distinct ring-folding from tetrahydroxychalcone, the result of CHS, due to the removal of the terminal carboxyl group in the STS reaction before the closure of the A ring. Thus, CHS and STS regulate the entrance points into the flavonoid and stilbene pathways, respectively, and compete for the same substrates.

Effects of Agrochemicals and Plant Activators on Grape Polyphenols

Exogenous elicitors can affect anthocyanin biosynthesis, and many chemical substances have been used to boost anthocyanin levels in grape fruit. It has been demonstrated that the use of hormones such abscisic acid (ABA), jasmonate molecules, ethylene, and salycilic acid, as well as non-hormone substances like ethanol and eutypine, can enhance the manufacture of anthocyanins. Additionally, recent research revealed that grapes treated with benzothiadiazole (BTH) and a BTH/methyl jasmonate combo had higher anthocyanin content. Additionally, it has been demonstrated that pectin-derived oligosaccharides and chitosan, a linear polysaccharide, increase the polyphenolic and anthocyanic content of grapes. In the vine, agrochemicals and plant activators function as elicitors for the production of stilbenes. The most widely used agrochemical is fosetyl-Al, a systemic fungicide that works against Oomycetes fungi such as Plasmopara viticola and causes the leaves to produce more ε-viniferin and trans-resveratrol. Numerous organisms, including Aspergillus carbonarius, Aspergillus japonicus, Rhizopus stolonifer, Tricoderma viride, Erysiphe necator, Laminarina spp., Bacillus spp., and others, as well as numerous products, including aluminum chloride, ozone, sucrose, dimethyl-β-cyclodextrin, MeJ, BTH, chitosan oligomers, salicylic acid, ABA, beta-amino-butyric acid (BABA), emodin, and UV treatments, are examples of plant activators. Although SAR (systemic acquired resistance) is known to affect how agrochemicals and plant activators affect grape stilbenes, flavan-3-ols, and anthocyanins, and activators cause the expression of phenylpropanoid genes in grapevines, little is known about how they affect flavonols. Given the proximity of the two biosynthetic routes, an induction of their synthesis, akin to that seen for anthocyanins, could be anticipated. Iriti et al. found that the level of total flavonoids was very poorly affected in a "one vintage (2004)/one variety (Merlot)/one activator (BTH)" experiment. Nevertheless, flavonols were not the focus of this investigation. An increase in flavonol concentration was noted in a two-year study on treatment with BTH and methyl jasmonate (MeJ) on Monastrell grape (2009–2010), however the results are not entirely consistent for the two years under investigation due to the large biological variability. While MeJ treatment only boosted flavonols in one year, albeit with a more noticeable effect (+131%), BTH-treated grapes had greater flavonol concentrations in both years (+17% and +56%). Although there was no difference in the flavonol profile, the differences between treated and untreated samples were more noticeable in the 2010 colder, humid year, when the environment was more conducive to the growth of infections. In any event, further research is required to fully understand the role that agrochemicals and plant activators play in the synthesis of flavonoids.

Conclusion

The chemotaxonomy of grapes can benefit greatly from an intimate structural understanding of anthocyanins, which is also a useful tool for assessing polyphenolic ripening in grapes and creating new research on biosynthetic pathways. Additionally, it can be applied enologically to enhance winemaking and wine aging techniques. Certain non-V vinifera grape varietals have been found to be both subjectively and quantitatively extremely rich in anthocyanins. Because these grapes extracts contain significant amounts of anthocyanin-acyl derivatives, which maintain a more stable color in slightly acidic or neutral solutions, their use in the natural colorant business has a lot of potential. Stilbenes in grape compounds have a very complicated chemistry. In reaction to biotic and abiotic stress, plants produce these substances, which function as phytoalexins. They are also linked to the health benefits of wine consumption, and studies can be conducted to improve farming and winemaking methods to boost their presence in grapes and wines. Finally, because the pharmaceutical and nutraceutical industries are increasing their need for stilbenes, it is also important to produce them from sustainable sources. All species of Vitis include flavonols, which are common secondary metabolites among flavonoids. Their profiles in grapes are greatly altered by genetic factors at the cultivar level, making them effective markers in Vitis vinifera chemotaxonomy. Thus, both white and red types can be categorized using flavonoid profiles. They have a strong correlation with anthocyanin profiles in red cultivars. In terms of concentration, the flavonoid class is also among the most significant grape flavonoids, particularly in white grapes. Because they have a photo-protective action against excessive direct sunlight, environmental factors primarily affect their abundance in ripe berries. It is important to remember that grapes and wine are two of the best sources of flavonols in the human diet, and their presence has frequently been linked to their quality. Our understanding of the chemistry of grape and wine polyphenols has significantly increased with the advent of sophisticated MS technology. The momentum of using these analytical methods to investigate grapes and wines over the coming years will greatly advance our understanding of the subject and yield major advantages for the domains where these polyphenolic compounds are relevant.

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