Plant Secondary Compounds

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In the plant cell, all the enzyme-catalysed transformation processes that allow and regulate growth, development and reproduction are summarized as metabolism. This process can be divided into anabolic and catabolic metabolism. In anabolism, smaller cell components are built up or assimilated to give more complex molecules. Important examples for anabolic processes are carbon fixation or fatty acid synthesis. During catabolism, organic material is broken down (dissimilation), e.g. during glycolysis, respiration or fatty acid beta-oxidation. Other examples of catabolism include the breakdown of polysaccharides to monosaccharides or the breakdown of nucleic acids to nucleotides. Plants also use catabolic processes to release previously stored energy, metabolic intermediates or nutrients for new anabolic reactions. Thus, metabolism comprises the energy exchange between energy-producing catabolic processes and energy-consuming anabolic processes (Bresinsky et al. 2013).

3.1 Primary Metabolites

Substances produced by the plant can be grouped into primary and secondary compounds, the so-called metabolites (Bresinsky et al. 2013). Before dealing with secondary metabolites, which are the focus of this textbook, primary compounds will be briefly highlighted in this chapter because an understanding of primary metabolism is essential for an understanding of secondary metabolism. *Primary compounds* are necessary for the growth and the propagation of plants, as these compounds are involved in overall cellular maintenance. These metabolites include carbohydrates, lipids and proteins and are found in relatively large quantities in almost all organisms and cells in which they fulfil important physiological functions (Yang et al. 2018). Most importantly, they ensure basic cellular homeostasis. Homeostasis can be understood as the capacity of a system in which parameters are steered in a way such that internal conditions remain stable and relatively constant (Torday 2015). The metabolic processes by which compounds (metabolites) are synthesized and broken down are called pathways. Remarkably, primary compounds and their metabolic pathways are relatively well conserved in all living organisms (McMurry and Begley 2016).

Among the primary metabolites, *carbohydrates* serve in plants for the storage and transport of energy (e.g. starch and sucrose) and as structural elements (e.g. cellulose). They are also part of molecules that carry genetic information (RNA and DNA) or are part of coenzymes (e.g. ATP). They consist of carbon, hydrogen (which is why we call them carbohydrates) and oxygen (Berg et al. 2015).

Proteins also have many functions. They serve, for example, in the replication of DNA, in the catalysis of metabolic processes, in cell signalling, in immune responses and in transport processes within the plant. The formation of proteins takes place in the cytoplasm, in the endoplasmic reticulum and in the Golgi, where they are synthesized from amino acids, their sequence being genetically determined. Biochemically, amino acids consist of amino group(s), carboxyl group(s) and a variable side chain. After being synthesized, enzymatically active proteins in plant cells are permanently degraded and replaced, and their amino acids are reutilized in order to adapt to the relevant development stages and any new environmental conditions (Berg et al. 2015).

Lipids are molecules consisting of hydrocarbon chains. They store energy, are involved in cell signalling and are components of cell and organelle membranes. The several categories of lipids include fatty acids and triglycerides. In most instances, the relevant metabolic pathways take place in the cytoplasm and endoplasmic reticulum in the case of triglycerides and in the plastids in the case of fatty acids (Bresinsky et al. 2013).

3.2 Secondary Metabolites

Secondary compounds (or *metabolites*) fulfil many ecophysiological functions. More than 200,000 secondary compounds have been identified to date (Wink 2016). However, they are less relevant for facilitating cellular homeostasis than primary metabolites. With regard to their chemical structure, secondary metabolites are highly diverse. The various plant species contrast in their metabolic composition, and, thus, species are characterized by a typical spectrum of chemical molecules. Whereas some secondary compounds are always present (constitutive), others depending on the individual species are elicited by certain biotic or abiotic environmental factors (▶ Chap. 3). Secondary compounds are usually found in small concentrations far below 1% of the dry weight of a plant (Akula and Ravishankar 2011). In principle, a tradeoff occurs between primary and secondary metabolism. First, primary metabolic activity is reduced when the production of secondary compounds is induced, e.g. by a stressful environment. This is not surprising given the fact that the production of secondary compounds requires substantial resources such as energy and metabolic precursors. Second, the enzymes that build up the metabolites have to be produced, and this alone consumes energy and precursors such as amino acids. Third, secondary metabolites have to be translocated from source (cell organelles of synthesis or storage) to sink (tissue of usage) organs (Wink 2010). Furthermore, specific storage organs for secondary metabolites have to be formed.

From a plant perspective, secondary compounds present a strategy to react fast and flexibly to various environmental cues (e.g. stresses). They may serve as attractants or repellents, inhibit herbivores, function as antimicrobial agents, provide a shield from excess light or act as inhibitors against other competing plant species (allelopathic function, Wink 2015a; please refer to ▶ Chap. 16). Secondary compounds are frequently relevant for plant fitness. Plants including hydrophytes have developed defence strategies in which secondary metabolites play a paramount role. After an infection, plants produce antibacterial or antifungal compounds and fortify their cell walls by lignin (Malinovsky et al. 2014). Often, secondary metabolites operate not only cumulatively but also synergistically, forming powerful chemical protection against pathogens. Therefore, it is very challenging for pathogens including viruses, bacteria, fungi and herbivores to become chemically resistant (Wink 2015a). Domesticated crops such as rapeseed are manipulated by breeding activities in such a way that they synthesize and accumulate less of a certain secondary compound, e.g. the bitter substance sinapine, because of its repellent taste or other properties that livestock does not appreciate (Bhinu et al. 2008). A disadvantage is that these crops lose their initial self-protective properties; this goes hand in hand with the necessity to use chemical pest management.

In general, and from a human perspective, plant secondary metabolites are extremely important. In agriculture, they are applied as biopesticides because of their antiviral, insecticide, fungicide and herbicide properties (Gutzeit and Ludwig-Müller 2014). In medicine, e.g. phytotherapy, they are appreciated for their anesthetic, antioxidant, anti-inflammatory, antibacterial, antidepressive, antiviral, relaxing or digestive functions (Table 3.1; Wink 2015b). Furthermore, plant secondary compounds are used as flavouring agents, fragrances, colourants, artificial sweeteners and hallucinogens (Seigler 1998; Erdogan Orhan 2012). However, certain substances may harm human health, e.g. atropine is toxic, coumarin is carcinogenic, furanocoumarin is allergenic, and pyrrolizidine alkaloids are hepatotoxic (Table 3.2; Neuman et al. 2015). Throughout human

Table 3.1 Properties and bioactivity of selected secondary metabolites that are applied as isolated compounds in medicine

Plant species	Substance (class)	Properties/applications
Aconitum napellus	Aconitine (A)	Analgesic
Atropa belladonna	L-hyoscyamine (A)	Parasympathomimetic
Campthotheca acuminate	Camptothecin (A)	Tumour therapy
Cannabis sativa	Tetrahydrocannabinol (T)	Analgesic
Catharanthus roseus	Dimeric Vinca alkaloids (A)	Tumour therapy
Chondrodendron tomentosum	Tubocurarine (A)	Muscle relaxant
Cinchona pubescens	Quinidine (A)	Antiarrhythmic
Coffea arabica	Caffeine (A)	Stimulant
Colchicum autumnale	Colchicine (A)	Gout treatment
Crotalaria	Pyrrolizidine (A)	Hepatic veno-occlusive disease
Cytisus scoparius	Sparteine (A)	Antiarrhythmic
Digitalis lanata	Digitoxin, digoxin (A)	Heart insufficiency therapy
Erythroxylum coca	Cocaine (A)	Analgesic, stimulant
Galanthus woronowii	Galantamine (A)	Alzheimer's disease treatment
Lycopodium clavatum	Huperzine (A)	Alzheimer's disease treatment
Papaver somniferum	Morphine (A)	Analgesic, hallucinogen
Physostigma venenosum	Physostigmine (A)	Alzheimer's disease treatment
Pilocarpus jaborandi	Pilocarpine (A)	Glaucoma treatment
Rauvolfia serpentina	Reserpine (A)	Hypertonia treatment
Sanguinaria canadensis	Sanguinarine (A)	Antibacterial, antiviral
Strophanthus gratus	Ouabain (T)	Heart insufficiency therapy
Taxus brevifolia	Paclitaxel (taxol) (A)	Tumour therapy

Modified after Wink (2015b); Van Wyk and Wink (2015, 2017); Van Wyk et al. (2015); Wagner et al. (2007)
A alkaloid, *T* terpenoid

Table 3.2 Estimated number of described secondary metabolites and their main functions in plants

Class	Estimated numbers of structures	Toxic or repellent for herbivores	Antimicrobial activity	Attraction of pollinators or fruit dispersers
With nitrogen				
Alkaloids	27,000	++++	++	–
Nonprotein amino acids	700	++++	+++	–
Cyanogenic glucosides/HCN	60	++++	+	–
Glucosinolates	150	++++	++++	–
Amines	100	+++	+	+++
Lectins, peptides, AMPs	2000	+++	+++	–
Without nitrogen				
Terpenes				
Monoterpenes (incl. iridoid glucosides)	3000	++	+++	+++
Sesquiterpenes	5000	+++	+++	++
Diterpenes	2500	+++	+++	–
Triterpenes, steroids, saponins	5000	+++	+++	–
Tetraterpenes	500	+	+	+++
Phenols				
Phenylpropanoids, coumarines, lignans	2000	+++	+++	++
Flavonoids, anthocyanins, tannins	4000	+++	+++	++
Polyketides (anthraquinones)	800	++++	+++	–
Others				
Polyacetylenes	1500	++++	++++	–
Carbohydrates, organic acids	600	+	++	–

Modified after Wink (2015b); Van Wyk and Wink (2017); Van Wyk et al. (2015)
Activity: – no or very few secondary metabolites (SM) active; + few SM active; ++ many SM active; +++ most SM active; ++++ all SM active

history, horticulturists have therefore tried to improve not only the quantity and quality of primary plant metabolites but also the properties of secondary compounds.

Another major function of secondary metabolites for flowering plants is to attract pollinators and other animals that disseminate their seeds. For this purpose, plants produce colourants in petals (e.g. anthocyanins, carotenoids) or volatile fragrances in blossoms (e.g. terpenes, amines). In order to prevent pollinators eating the entire flower, plants produce nectar, pollen or essential oils as a reward. For the promotion of the dispersal of seeds, plants produce fruits that contain secondary metabolites that confer certain colours, fragrances and flavours to attract animals that eat and thus disperse the seeds.

Another feature of importance is that plants synthesize secondary metabolites, e.g. antioxidant agent, in order to protect themselves against abiotic stress (e.g. heat, drought, UV radiation). Secondary metabolism is dynamic and flexible: upon stress, namely, attacks or infection, molecular cues (themselves being secondary metabolites) induce plants rapidly to increase the production of the necessary compounds. Production, storage and transport within the plant are energy-consuming and require considerable amounts of ATP or reduction equivalents, thus decreasing growth and development, explaining the tradeoff between primary and secondary metabolism as explained above (Züst and Agrawal 2017).

Many secondary compounds are synthesized in the cytoplasm of the cell. However, they can also be produced in chloroplasts (some terpenes and some alkaloids), in mitochondria (some amines, a few alkaloids) or in vesicles (Wink 2015a). Plants not only build up secondary compounds upon stimulation but also store them in considerable amounts in their vacuoles, in which they fulfil their function (e.g. vacuolic anthocyanins act as pigment to shield the cell from solar radiation) or where they can be released when they are needed (e.g. vacuolic glucosinolates are needed during herbivore attack). The site of storage of a compound also depends on its polarity. Hydrophilic compounds can be sequestered and stored in water-based compartments (e.g. the vacuole), whereas lyophilic compounds cannot. Lipophilic compounds such as terpenes are stored in specific cells, in small oil reservoirs or in the cuticle or in trichomes (Wink 2015a). Papaveraceae (poppy flowers) and many Euphorbiaceae (surge flowers) plants, which produce lactiferous compounds that include toxic alkaloids, sesquiterpenes or diterpenes, store them in particular tubes, called laticifers (Wink 2015a). Plants manufacture and store specific blends of secondary metabolites derived from various groups of compounds. Even within a single plant, one organ may contain a compound mix differing from that of another. The metabolite composition differs also according to the plant's developmental stage (e.g. germination or flowering stage) and among or within populations of the same species (Wink 2015a).

The most widespread groups of secondary metabolites are phenols, terpenoids and alkaloids:

- *Phenols* are characterized by several phenolic rings and phenolic OH groups (Wink 2015b). Several metabolic pathways lead to phenols, including the shikimate pathway, the acetate-malonate pathway and the terpenoid synthetic pathway (▣ Fig. 3.1) (Wink 2010; Crozier et al. 2006). According to the number of carbon atoms, the most important subgroups of phenols are:
  - *Coumarines*: these phenols are fragrant with a sweet odour and naturally found in many plants, particularly in the Tonka bean (*Dipteryx odorata*), vanilla grass

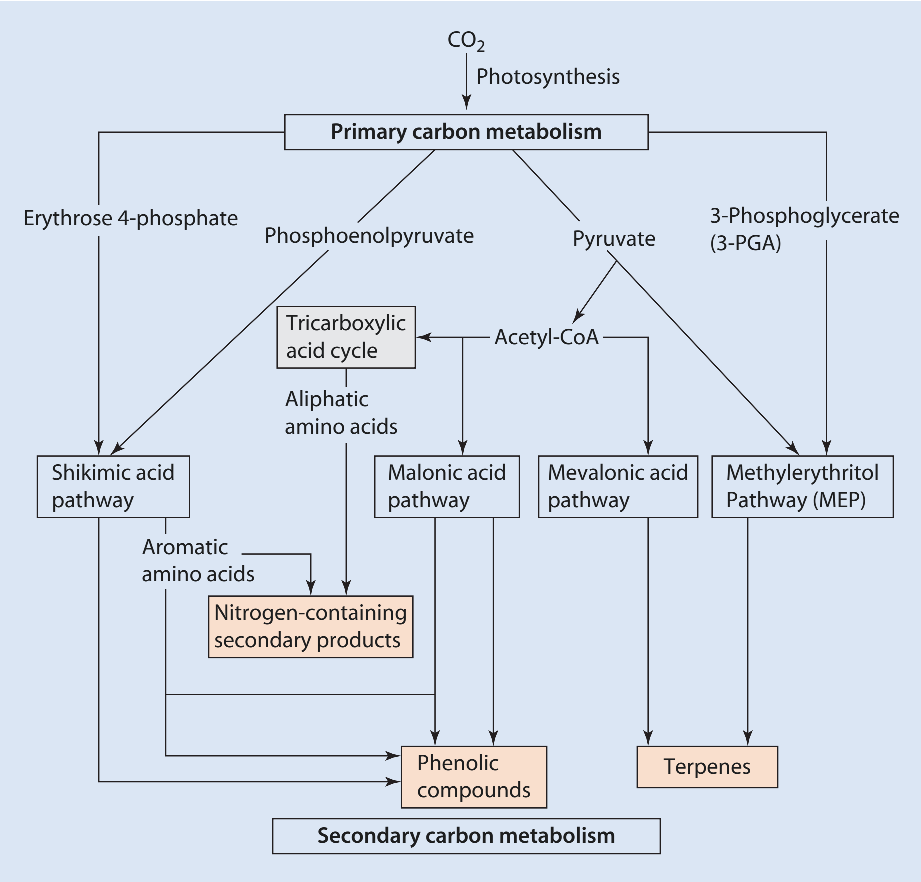

Fig. 3.1 A simplified general overview of the biosynthetic pathways involved in the biosynthesis of secondary metabolites showing a tight association with the product of primary/central metabolism. *Pink boxes* represent secondary metabolites, whereas primary metabolites are given without a frame. The pathways in *unshaded boxes* represent secondary metabolism and that shaded *grey* is part of primary metabolism (most not shown). (Figure taken from Ncube and van Staden (2015). © 2015 by Ncube and van Staden; licensee MDPI, Basel, Switzerland. Open access article distributed under the terms and conditions of the Creative Commons Attribution license (▶ http://creativecommons.org/licenses/by/4.0/))

(*Anthoxanthum odoratum*), sweet woodruff (*Galium odoratum*), sweet grass (*Hierochloe odorata*) and cassia cinnamon (*Cinnamomum cassia*). Furanocoumarins (e.g. celery, parsley) have to be activated by UV-A light (320–400 nm) in order to be toxic for plant pathogens (Hänsel and Sticher 2010; Petersen et al. 2010).

- *Flavonoids and anthocyanins*: flavonoids appear in many plants and fulfil important functions. Flavonoids are (or are precursors for) yellow, red and purple plant pigments for flower coloration and serve in the attraction of pollinators. They are also involved in UV filtration and symbiotic nitrogen fixation and provide protection against oxidation. Anthocyanins (glycosides of anthocyanidin) are water-soluble vacuolar pigments in leaves (red cabbage), flowers (e.g. roses, delphiniums, corn cockles, begonias) and fruits (e.g. apple) and, sometimes, in roots (balsams). Among the best known flavonoids are quercetin and kaempferol (Hänsel and Sticher 2010).

  - *Lignins*: they stabilize the tissues of vascular plants and algae and support wood formation. Upon infection by pathogens or wounding by feeding insects, additional lignification is induced in plants followed by a thickening of the cell walls through the accumulation of lignin (Hänsel and Sticher 2010; Lattanzio et al. 2006).
  - *Polyphenols*: these occur in most plant families and are often concentrated in leaf tissue, the epidermis, bark layers, flowers and fruits. They may release and suppress growth hormones, deter herbivores and prevent microbial infections and may function as signalling molecules (Hänsel and Sticher 2010).

- *Terpenes* are derived from five-carbon isoprene units by biosynthesis from isopentenyl pyrophosphate and are modified in multiple ways. They are produced in the acetate-mevalonate pathway or the non-mevalonate pathway: MEP/DOXP (2-C-methyl-D-erythritol 4-phosphate/1-deoxy-D-xylulose 5-phosphate) (Hänsel and Sticher 2010). They form a large class of naturally occurring organic substances. Cytokinins, major plant hormones involved in cell growth and differentiation, belong to the terpenoids (please refer to ▶ Chap. 15). Isoprenes are thought to protect the photosynthetic membranes from heat damage (Hänsel and Sticher 2010; Ashour et al. 2010).
  - *Monoterpenes* are substantial components of ethereal oils, which attract or repel insects, and are characterized by their aromatic smell. They are synthesized in multiple plant species, e.g. in the Asteraceae (sunflower family), Apiaceae (celery family), Lamiaceae (mint family), Myristicaceae (nutmeg family) and Poaceae (grasses) and are present in the resins of conifers (Wink 2015b).
  - An important subgroup of terpenoids are the *saponins*. Because of their detergent properties, they are toxic, particularly to fish. Historically, humans used them as soaps. In plants, saponins are found in leaves, stems, bulbs, roots, blossoms and fruits. They are present in several monocot and dicot families including the Amaranthaceae (amaranth) and Sapindaceae (soapberry) families. In plants, they serve as antifeedants and protection against microorganisms by damaging their membranes (Hänsel and Sticher 2010).
  - *Tetraterpenes* are needed for the biosynthesis of carotenoids. Carotenoids are dominant pigments in flowers (e.g. violaxanthin in viola) and in fruits (the red pigment of tomato, lycopene) and appear in other organs (e.g. ß-carotene in the taproot of carrots). Carotenoids absorb light energy for photosynthesis, protect chlorophyll from radiation damage and serve as antioxidants (Hänsel and Sticher 2010). Other well-known terpenoids are the cannabinoids synthesized in cannabis plants (Kinghorn et al. 2017), the carotenes and xanthophylls and the essential oils synthesized, for example, by peppermint, chamomile and eucalyptus (Lüttge and Kluge 2012).

- *Alkaloids*: with 27,000 substances, this is the largest group of identified secondary plant compounds (Wink 2015b). Alkaloids are particularly frequent in the

Solanaceae (nightshades), Papaveraceae (poppies), Apocynaceae (dogbanes) and Ranunculaceae (buttercups or crowfeet) families. They are characterized by heterocyclically bound nitrogen and are synthesized from amino acids. Alkaloids mostly taste bitter. They are usually toxic, a feature that protects the plant against herbivory attacks. Upon attack by pathogens, plants might synthesize these antimicrobial compounds. Plants produce alkaloids mainly in their leaves, fruits, seeds, roots or bark, although different parts of the plant may contain different alkaloids.

  - The main alkaloid subgroups are the tropanes (e.g. atropine, cocaine), pyridines (e.g. nicotine), isoquinoline (e.g. morphine, codeine), purine (e.g. caffeine) and colchicine. Alkaloids include morphine, mescaline and cocaine, which provoke specific effects on the nervous systems of animals and humans (Roberts et al. 2010). Other alkaloids (e.g. caffeine, nicotine) show stimulating effects. Because of their pharmacological properties, they are used in traditional and modern medicine. Such properties include antimalarial, anticarcinogen, analgesic, antibacterial and antiarrhythmic effects (Hänsel and Sticher 2010).

- *Glycosides* are 'originally mixed acetals resulting from the attachment of a glycosyl group to a non-acyl group RO- (which itself may be derived from a saccharide and chalcogen replacements thereof (RS-, RSe-)' (IUPAC 2014). Their functions in plants are related to detoxification processes and protection against herbivory (Brito-Arias 2016). Because of their membrane-damaging property, glycosides may be toxic to bacteria and fungi. An important subgroup is the glucosinolates, which are composed of specific amino acids (Selmar 2010). They contain sulphur and nitrogen and are found particularly in the Brassicaceae (e.g. cabbage, horseradish, mustard), Capparidaceae (e.g. caper) and Tropaeolaceae (e.g. garden nasturtium). Typical is the spicy smell and taste of mustard oil. Increasing indications suggest that they protect people from colon cancer (Schneider et al. 2017).
- *Plant hormones* (also called phytohormones) are extremely important secondary metabolites. They serve as signal molecules, regulate gene expression, control cellular processes and determine the formation of major plant organs (Gray 2004; Depuydt and Hardtke 2011). Moreover, they are involved in stress responses. Plants usually produce them in low concentrations. The most important phytohormones include the abscisic acids (ABA), auxins, cytokinins, ethylene and gibberellins. However, Taiz et al. (2018) group phytohormones as primary metabolites, since all plants require them for growth and development. In general, phytohormones are derivatives of secondary metabolite pathways, except for the auxins and ethylene whose precursors are synthesized in primary metabolism: A more detailed review on the role of phytohormones in CEH is given in ▶ Chap. 15.

3.2.1 Improving Quality by Adjusting Metabolites Through the Regulation of Controlling Environmental Factors

Although the metabolome is predetermined by the genetic background of the plant, metabolism is a dynamic process and is not fixed. This is because during growth and development, plants are permanently adapting to their changing environment. Thus, the metabolomic composition is dynamic in terms of quality (this means in both the pattern and biological properties of the metabolites) and quantity (Gorelick and Bernstein 2014). Environmental factors contribute highly to changes in the secondary metabolome (Gorelick and Bernstein 2017; Yang et al. 2018). These factors include light (see ► Chap. 5), nutrient deficiency (see ► Chap. 6), salinity (see ► Chap. 7), water availability (see ► Chap. 8), temperature (see ► Chap. 9) or wounding (see ► Chap. 10). Apart from these physical stressors, hormonal elicitors also exert their effects, such as jasmonic acids, salicylic acids, brassinosteroids, abscisic acids and auxins, and inorganic chemical elicitors in the form of heavy metals.

Definition of Metabolome

A metabolome is a set of small molecules within a biological organism (e.g. cell, organ, tissue). A metabolome can include primary and secondary metabolites and substances not necessarily produced naturally by organisms, e.g. toxins. Very small molecules, including metabolites, are part of the metabolome. Macromolecules, such as DNA and RNA, are not.

Moreover, also biotic stressors, e.g. pathogens, fungi or insects, elicit the production of specific secondary compounds in plants. After exposure of a plant to stressors, enzymatic pathways are induced that alter the content of bioactive secondary compounds, namely, alkaloids, terpenoids and phenylpropanoids. By regulating controllable stressors, growers can shift metabolism towards the accumulation of favourable compounds. However, an important point to note is that plants might reduce their production of primary metabolites when stress factors induce the production of secondary compounds. This might result in a reduction of the biomass, an effect that has to be avoided in horticultural production as crops need to be marketable. Stress exposure must be strong enough to adjust the metabolism towards compounds that are favourable in the human diet but, at the same time, must be so mild that biomass and yield formation is not reduced. This is the challenge for the horticulturist.

Since the production of secondary metabolites often depends on the physiological and developmental stage of the plant and on environmental conditions, the time of harvest is of great importance, as is the postharvest treatment (e.g. drying technology and storage conditions) (Ncube et al. 2012). In order to avoid yield losses, it makes sense to start the controlled stress treatment shortly before harvest, by which time yield has been set (Schreiner et al. 2003; Pareek 2017). Usually, the desired secondary compounds accumulate within hours and days. In other words, the crop can be gently stressed to induce the synthesis of favourable secondary compounds, for example, at 1 or 2 days before harvest. However, this cannot be generalized, and case studies and metabolite-specific strategies are introduced in this textbook.

The reader is warned that the initial effect might be different, if an individual stressor interacts with other factors. For example, high irradiation often accompanies elevated temperature and drought stress (Selmar and Kleinwächter 2013). Thus, an advantageous ploy might sometimes be to enrich secondary compounds under totally controlled conditions. This is particularly the case with medicinal and pharmaceutical plants for which market requirements have to be fulfilled extremely precisely (Naik and Al-Kharyri 2016), e.g. in prescription medicines containing cannabinoids (Potter 2013). In addition, even if environmental stressors can be well controlled, certain substances might nevertheless operate differently in isolated conditions than when acting together with other substances in the same plant (Bhatia and Bera 2015).

Next, we show an example of the way that pharmaceutically active secondary compounds, in particular cannabinoids, can be enriched in the medicinal plant cannabis (*Cannabis sativa* L.) by inducing controlled stress. Cannabis belongs to the Cannabaceae family and is an annual and dioecious plant (male and female flowers are sited on separate plants). After the pollination of female flowers, the male plants die (Flores-Sanchez and Verpoorte 2008). Several native species exist in Central Asia, but, nowadays, they are spread all over the world. They are a source of food, energy and fibre, and several components of the plant are used medicinally or pharmaceutically.

During the flowering period, cannabis plants produce many valuable unique metabolites including cannabinoids, terpenes and phenolic compounds. They protect the developing flowers from insects (sticky resinous oils and volatiles) and from excessive heat under shifting solar conditions. The predominant cannabinoids are Δ9-tetrahydrocannabinolic acid (THCA), cannabidiolic acid (CBDA) and cannabinolic acid (CBNA), followed by cannabigerolic acid (CBGA), cannabichromenic acid (CBCA) and cannabinodiolic acid (CBNDA) (Hazekamp et al. 2010; ElSohly et al. 2017). They are found in the secretory cavity of the glandular trichomes.

These acids are decarboxylated in the living plant, a process that is particularly induced upon heating, e.g. after harvest (Flores-Sanchez and Verpoorte 2008). Only thereafter can THC unfold its psychoactive properties (André et al. 2016). The concentrations of secondary metabolites depend on tissue type, age, variety, growth conditions (nutrition, humidity and light levels), harvest time and storage conditions (André et al. 2016). Their impacts on humans are observed as psychotropic, antinociceptive, antiepileptic, cardiovascular, immunosuppressive, antiemetic, appetite stimulating, antineoplastic, antimicrobial, anti-inflammatory and neuroprotective. Positive effects in psychiatric syndromes, such as depression, anxiety and sleep disorders, are well described (Kinghorn et al. 2017; Musty 2004; Cascio et al. 2017; Pertwee 2014). The precursors of cannabinoids are synthesized from the deoxyxylulose phosphate/methylerythritol phosphate (DOXP/MEP) pathway and the polyketide pathway (Flores-Sanchez and Verpoorte 2008; André et al. 2016).

Whereas, in general, the outdoor cultivation of cannabis is limited to one harvest per year, three to four crops are possible under controlled environment conditions (Thomas and ElSohly 2016). For indoor cultivation, protocols are available for horticulturists: with regard to photo-radiation, cannabis prefers high photosynthetic photon flux densities (≈ 1500 μmol/m^2/s) in order to exchange gas and water vapour efficiently between leaves and their surroundings. Several lamp types can be used, including high-pressure sodium (HPS) lamps and light-emitting diodes LEDs. During vegetative growth, an 18-h photoperiod is recommended, which is reduced to 12 h for the evocation of flowering. According to Gorelick and Bernstein (2017), UV-B light increases the THC content.

The amount, type and quality of cannabinoids rely on genetic background and can be induced by changing stressful conditions. ◘ Table 3.3 summarizes the effects of elicitors on cannabinoid production.

Table 3.3 Effects of selected biotic and abiotic elicitors on the production of cannabinoids

Stressor	Elicitation	Induced effect
Nutrients	Increased content of N, Ca, Fe, Mg	Increase of THC content
	Nutrient deficiency because of poor soil condition	Increase of cannabinoid content
	P deficiency	Increase of THC content
Drought	Drought stress	Increase of trichome density and thereby increase of cannabinoid production
	Increased humidity	Increase of THC content
Temperature	Increased temperature	Conflicting results about effects on cannabinoid content
Photo-radiation	Increased irradiance	Increase of THC concentration due to its defensive role against UV radiation
	Increased intensities of UV-B radiation	Increase of THC without changes of other cannabinoids
	Increased UV-C radiation	No effect on cannabinoid production but increase of stilbenes and cinnamic acid derivatives
Metals	Moderate concentrations of Cd, Ni, Cr	Tolerance regarding plant growth and physiology and only slight effect on THC content
Wounding	Insect herbivory	Increases cannabinoid and terpene content due to their role as natural insecticide
Pathogens	Fungal (e.g. *Phomopsis ganjae*) and bacterial (e.g. *Staphylococcus aureus*) pathogens	Modulate cannabinoid biosynthesis attributable to their antibiotic and antifungal properties
Hormones	Jasmonic acid, methyl jasmonate, salicylic acid	Increase of secondary metabolite production in cannabis cell suspension culture but no change in cannabinoid content
	Abscisic acid	Decrease of THC and CBD in vegetative plants but increase of THC content in flowering female plants

Modified after Gorelick and Bernstein (2017)

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