Selected Phenolic Compounds in Cultivated Plants: Ecologic Functions, Health Implications, and Modulation by Pesticides

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Phenolic compounds are widely distributed in the plant kingdom. Plant tissues may contain up to several grams per kilogram. External stimuli such as microbial infections, ultraviolet radiation, and chemical stressors induce their synthesis. The phenolic compounds resveratrol, flavonoids, and furanocoumarins have many ecologic functions and affect human health. Ecologic functions include defense against microbial pathogens and herbivorous animals. Phenolic compounds may have both beneficial and toxic effects on human health. Effects on low-density lipoproteins and aggregation of platelets are beneficial because they reduce the risk of coronary heart disease. Mutagenic, cancerogenic, and phototoxic effects are risk factors of human health. The synthesis of phenolic compounds in plants can be modulated by the application of herbicides and, to a lesser extent, insecticides and fungicides. The effects on ecosystem functioning and human health are complex and cannot be predicted with great certainty. The consequences of the combined natural and pesticide-induced modulating effects for ecologic functions and human health should be further evaluated. — *Environ Health Perspect* 107(Suppl 1):109–114 (1999). *http://ehpnet1.niehs.nih.gov/docs/1999/Suppl-1/109-114daniel/abstract.html*

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Plants synthesize primary carbohydrates, lipids, and proteins. Secondary plant compounds are synthesized from lipid precursors and aromatic amino acids. Phenolic compounds build an important portion of the secondary plant compounds. Lignin, a complex polymer of phenylpropane units is, quantitatively, the most important phenolic compound in plants. Other abundant compound classes are the flavonoids, stilbenes, coumarins, and polyflavonoids (condensed tannins). Phenols in plants have diverse functions such as stabilization of the structure, protection from herbivory, protection from ultraviolet (UV) light, exchange of information with symbionts, coloration of blossoms, and biocidal effects against bacteria and fungi (*1*). After the death of plants, phenolics may persist for weeks or months and affect decomposer organisms and decomposition processes in soils (*2*). Therefore, their effects are not restricted to single plants but may extend to the functioning of whole ecosystems.

Phenolic plant compounds may have both beneficial (*3*) and toxic (*4*) effects on human health. Only a few of the thousands of phenolic compounds have been investigated for their beneficial and health-damaging effects. Compared to pesticides and other anthropogenic chemicals, the knowledge of toxicologic effects of most phenolic compounds in plants is limited.

External stimuli can modulate the synthesis and therefore change the chemical composition or quantities of phenolic compounds in the plants. External stimuli include microbial infections, UV light, and mechanical wounding of the plant (*5–8*), as well as chemical stressors such as heavy metals and pesticides. Furthermore, genes coding for enzymes of the secondary metabolism have been transfected into plants to increase the resistance against pathogens. The purpose of this paper is to investigate how plant phenolics influence ecologic functions, to what extent human health is affected, and whether modulation of plant phenolics by pesticides impairs ecologic functions and human health.

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Abbreviations used: HDL, high-density lipoproteins; LDL, low-density lipoproteins; 5-MOP, 5-methoxypsoralen; 8-MOP, 8-methoxypsoralen; NO, nitric oxide; PAL, phenylalanine ammonia-lyase; PUVA, psoralen ultraviolet A; SOM, soil organic matter; UV, ultraviolet.

Distribution and Ecologic Functions

Biochemistry of Selected Plant Phenols

Phenylalanine, synthesized in the shikimate pathway, is a starting point of the synthesis of plant phenols (Figure 1) (*1*). Phenylalanine ammonia-lyase (PAL) catalyzes the deamination of phenylalanine to cinnamate. Coumaric acid is built by introducing a hydroxy group in the phenyl ring of the cinnamic acid, a process catalyzed by cinnamate-4-hydroxylase (a P450-monooxygenase). This phenylpropanoid is a precursor of stilbenes, flavonoids, and furanocoumarins.

Stilbenes are synthesized from coumaroyl-CoA and three molecules of malonyl-CoA under cleavage of four molecules of carbon dioxide. The formation of chalcone, which can be transformed into flavonon by chalcone isomerase, is performed by chalcone synthase from coumaroyl-CoA and three molecules of malonyl-CoA under cleavage of three molecules of carbon dioxide. Flavanone is a precursor of a variety of flavonoids such as flavones, flavonoles, anthocyanidins, and isoflavones. The precursor of furocoumarins is coumaric acid. Psoralen is built by the introduction of a C2-group in hydroxycoumarin, which is formed by hydroxylation and the building of an inner ester of cumaric acid.

Resveratrol

trans-Resveratrol, a *trans*-3,5,4′-trihydroxystilbene (Figure 1), occurs in grape species and a few other plant species (*9*). The peanut is the only other plant species used for human nutrition that contains *trans*-resveratrol in significant amounts (*10*). In the grape species resveratrol reaches concentrations of 50 to 400 µg/g fresh weight in the leaves. Resveratrol is also synthesized in the berries and in lignified plant tissues (e.g., stalks and kernels of the berries) (*9*). Concentrations in the skin (pericarp) of the berries are high compared to those in the flesh (*11*). During mashing, a part of the resveratrol from the skins is dissolved in the

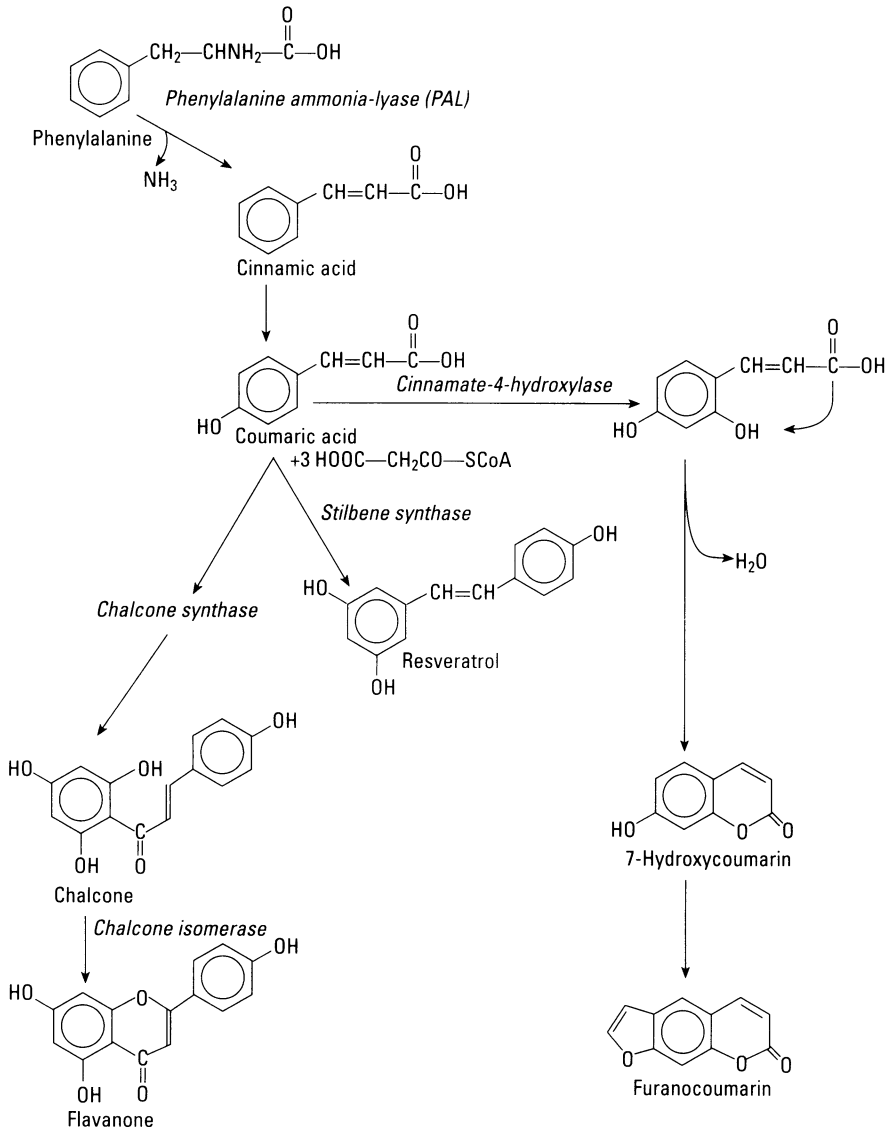

Figure 1. Schematic biochemical pathway of the synthesis of flavanone, resveratrol, and furanocoumarin.

must. In a Spanish study concentrations of *trans*-resveratrol were 0.011 to 0.547 mg/liter in white wine and 0.07 to 1.06 mg/liter in rosé (*12*). Slovenian white wine contained up to 0.6 mg/liter and red wine 0.9 to 8.7 mg/liter (*13*). The reason for the higher content of *trans*-resveratrol in red wine is the long contact time between the berry skin and must. Must for white wine is immediately separated from berry residues after mashing, whereas must for red wine is fermented with the berry skins for several days.

Resveratrol also exists in the chemical form of *cis*-resveratrol and as *trans*- and *cis*-piceid (β-glucosides of resveratrol) (*12,14*). Both isomeric forms were detected as glyca and aglyca in white wine, rosé, and red wine (*12,15,16*). *cis*-Isomers are probably synthesized during the vinification process but do not reach the concentration of *trans*-isomers in the wine (*12,17*). Polymerization of resveratrol leads to α-viniferin and ε-viniferin. The ability to synthesize resveratrol decreases with ripening. Therefore, increased contents at harvest time cannot originate from an increased synthesis (*13*). Because resveratrol increases simultaneously with a decrease of piceid (*17*), the increase of resveratrol might be related to the hydrolysis of the resveratrol glycoside.

The synthesis of *trans*-resveratrol in the plants can be induced by microbial infections (*9*) and UV radiation (*17,18*). The content of resveratrol in grape determines the natural resistance to the fungus *Botrytis cinerea* (gray mold) (*19*). After an infection of a single berry, the resveratrol content in the neighboring berries increases within a short time. The fungus is able to synthesize an enzyme that degrades resveratrol. This results in a low concentration of resveratrol in heavily infected plant tissue. However, because the synthesis of the enzyme is slower than the induction of resveratrol, the plant can usually prevent or limit a fungal infection (*20*). This mechanism of defense has only been adopted by few plant species because most lack the enzyme stilbene synthase (resveratrol synthase). To allow the synthesis of stilbenesynthase in tobacco, rape, tomatoes, potatoes, and rice, the gene coding for stilbene synthase has been engineered into these plant species (*21*). Resistance to fungal infections was increased in these transgenic plants (*21*).

Flavonoids

Flavonoids, which are derived from flavanone (Figure 1), include more than 4000 chemical structures and are widely distributed in the plant kingdom. Examples are the flavanones naringenin and sakuranetin in rice (*22,23*), the isoflavonoids kievitone and phaseollin in beans (*24–26*), and the catechins epicatechin and epigallocatechin in green tea (*27*). Catechins and flavan-3,4-diols can polymerize to polyflavonoids called condensed tannins. Flavonoles are the most abundant flavonoids. The concentration of quercetin, the most important flavonole, is < 10 mg/kg in the edible parts of most vegetables (*28*). Exceptions include onions, kale, French beans, apples, cherries, and broccoli, where concentrations may amount to 30 to 490 mg/kg (*28,29*). Like most other flavonoids, quercetin in plant tissue is found mainly in its glycosidic form, e.g., as rutin.

Flavonoids in plants fulfill a variety of functions. They determine the color of the blossoms, protect the plant from UV radiation, and have biocidal effects on bacteria and fungi. Certain isoflavonoids attract insects, promote growth of several microorganisms, and induce the nodulation gene in the nitrogen-fixing bacteria *Rhizobium* sp. and *Bradyrhizobium* sp. (*30*). Other flavonoids help to repel herbivorous insects. In woody plants these are mainly the condensed tannins, whereas in herbs the flavonoles (quercetin, rutin) predominate.

The synthesis of flavonoids can be induced by external stimuli. UV radiation and microbial infections are effective stimuli, but chemical stress may also increase the synthesis of flavonoids. Phaseollin,

phaseollinisoflavane, and kievitone in bean seeds increased to 5.78, 5.11, and 9.90 µg/g following exposure to 1 mM silver nitrate (*25*). In response to a treatment with mercuric chloride, phaseollin and kievitone accumulated in cotyledons, hypocotyls, and roots of the seedlings (*24*). The exposure to silver nitrate and mercuric chloride had a more pronounced effect on the synthesis of flavonoids than had microbial infections.

Furanocoumarins

Furanocoumarins (Figure 1) are found in approximately 15 plant families. Several of these plants, e.g., lemon, celery, pastinak, parsley, and carrots, are part of the human diet. In lemons the furanocoumarins bergamottin and isopimpinellin predominate. Concentrations in the rind are up to 100 times higher than in the flesh (*31*). In celery the furanocoumarins psoralen, xanthotoxin (8-methoxypsoralen [8-MOP]), and bergapten (5-methoxypsoralen [5-MOP]) are most abundant. Green leaves usually contain greater amounts of furanocoumarins than yellow leaves; outer leaves usually contain more than inner leaves. Leaves usually contain more than stalks and stalks usually contain more furanocoumarins than roots (*32,33*).

After infection of celery by the pathogenic bacterium *Erwinia carotovora* the content of psoralen, xanthotoxin, and bergapten increased from < 2.5 mg/kg (fresh weight) in healthy tissues to > 20 mg/kg in putrefied tissues (*34*). Therefore it was hypothesized that furanocoumarins may play an important role in the defense of microbial infections. After infection the concentration of marmesin initially increased to approximately 200 mg/kg (fresh weight). After 4 days the concentration of marmesin decreased and the concentration of the psoralens (psoralen, bergapten, xanthotoxin, and isopimpinellin) increased (*35*). During postharvest storage of celery, marmesin contents decreased and psoralen content increased at the same rate (*35*). Marmesin presumably loses its hydroxypropyl group via oxidation catalyzed by a P450 monoxygenase to yield psoralen (*32*). Because marmesin is at least 100 times more effective against infections by various fungi (*Botrytis cinerea, Alternaria alternata, Sclerotinia sclerotiorum*), it was postulated that marmesin is the primary antimicrobial agent.

UV radiation (< 400 nm) induces the synthesis of furanocoumarins, which are often produced in tissue exposed to the sun. It is assumed that furanocoumarins protect the plant against UV-induced mutations (*32*). Leaves of giant hogweed (*Heracleum lanatum*) extrude furanocoumarins to the surface. Because furanocoumarins absorb UV radiation, their function on the leaves is similar to that of a suntan lotion (*36*). Additional abiotic factors inducing the synthesis of furanocoumarins include mechanical damage, low temperature, and chemicals (sodium hypochlorite, copper sulfate) (*37*).

Psoralen, xanthotoxin, and bergapten in plant tissue ingested by animals may induce phototoxic effects and protect plants from herbivory. For example, the shrub *Pituranthos triradiatus* growing in the Negev desert in Israel contains several furanocoumarins and is rarely used as food resource by hyraxes (small mammals of the order Hyracoidea). Starved hyraxes offered *P. triradiatus* as their only food and then exposed to the sun showed phototoxic symptoms such as apathy, photophobia, and injuries around the eyes and the back 3 to 4 hr after feeding (*38*). The polyphageous southern armyworm *Spodoptera eridania* (Noctuidae) dies after eating a diet with 0.1% (fresh weight) xanthotoxin if exposed to UV radiation (*39*). Some insects prevent UV radiation by living in rolled leaves and survive by feeding on plants with high contents of furanocou-marins (*39*). Other insects have evolved a biochemical resistance to furanocoumarins. Studies on lepidoptera of the genus Papilio and plants of the family Umbelliferae indicate that the evolution of species in these taxa was closely interdependent. In this coevolution new coumarins in the plants allowed for a better defense against herbivores, and biochemical pathways adapted to the detoxification of these coumarins enabled insects to explore new food resources not yet exploited by other insect species (*40*).

Beneficial and Toxic Effects on Human Health

Resveratrol

Resveratrol. Resveratrol is consumed with red wine and, in smaller amounts, with white wine or peanuts. Low to moderate drinking of red wine is associated with decreased mortality, mainly because of a reduction of coronary heart diseases (*41*). Coronary heart diseases in France are less frequent than in other countries with similar risk factors (smoking, fat content in the diet, lack of exercise). This phenomenon has been called the French paradox (*41*). It has been hypothesized that the reduction of the incidence of coronary heart diseases was related to the pharmacologic properties of resveratrol (*20*).

Low-density lipoproteins (LDL) transport cholesterol to the extrahepatic tissues and high-density lipoproteins (HDL) are responsible for the reverse transport of cholesterol (*42*). Following oxidation LDL are accumulated in subendothelial cells, the reverse transport of cholesterol is blocked, and the adhesion of platelets to the epithelium is promoted (*20*). Antioxidative compounds hinder these processes and reduce the risk of coronary heart diseases. Resveratrol has antioxidative properties *in vitro* (*20*). *In vivo*, however, the antioxidative properties of red wine were not correlated with resveratrol, but with gallic acid, catechin, myricetin, quercetin, and caffeic acid (*41*). Phenolic compounds can also modulate the production of nitric oxide (NO) by endothelial cells and thereby affect vasodilation and the adhesion of platelets to the endothelium. In contrast to quercetin and tannin, however, resveratrol does not affect NO production (*43*).

Resveratrol, but not quercetin, inhibits the *in vitro* synthesis of thromboxanes and prostaglandins in the cyclooxygenase pathway; quercetin inhibits the production of leukotrienes in the lipoxygenase pathway (*20*). Thromboxane A_2, which is built from prostaglandin H_2 in the platelets, induces the aggregation of platelets. Prostaglandin I_2 (prostacycline), which is built from prostaglandin I_2 in the endothelium, inhibits the aggregation of platelets (*42*). Disturbances in the proportion of thromboxane A_2 and prostaglandin I_2 contribute to the generation of thromboses. Leukotrienes are mediators of inflammatory reactions, are built in the neutrophil leukocytes, and are presumably related to the occurrence of arteriosclerosis.

Clinical studies with healthy male volunteers drinking 375 ml red or white wine per day did not confirm the results of the *in vitro* studies. Both red and white wine ingestion were related to an increase of HDL cholesterol and a decrease of platelet aggregation (*20*). This was surprising because not only resveratrol but also other phenolic compounds (e.g., catechin, epicatechin, and quercetin) differed between red and white wine by a factor of 10 to 20. This study supports the hypothesis that alcohol itself may be the dominant biologically active component. The reason for the missing *in vivo* effect of the phenolic compounds may be a low absorption in the gastrointestinal tract (*20*). The beneficial effects of resveratrol cannot be quantified definitively. However, it seems that flavonoids are of greater importance as protective factors against coronary heart diseases.

Flavonoids. Flavonoids are found in almost every food item or beverage of plant origin. Studies in the United States in the 1970s showed that the average daily intake of flavonoids in glycosidic form was approximately 1 g/person (*44*). The largest contributors were cola, cocoa, coffee, beer, wine (420 mg/day), and fruits (including fruit juices) (290 mg/day). Minor quantities resulted from the consumption of cereals (44 mg/day); potatoes, bulbs, and roots (79 mg/day); peanuts and other nuts (45 mg/day); and vegetables and herbs (162 mg/day). An average of 5% of the flavonoids were taken up as quercetin. In vegetables concentrations of quercetin may be high, i.e., 284 to 486 mg/kg in garlic, 110 mg/kg in kale, and 30 mg/kg in broccoli (measured as aglycon). In red wine quercetin amounts to 4 to 16 mg/liter; in fermented tea to 10 to 25 mg/liter (*45*). Total amounts in green tea reach 1 g/liter (*46*) with up to 293 mg/liter in the form of (-)-epigallocatechin gallate (*47*). Inhabitants of Japan, who drink up to 10 cups (approximately 180 ml) of green tea per day, have a daily intake of 540 mg (-)-epigallocatechin, which corresponds to approximately 1.8 g total flavonoids. The absorption of flavonoids, especially of the glycosidic forms, from the gastrointestinal tract is usually low.

Flavonoids have a variety of pharmacologic effects. They protect LDL from oxidation (reduction of arteriosclerotic plaques), inhibit the aggregation of platelets (antithrombotic effect), promote vasodilation (antihypertensive and antiarrhythmic effects), and modify the synthesis of eicosanoids (antiprostanoid and antiinflammatory effects) (*45*). Quercetin and myricetin have the most pronounced antioxidative effects.

Some flavonoids are mutagenic *in vitro* and are discussed as potential carcinogens by Brown (*48*). Their mutagenic activity depends on the metabolism and type of flavonoid. Quercetin and myricetin are mutagenic without enzymatic activation by cytochrome P450; an activation increases their mutagenicity. Kampferol is only mutagenic following activation. Some mutagenically active flavonoids may even be inactivated by cytochrome P450. Approximately 15 different quercetin feeding studies with mice, rats, and hamsters did not show an increase of tumor incidence except for bladder and intestine tumors in rats (*45*). It appears that depending on the chemical structure and the dose, flavonoids have both positive and negative effects on human health. This complex behavior of flavonoids seems reasonable because many flavonoids in plants have evolved because of their toxic effects on herbivores and pathogens. For humans the positive effects are assumed to predominate because epidemiologic studies report mostly positive effects of a flavonoid-rich diet.

Furanocoumarins. The uptake of furanocoumarins by humans in the United States was calculated to be 1.3 mg/person/day. Calculations were based on data of average consumption and furanocoumarin contents of different food items (*49*). The uptake was age dependent with maximum uptake at 16 to 20 years of age (2.48 mg/person/day). The biggest contribution to the uptake was from limes, which had a total concentration in the oil of 46.7 mg/kg bergamottin, isopimpinellin, and other furanocoumarins. An important contributor to the uptake via limes results from individuals drinking lemon–lime-flavored carbonated beverages.

Several furanocoumarins, especially those with a linear structure, can be activated by UV radiation and are phototoxic. After the uptake of furanocoumarins, exposed skin areas may develop erythematous sunburnlike symptoms and, in severe cases, blistering. These symptoms are well known from celery workers or from people who were exposed to giant hogweed (*Heracleum montegazzianum*) (*33*). The consumption of food containing furanocoumarins can produce the same symptoms. For example, a 65-year-old woman was hospitalized with severe burns after consuming approximately 500 g celery and visiting a suntan parlor afterward (*50*). Some furanocoumarins are genotoxic and possibly carcinogenic in combination with exposure to UV radiation (*33*).

The photoactivation of furanocoumarins depends on UV radiation putting electrons into higher energetic levels. Most electrons return to the ground state by emitting fluorescent or phosphorescent light. The translocation of energy from electrons to other molecules is the basis of phototoxic effects. This includes the covalent binding of furanocoumarins to the DNA, the production of reactive oxygen species, and the binding to receptors that regulate growth and differentiation of cells (*33*). Target cells of these effects are epithelial cells of the skin (e.g., keratinocytes, melanocytes) and the eye (cornea) as well as lymphocytes circulating in the peripheral blood vessels.

Eating average portions of celery and other vegetables containing furanocoumarins does not cause phototoxic burns. However, exposure to UV radiation (e.g., remaining on ice, snow, or water) should be kept low after eating extremely large portions of celery. The carcinogenic potential of furanocoumarins is presumably low but cannot be accurately quantified.

8-MOP and 5-MOP are used in the psoralen UVA (PUVA)-therapy of psoriasis. Patients are given doses of 30 to 60 mg 8-MOP or 60 to 120 mg 5-MOP 3 times per week over 3 to 6 weeks, followed by controlled exposure to UVA approximately 2 hr after the intake. The PUVA-therapy can also be used against vitiligo, cutaneous T-cell lymphoma, and eczema. Depending on the patient's skin type, the PUVA therapy increases the risk of nonmelanomous skin cancers. A previous exposure to arsenic or ionizing radiation additionally increases the risk (*51*). Therefore, benefit and risk of a PUVA therapy should be carefully balanced against each other.

Modulation of Phenolic Compounds in Plants by Pesticides

Herbicides are able to modulate concentrations of secondary compounds at application regimes that are not lethal for plants (*52*). Herbicides often reduce the carbon fixation by plants (Table 1). This may decrease the proportion of carbon allocated to the synthesis of secondary compounds (*52*). Other herbicides have more specific effects. Glyphosate blocks the shikimate pathway, thereby reducing the synthesis of aromatic amino acids (phenylalanine, tyrosine, tryptophan) and causing an accumulation of shikimate (*52,53*). Gallic acids, which are components of hydrolyzable tannins, are mainly synthesized from shikimate. Their contents in soy leaves and beans are increased after the application of glyphosate (Table 2). The contents of many other phenols, which are synthesized from aromatic amino acids, are reduced (Table 1). By other mechanisms, e.g., an increased PAL synthesis, contents of secondary compounds in plants may increase (Table 2). Herbicides such as dichlobenil, amitrol, acifluorfen, and paraquat can increase the PAL synthesis (*54*). Diphenyl ethers (e.g., acifluorfen) exert herbicidal effects mainly by oxidative damages (singlet oxygen of protoporphyrin) (*52*). Possibly as a protective reaction to the oxidative damages, plants increase the PAL synthesis and synthesize more flavonoids. For example, xanthotoxin content in celery is increased after the application of acifluorfen (Table 2) (*55*). Other herbicides that inhibit photosystem II (triazine, urea,

Table 1. Herbicides reducing the content of secondary compounds in plants.

Herbicide	Plant	Secondary compound	Factor of reduction	Reference
Atrazine	Soja	Anthocyan	4	Lydon and Duke (*53*)
Buthidazol	Maize	Anthocyan	4.3	Lydon and Duke (*53*)
Dinoterb	Pea	Flavonols	3.3	Lydon and Duke (*53*)
Glyphosate	Bean	Phaseollin	1.4–1.5	Lydon and Duke (*53*)
	Soja	Anthocyan	2	Lydon and Duke (*52*), Hoagland (*54*)
		Glyceollin	5	Lydon and Duke (*52*)
Metribuzin	Potato	Phenols, total	1.1–3.3	Lydon and Duke (*52,53*)
Sethoxydim	Raspberry	*o*-Phenols	2	Lydon and Duke (*53*)

Table 2. Herbicides increasing the content of secondary compounds in plants.

Herbicide	Plant	Secondary compound	Factor of increase	Reference
Acifluorfen	Pea	Pisatin	19	Lydon and Duke (*52,53*), Kömives and Casida (*55*)
	Bean	Phaseollin	>47	Lydon and Duke (*53*), Kömives and Casida (*55*)
	Broad bean	Medicarpin	>45	Lydon and Duke (*52,53*), Kömives and Casida (*55*)
		Wyeron	>15	Kömives and Casida (*55*)
	Celery	Xanthotoxin	4	Kömives and Casida (*55*)
	Soja	Glyceollin I-III	>79	Lydon and Duke (*52*), Kömives and Casida (*55*), Cosio et al. (*56*)
	Pisatin	Glyceofuran	>16	Kömives and Casida (*55*), Cosio et al. (*56*)
Chlomethoxyfen	Rice	Biphenyl-2-ol	3	Lydon and Duke (*52,53*)
Chlorsulfuron	Soja	Anthocyan	1.77	Lydon and Duke (*52*), Hoagland (*54*)
Glyphosate	Oat	Phenols, total	1.43	Lydon and Duke (*53*)
	Soja	Gallate	9	Lydon and Duke (*52,53*)
		Glyceollin	2.22	Lydon and Duke (*53*)
		Protocatechuate	102	Lydon and Duke (*52,53*)

amide, and carbamate classes) are able to reduce the PAL synthesis in soy seedlings (*54*). Some fungicides can also modulate concentrations of secondary compounds. Maneb, benomyl, and nabam induced the synthesis of hydroxyphaseollin in soy (*53*). The effect of insecticides on plant phenolics was not extensively investigated (*52*). Methomyl modulates the contents of tannin in cotton (*52*). Most pesticides are applied together with adjuvants, which enhance the effects of insecticides. Triton X-100, which is used as adjuvant at 0.01% in 50 to 100 ml/liter acifluorfen, increases the synthesis of flavonoids in soy leaves (*52,56*).

The examples demonstrate that concentrations of plant phenolics may be both increased (Table 1) and decreased (Table 2) because of pesticide application. However, these examples cover only a minor portion of pesticides used and plant phenolics potentially affected. It is unknown whether other pesticides also have modulating effects and whether the many plant phenolics not yet investigated also can be modulated.

The diverse modulating effects of pesticides on phenolic compounds in plants may affect the functioning of ecosystems. Direct effects involve the colonization of plants with pathogenic microorganisms or the change of the plant's attractivity to herbivores (*54*). Pesticides can also affect the secondary metabolism indirectly by eliminating nontarget plants, that compete for light and nutrients or serve as habitats for pathogens and herbivores (*53*). Plant phenolics affect not only aboveground organisms and ecologic functions. They might also be important belowground if they are applied to the soil or if plants exposed to pesticides are left in the field. During decomposition these plant tissues may either decay to carbon dioxide or become part of the soil organic matter (SOM) (*57*). Part of the SOM is degradable by the soil microflora and animals; another part may persist for centuries because of its chemical properties or physical protection (*58*). Plant phenolics may persist for weeks or months after plant death (*2*) and affect suitability as a food resource for soil organisms and decomposition rates of the SOM (*59*). SOM is an important component of soil fertility because it affects nutrient cycles and aggregate stability as well as water, temperature, and oxygen distribution in the soil. The sustainable use of SOM is a key factor in the management of agricultural ecosystems. Therefore, it would be interesting to know whether modulated plant phenolics in the SOM change the suitability as a food resource for the soil microflora and animals and thereby have an impact on the functioning of ecosystems.

Pesticide residues in plants are regulated to protect human health. They are measured when plants enter the market. However, this procedure does not consider that pesticides modulate secondary metabolism in plants when they are applied. The question arises whether it is conceivable that pesticide-induced changes in the chemical composition of plants influence human health. The examples resveratrol, flavonoids, and furanocoumarins indicate that plant phenolics may have subtle effects on physiologic processes that are relevant to human health. These effects may be beneficial, e.g., due to the inhibition of the oxidation of macromolecules and platelet aggregation or by their pharmacologic properties. Depending on concentration and specific chemical composition, however, plant phenolics may also be toxic, mutagenic, or cancerogenic. The consequences of a modulation of plant phenolics on human health are complex and cannot be predicted with certainty. It may be that the modulation of plant phenolics at the time of application and not the usually low level of pesticide residues at the time of consumption is critical for human health. Another complication in the interpretation of the role of pesticides originates from the additional effects of natural factors such as UV radiation, nutrient availability, pathogens, and herbivore pressure. The consequences of these modulating effects for ecosystem functioning and human health should be evaluated further.

REFERENCES AND NOTES

1. Heldt HW. Pflanzenbiochemie. Heidelberg: Spektrum Akademischer Verlag, 1997.
2. Horner JD, Gosz JR, Cates RG. The role of carbon-based plant secondary metabolites in decomposition in terrestrial ecosystems. Am Nat 132:869–883 (1988).
3. Bitsch R. Pflanzenphenole und ihre gesundheitliche Wirkung. Natwiss Rundsch 2:47–51 (1996).

4. Schlatter J, Luethy J. Vorkommen und Toxizitaet von Phytoalexinen in pflanzlichen Lebensmitteln. Mitt Geb Lebensmittelunters Hyg 77:404–419 (1986).
5. Grisebach H, Ebel J. Phytoalexine, chemische Abwehrstoffe hoeherer Pflanzen? Angew Chem 90:668–681 (1978).
6. Benhamou N. Elicitor-induced plant defence pathways. Trends Plant Sci 1:233–240 (1996).
7. Matsuki M. Regulation of plant phenolic synthesis: from biochemistry to ecology and evolution. Aust J Bot 44:613–634 (1996).
8. Kuc J. Phytoalexins, stress metabolism, and disease resistence in plants. Annu Rev Phytopathol 33:275–297 (1995).
9. Langcake P, Pryce RJ. The production of resveratrol by *Vitis vinifera* and other members of the vitaceae as a response to infection or injury. Physiol Plant Pathol 9:77–86 (1976).
10. Goldberg DM. Does wine work? Clin Chem 41:14–16 (1995).
11. Jeandet P, Bessis R, Gautheron B. The production of resveratrol (3,5,4′-trihydroxystilbene) by grape berries in different developmental stages. Am J Enol Vitic 42:41–46 (1991).
12. Romero-Perez AI, Lamuelaraventos RM, Waterhouse AL, Delatorreboronat MC. Levels of *cis*- and *trans*-resveratrol and their glucosides in white and rose *Vitis vinifera* wines from Spain. J Agric Food Chem 44:2124–2128 (1996).
13. Vrhovsek U, Eder R, Wendelin S. The occurrence of *trans*–resveratrol in Slovenian red and white wines. Acta Aliment 24:203–212 (1995).
14. Waterhouse AL, Lamuela-Raventos RM. The occurrence of piceid, a stilbene glucoside, in grape berries. Phytochemistry 37:571–573 (1994).
15. Romero-Perez AI, Lamuelaraventos RM, Buxaderas S, Delatorreboronat RC. Resveratrol and piceid as varietal markers of white wines. J Agric Food Chem 44:1975–1978 (1996).
16. Roggero J-P, Archier P. Quantitative determination of resveratrol and of one of its glycosides in wines. Sci Aliments 14:99–107 (1994).
17. Roggero JP, Garciaparrilla C. Effects of ultraviolet irradiation on resveratrol and changes in resveratrol and various of its derivatives in the skins of ripening grapes. Sci Aliments 15:411–422 (1995).
18. Creasy LL, Coffee M. Phytoalexin production potential of grape berries. J Am Soc Hort Sci 113:230–234 (1988).
19. Sbaghi M, Jeandet P, Faivre B, Bessis R, Fournioux JC. Development of methods, using phytoalexin (resveratrol) assessment as a selection criterion to screen grapevine *in vitro* cultures for resistance to grey mould (*Botrytis cinerea*). Euphytica 86:41–47 (1995).
20. Soleas GJ, Diamandis EP, Goldberg DM. Resveratrol: a molecule whose time has come? and gone? Clin Biochem 30:91–113 (1997).
21. Stark-Lorenzen P, Nelke B, Hänssler G, Mühlbach HP, Thomzik JE. Transfer of a grapevine stilbene synthase gene to rice (*Oryza sativa* L). Plant Cell Rep 16:668–673 (1997).
22. Rakwal R, Hasegawa M, Kodama O. A methyltransferase for synthesis of the flavanone phytoalexin sakuranetin in rice leaves. Biochem Biophys Res Commun 222:732–735 (1996).
23. Kodama O. Biochemical studies of rice phytoalexins. Mycotoxins 42:7–11 (1996).
24. Goossens JF, Stabel A, Vendrig JC. Relationships between kievitone and phaseollin accumulation in different tissues of *Phaseolus vulgaris* in response to treatment with mercuric chloride, a fungal cell wall elicitor and abscisic acid. Physiol Mol Plant Pathol 30:1–12 (1987).
25. Stössel P, Magnolato D. Phytoalexins in *Phaseolus vulgaris* and *Glycine max* induced by chemical treatment, microbial contamination and fungal infection. Experientia 39:153–154 (1983).
26. Liu L, Punja ZK, Rahe JE. Effect of *Pythium* spp and glyphosate on phytoalexin production and exudation by bean (*Phaseolus vulgaris* L.) roots grown in different media. Physiol Mol Plant Pathol 47:391–405 (1995).
27. Ishikawa T, Suzukawa M, Ito T, Yoshida H, Ayaori M, Nishiwaki M, Yonemura A, Hara Y, Nakamura H. Effect of tea flavonoid supplementation on the susceptibility of low-density lipoprotein to oxidative modification. Am J Clin Nutr 66:261–266 (1997).
28. Hertog MGL, Hollman PCH, Katan MB. Content of potentially anticarcinogenic flavonoids of 28 vegetables and 9 fruit commonly consumed in The Netherlands. J Agric Food Chem 40:2379–2383 (1992).
29. Watzl B, Leitzmann C. Bioaktive Substanzen in Lebensmitteln. Stuttgart:Hippokrates Verlag, 1995.
30. Dakora FD, Phillips DA. Diverse functions of isoflavonoids in legumes transcend anti-microbial definitions of phytoalexins. Physiol Mol Plant Pathol 49:1–20 (1996).
31. Nigg HN, Nordby HE, Beier RC, Dillman A, Macias C, Hansen RC. Phototoxic coumarins in limes. Food Chem Toxicol 31:331–335 (1993).
32. Diawara MM, Trumble JT. Linear furanocoumarins. In: Plant and Fungal Toxicants (Felix D'Mello JP, ed). Boca Raton, FL:CRC Press, 1997;175–189.
33. Anonymous. Furocoumarins in Plant Food-Exposure, Biological Properties, Risk Assessment and Recommendations. Copenhagen: Nordic Council of Ministers, 1996.
34. Surico G, Varvaro L, Solfrizzo M. Linear furocoumarin accumulation in celery plants infected with *Erwinia carotovora* pv. *carotovora*. J Agric Food Chem 35:406–409 (1987).
35. Afek U, Aharoni N, Carmeli S. The involvement of marmesin in celery resistance to pathogens during storage and the effect of temperature on its concentration. Phytopathology 9:1033–1036 (1995).
36. Zobel AM, Crellin J, Brown SA, Glowniak K. Concentrations of furanocoumarins under stress conditions and their histological localization. Acta Hortic Wageningen 381:510–516 (1994).
37. Beier RC, Oertli EH. Psoralen and other linear furocoumarins as phytoalexins in celery. Phytochemistry 22:2595–2597 (1983).
38. Ashkenazy D, Kashman Y, Nyska A, Friedman J. Furocoumarins in shoots of *Pituranthos triradiatus* (Umbellieferae) as protectants against grazing by hyrax (Procavidea: *Procavia capensis syraca*). J Chem Ecol 11:231–239 (1985).
39. Berenbaum M. Toxicity of a furanocoumarin to armyworms: a case of biosynthetic escape from insect herbivores. Science 201:532–534 (1978).
40. Berenbaum M. Coumarins and caterpillars: a case for evolution. Evolution 37:163–179 (1983).
41. Soleas GJ, Diamandis EP, Goldberg DM. Wine as a biological fluid: history, production, and role in disease prevention. J Clin Lab Anal 11:287–313 (1997).
42. Löffler G, Petrides PE. Biochemie und Pathobiochemie. Berlin:Springer, 1997.
43. Fitzpatrick DF, Hirschfield SL, Coffey RG. Endothelium-dependent vasorelaxing activity of wine and other grape products. Am J Physiol 265:H774–H778 (1993).
44. Pierpoint WS. Flavonoids in the human diet. In: Plant Flavonoids in Biology and Medicine: Biochemical, Pharmacological, and Structure-activity Relationships (Cody V, Middleton EJ, Harborne JB, eds). New York:Alan R. Liss, 1986;125–140.
45. Formica JV, Regelson W. Review of the biology of quercetin and related bioflavonoids. Food Chem Toxicol 33:1061–1080 (1995).
46. Fujiki H, Suganuma M, Okabe S, Komori A, Sueoka E, Sueoka N, Kozu T, Sakai Y. Japanese green tea as a cancer preventive in humans. Nutr Rev 54:67–70 (1996).
47. Nakachi K, Imai K, Suga K. Epidemiological evidence for prevention of cancer and cardiovascular disease by drinking green tea. In: Proceedings of International Conference on Food Factors: Chemistry and Cancer Prevention (Osawa T, ed). Tokyo:Springer Verlag, 1997;105–108.
48. Brown JP. A review of the genetic effects of naturally occurring flavonoids, anthraquinones and related compounds. Mutat Res 75:243–277 (1980).
49. Wagstaff DJ. Dietary exposure to furocoumarins. Regul Toxicol Pharmacol 14:261–272 (1991).
50. Ljunggren B. Severe phototoxic burn following celery ingestion. Arch Dermatol 126:1334–1336 (1990).
51. Anonymous. British photodermatology group guidelines for PUVA. Br J Dermatol 130:246–255 (1994).
52. Lydon J, Duke SO. Pesticide effects on secondary metabolism of higher plants. Pestic Sci 25:361–374 (1989).
53. Lydon J, Duke SO. The role of pesticides on host allelopathy and their effects on allelopathic compounds. In: Pesticide Interactions in Crop Production. Beneficial and Deleterious Effects (Altman J, ed). Boca Raton, FL:CRC Press, 1993; 37–56.
54. Hoagland RE. Chemical interactions with bioherbicides to improve efficacy. Weed Technol 10:651–674 (1996).
55. Kömives T, Casida JE. Acifluorfen increases the leaf content of phytoalexins and stress metabolites in several crops. J Agric Food Chem 31:751–755 (1996).
56. Cosio EG, Weissenböck G, McClure JW. Acifluorfen-induced isoflavonoids and enzymes of their biosynthesis in mature soybean leaves. Plant Physiol 78:14–19 (1985).
57. Swift MJ, Heal OW, Anderson JM. Decomposition in Terrestrial Ecosystems. Oxford:Blackwell Scientific, 1998.
58. Davidson E, Daniel O, Emeis K-C, Largeau C, Lee C, Mopper K, Oades JM, Reeburgh WS, Schimel DS, Zepp RG. What are the physical, chemical, and biological processes that control the formation and degradation of nonliving organic matter? In: The Role of Nonliving Organic Matter in the Earth's Carbon Cycle (Zepp RG, Sonntag C, eds). Chichester:John Wiley & Sons, 1995;305–324.
59. Kohli L, Daniel O, Schönholzer F, Hahn D, Zeyer J. *Miscanthus sinensis* and wild flowers as food resources of *Lumbricus terrestris* L. Appl Soil Ecol 334:1–9 (1998).