CHAPTER

20

Botanical Ingredients

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20.1 INTRODUCTION

A wide variety of botanical ingredients are used in cosmetics, and their application is increasing year over year. Botanical substances have different characteristics like the following when compared to synthetic ingredients or other natural ingredients such as animal-derived ingredients and mineral ingredients: (1) The image of the products is positive since they are natural ingredients; (2) In general, there is a public perception that botanical ingredients are safe and are environmentally friendly due to their biodegradability and renewability; (3) Their effect is usually mild and sustained; and (4) When the ingredients are not used as single components, the subcomponents often help slightly soluble components to become easily soluble.

Botanical substances are used for various purposes in cosmetics, but they are mainly used for their effectiveness and have expanded remarkably in recent years. However, botanical ingredients are also frequently used as base materials to meet trends such as nonadditive cosmetics. When botanical ingredients are used as base materials, they are usually used as a substitute for synthetic materials with similar effects. For example, botanical oils such as olive oil and jojoba oil are frequently used as a substitute for mineral oils, and dyes of safflower, lithospermum root, or gardenia are often used to substitute for tar-based dyes. Additionally, soy isoflavone, botanical glycerin, and hyaluronate are used as moisturizers to avoid use of petrochemical surfactants and synthetic polymers. Furthermore, rosemary extract and hinokitiol are used as alternatives to synthetic preservatives, and essential oils are used as aromatic substances instead of petroleum-based synthetic aromatic substances.

Plant-derived ingredients do not necessarily mean that they are safe, as they can cause specific plant allergies or rashes. Since the effect is mild, their preservative efficacy may be weaker than synthetic preservatives, and since natural ingredients are not limited to plants, safe ingredients are often found with other natural ingredients like those derived from animals or natural mineral dyes. In this chapter, we will learn the current trends and challenges of botanical ingredient development in cosmetics by understanding characterisrutics of plant-derived substances.

20.2 BOTANICAL SUBSTANCES

20.2.1 The Diversity of Botanical Substances

Botanical substances are classified into primary metabolites and secondary metabolites. Primary metabolites are substances that are vital to preserve the organism, such as amino acids, sugars, and nucleic acids, and are chemical substances common in each organism in a taxonomic group. On the other hand, secondary metabolites are low-molecular compounds derived from primary metabolites and are not necessarily essential to an organism. The fundamental difference between primary metabolites and secondary metabolites are that secondary metabolites are unique to each organism. Unlike animals, plants do not have the ability to move by themselves, and thus have evolved to produce a plethora of secondary metabolites in order to adapt to their surrounding environment, mainly to provide vital defense functions against organic and inorganic stress. Humans consume primary metabolites of plants as nutrients, but the botanical substances that have been used for medicinal purposes are all secondary metabolites. It is believed that there are a quarter million species of higher plants in the world,[1] and the variety of secondary metabolites is

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abundant, as well as their chemical structure. There is even a theory that estimates over a million plant-derived secondary metabolites exist in the entire plant kingdom.[2] In other words, the botanical substances that have been used by humans are only a small fraction of these extremely diverse chemicals, and countless possibilities for use still remain.

20.2.2 The Characteristics of Botanical Substances as Ingredients

Unless they are purified to isolate a specific substance, botanical ingredients are mostly used as plant extracts or as dried ingredients. In this case, the main characteristic is that the ingredient is a multicomponent system. It is important to specifically define the plant, the part of the plant, and the treatment to assure stable supply of botanical ingredients. Caution is required when using botanical ingredients, since plants sold in the market can differ by region even though they have the same name, and the scientific name sometimes changes as studies in the region progress. Additionally, the individual chemical composition may change depending on the region, season, and/or climate, and the quality may differ even if the originating plants are well managed and the parts are specified. If possible, cultivated plants should be preferred over wild plants in order to control the ingredients under fixed conditions. Moreover, it is important to consider refinement and other treatment methods to remove components that can have a negative effect on the ingredients' safety and stability. Setting quality standards for each material and controlling the quality is a key point in botanical ingredients. Botanical ingredients show a variety of effects due to their unique multicomponent systems. The reason for the botanical ingredient's effect to work mildly and sustained is thought to be from the activity balance between the active component's effect and subcomponent's effect working subcutaneously. In other words, stability of standardized substances, like the active components and subcomponents, is essential. Standardization and unification of the compositions in botanical ingredients remains a vital task in botanical ingredients.

20.3 REGULATIONS REGARDING BOTANICAL SUBSTANCES

There are international regulations and documentation such as the Washington Convention and the Red List that are stipulated for protecting rare organisms, and these must be checked when acquiring ingredients. The Convention on Biological Diversity, on the other hand, acknowledges the economic value of organisms. This perspective is an important topic in the cosmetics industry, and in this section we will provide an overview of this convention along with a narrative history.

20.3.1 The Convention on Biological Diversity[3]

The Convention on Biological Diversity (CBD) was adopted in 1992 and was one of the international environmental agreements that was signed and opened at the United Nations Conference on Environment and Development (UNCED) held on May 22, 1992 in Rio de Janeiro.[4] This convention came into effect in 1993, and as of 2015, 195 parties excluding the United States have joined as participating countries. The international conference of this convention is called the Conference of Parties (COP) and is held every two years. (COP − CBD). The last conference was the COP12 held in Pyeong Chang (South Korea) in 2014, and the next COP13 is scheduled to be held in Mexico.

The objectives of this convention are: (1) to preserve the diversity of organisms and their habitats on Earth; (2) to use biological resources with sustainability; and (3) to share the profits and benefits of the resources equally and fairly. The third item, access and benefit sharing (ABS), is especially for acknowledging the sovereign rights of the origin countries of natural resources including genetic resources, rather than preserving the common international benefit, and this item is strongly influenced by the hosting country.

To ensure ABS, a voluntary guideline (the Bonn Guideline) was set in the CBD held in 2002 (COP6) as a reference for establishing domestic legislation or documenting contracts.[5] This guideline was set to clarify the standards of "prior informed consent" and "mutually agreed terms." However, the Bonn Guideline was a voluntary guideline and was not obligatory, and has been criticized that it is too weak for properly distributing the benefits and profits gained from genetic resources back to the country of origin. With this criticism, discussion began at the Johannesburg Summit (the World Summit on Sustainable Development) held that year to establish a binding "international regime"). There have been many negotiations since this summit, but there has also been strong conflict between the countries of origin and countries utilizing the genetic resources, or between developing countries and developed countries, and the negotiations have been slow. However, in 2010 this conflict was dramatically resolved politically at the COP10 held in Nagoya, and the Nagoya protocol on Access to Genetic Resources and the Fair and Equitable Sharing of Benefits Arising from their Utilization to the Convention on Biological Diversity was established, which came into effect in

2014.[6] Developed countries must be cautious about the domestic legislations of the genetic resource−providing countries. Currently, it is assumed that the signing countries are setting the rules regarding supply of genetic resources.

Developing countries claimed these five items to be included in the Nagoya protocol: (1) All products derived from the genetic resources should be subject to benefit sharing; (2) Traditional knowledge should be clearly stated as subject to benefit sharing; (3) The ABS rules should have retroactive provision and should be applicable to resources accessed prior to the protocol; (4) Fair and equal access and use of genetic resources should be monitored by the resource utilizing countries; and (5) Protest against the developed countries' request to exclude pathogenic organisms from ABS.

These claims were reflected to the Nagoya protocol as follows: (1) "Derivatives" was not explicitly stated in the items, but the term "utilization of genetic resources" was used and the scope of the protocol was stipulated as the "utilization of genetic resources." (2) A standard for defining "traditional knowledge" was not stipulated, but the handling of traditional knowledge was acknowledged as the same level of genetic resources, including benefit sharing in accordance with domestic legislation. (3) For retroactive provision, the protocol did not acknowledge benefit sharing for resources accessed prior to the protocol. (4) The protocol added an item that at least one checkpoint must be set for the monitoring of utilization of genetic resources, but the checkpoints were not defined. (5) The protocol did not define its scope with pathogenic organisms. In the Nagoya protocol, additional important topics were set, such as efforts in balancing the compliance of the country of origin and country of utilization, and establishing a clearing house regarding access and benefit sharing.

20.3.2 Actual Industrial Use and Issues of Genetic Resources

Since the economic situation differs with each industrial field, the utilization of genetic resources and benefit sharing also differs drastically. The cosmetics industry and health food industry are unique in how they require a stable supply of genetic resources in large volumes. Also, unlike the medical drug industry, the value of traditional knowledge tends to have more value. In general, the utilizing company would feel that the benefit sharing is complete if the ingredients are directly bought from the genetic resource's country of origin. However, the distribution channel of genetic resources follows a complex and diverse route from the country of origin and follows many stages, often going through many countries. In this case, an entity to oversee the entire distribution of these various routes is required in order to properly distribute the benefits and access. Furthermore, a stable supply of genetic resources cannot be obtained only from Mother Nature, and cultivation systems must be developed. Development of cultivation systems will create employment in the countries of origin and can be viewed as nonmonetary benefit sharing. Regardless of how the resources are distributed, regulations with countries of origin are expected to become stronger even in the cosmetics industry, so consulting experts on distribution including ethical aspects should be effective for businesses.[7−9]

20.4 ORGANIC COSMETICS

20.4.1 The "Natural Cosmetics" Trend

"Natural" became a global trend in cosmetics in the 1970s, starting in the United States, where cosmetics with plant-derived ingredients were differentiated and marketed as phytocosmetics or herbal cosmetics, and naturally derived ingredients were largely admired.[10] As an example, some cosmetics during this period used ginseng extract as one of their ingredients.[11] In Japan, luffa cylindrica and hamamelis (witch hazel) were used traditionally and were used as ingredients for skin toners, but in the 1970s, as the first "natural cosmetics" fad began, extracts of herbal ingredients such as aloe, camomile, and glycyrrhiza were added in cosmetics.

20.4.2 What is Organic?

After 25 years of studying agriculture in India, Albert Howard was the first person to propose organic agriculture, in his text *An Agricultural Testament* in 1940.[12] Organic agriculture is often confused with "pesticide-free" or "chemical-free," but is a concept of agriculture to preserve biodiversity by nurturing the circulation of resources and by maintaining the balance of the habitat. The US Department of Agriculture's National Organic Program is a reference for organic certification.[13]

20.4.3 Green Chemistry

The demand for organic cosmetics and natural cosmetics from consumers has become stronger, and so has the concern for global environmental protection. Sustainability, reducing environmental impact, and effective use of energy resources has become more and more important, and the value of botanical ingredients that are environmentally and human friendly are expected to increase. The COSMOS-standard[14] recommends the use of natural or naturally derived ingredients, especially organic agriculture-derived ingredients, and strongly suggests that the concept of environmentally friendly "green chemistry" is also adopted. There are many activities that are being promoted globally to accomplish a sustainable society. The following are the 12 Principles of Green Chemistry proposed by Dr. Anastas of the United States.[15]

1. It is better to prevent waste than to treat or clean up waste after it has been created.
2. Synthetic methods should be designed to maximize the incorporation of all materials used in the process into the final product.
3. Wherever practicable, synthetic methods should be designed to use and generate substances that possess little or no toxicity to human health and the environment.
4. Chemical products should be designed to affect their desired function while minimizing their toxicity.
5. The use of auxiliary substances (e.g., solvents, separation agents, etc.) should be made unnecessary wherever possible and innocuous when used.
6. Energy requirements of chemical processes should be recognized for their environmental and economic impacts and should be minimized. If possible, synthetic methods should be conducted at ambient temperature and pressure.
7. A raw material or feedstock should be renewable rather than depleting whenever technically and economically practicable.
8. Unnecessary derivatization (use of blocking groups, protection/deprotection, temporary modification of physical/chemical processes) should be minimized or avoided if possible, because such steps require additional reagents and can generate waste.
9. Catalytic reagents (as selective as possible) are superior to stoichiometric reagents.
10. Chemical products should be designed so that at the end of their function they break down into innocuous degradation products and do not persist in the environment.
11. Analytical methodologies need to be further developed to allow for real-time, in-process monitoring, and control prior to the formation of hazardous substances.
12. Substances and the form of a substance used in a chemical process should be chosen to minimize the potential for chemical accidents, including releases, explosions, and fires.

Unlike previous activities that promote waste recycling, green sustainable chemistry is a concept that promotes reducing resource and energy from the beginning, and to design products and manufacturing processes to minimize or eliminate derivatization of waste and toxic materials. Along with these concepts, there are many certifications that relate to botanical ingredients such as on the effective use of organisms, preservation and cultivation, and economic return to resource-possessing countries, which are also important to the cosmetic industry.

20.4.4 The International Trend of Organic Cosmetics and Natural Cosmetics

Consumers have become more conscious about the safety of cosmetics, leading to a stronger demand for cosmetics using botanical ingredients, and many organic cosmetics and natural cosmetics are now being manufactured and are spreading globally. However, even though there are many certification authorities around the world, a trusted unified standard to certify organic cosmetics and natural cosmetics is yet to be established and is still in a changing phase. A global standard has been established for organic foods, and the foundation of this standard was proposed by an international nongovernmental organization, the International Federation of Organic Agriculture Movements. However, these standards are not applied to cosmetics, and various certification authorities certify cosmetics based on their own standards. Historically speaking, the first natural cosmetics standard was set by Germany-based BDIH, founded by a task force of 19 German natural cosmetics manufacturers in 2001. Following the establishment of BDIH, many certification authorities started their own certification, and in 2010 a certification authority named COSMOS was founded in the European Union (EU) and the COSMOS-Standard was established.[16] This was the world's first international standard that defined organic cosmetics and natural cosmetics. The participating certification authorities are BDIH,[17] Cosmebio,[18] Ecocert Greenfile,[19] ICEA,[20] and Soil Association.[21] The arrangement between the participating authorities required some time, and the standard was aimed to be effective by 2015, but has been postponed. Additionally, use of some petroleum-based synthetic substances such as synthetic preservatives and synthetic surfactants is approved under the COSMOS-Standard, and has been under question from a consumer standpoint.

20.5 THE EFFECTIVENESS OF BOTANICAL SUBSTANCES

20.5.1 The Effectiveness of Botanical Materials

One of the main reasons to use botanical ingredients in cosmetics is for their physiological effects. Skin care cosmetics are used primarily for moisturization, but many dermatological effects are added to differentiate the products. Expectations are strong with botanical materials to meet these additional effects. Such effects may be their antiinflammatory action, tightening of the skin, ultraviolet (UV) absorption, antioxidative effects, cell growth stimulation, melanin inhibition to prevent senile pigment freckles, metabolism stimulation, circulation improvement and revitalization of the skin, and prevention of small wrinkles. However, only limited effects can be promoted unless the evaluation methods are unified. This is why botanical materials are being developed to add effective physiological effects as dermatological research evolves and new physiological systems of the skin are revealed. Extracting stable substances from botanical ingredients can be challenging. The components of botanical extracts are complex and can be largely affected by the season, agricultural environment, geographical condition, manufacturing method, and storing conditions. Thus, it is vital to determine the active components of botanical extract and control the component quality standards for a stable supply with reliable functions and effects.

We will use the widely researched glycyrrhiza (licorice root) as an example of how botanical materials are researched and developed. Glycyrrhiza has a long history in both Eastern and Western cultures as an effective plant, and has also been used as a botanical ingredient in modern drugs ever since the structure of its active component, glycyrrhizic acid, was determined in 1937.

20.5.1.1 Glycyrrhiza

Licorice is the root or stolon of plants in the *Glycyrrhiza* genus in the perennial herbaceous plant legume family, distributed from northeast China to mid-Asia and southern Europe, with saponin glycoside as its main substance. It is believed that they were used for tonics and beauty agents since they were first found in the Mesopotamian valley 4000 years ago. The dried roots and stolon (Fig. 20.1) themselves are also referred to as licorice, and the rough extract or rough purified products using water or alcohol, or their main active substance, a triterpene glycoside called glycyrrhizic acid (Fig. 20.2), is used in a considerable 70% of Chinese herbal medicine prescriptions. They are widely used in medicine and cosmetics for their antiinflammatory and antihepatitis action. Glycyrrhizic acid is known to be 200 times sweeter than sucrose (sugar), and there is a strong demand in the food industry for glycyrrhiza as a natural sweetener.[22] In Japan, they are commonly added to soy sauce, miso, and pickles since the sweetener has an effect of reducing saltiness. They are also commonly used globally as a fragrant for cigarettes.[23]

Among the *Glycyrrhiza* genus, the most commonly used species are *Glycyrrhiza glabra*, *Glycyrrhiza inflata*, and *Glycyrrhiza uralensis*. The species of glycyrrhiza and the habitats are shown in Table 20.1. The effects of glycyrrhizic acid are not limited to antiinflammatory action, and show antiallergy, detoxification, and antiulcer action. They also

FIGURE 20.1 The root and stolon of the *Glycyrrhiza glabra*.

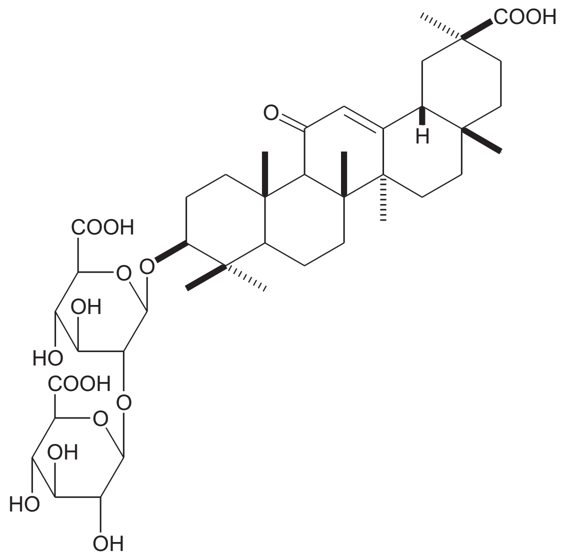


FIGURE 20.2 Structure of glycyrrhizin.

TABLE 20.1 Principal Component and the Licorice of Origin

Locality	Glycyrrhiza Species	Water-Soluble Component	Major Species-Specific Flavonoid Component
China Northeast ~Inner Mongolia ~Northwest	*G. uralensis*	Glycyrrhizic acid, liquiritigenin, isoliquiritigenin, and their glycosides	Licocoumarone, glycycoumarin, licoricidin
Shinkyo	*G. inflata*	Glycyrrhizic acid, liquiritigenin, isoliquiritigenin, and their glycosides	Licochalcone A, licochalcone B
Mongolia	*G. uralensis*	Glycyrrhizic acid, liquiritigenin, isoliquiritigenin, and their glycosides	Licocoumarone, glycycoumarin, licoricidin
Central Asia, such as Afghanistan and Uzbekistan	*G. glabra*	Glycyrrhizic acid, liquiritigenin, isoliquiritigenin, and their glycosides	Glabridin, glabrene
Europe, such as Italy	*G. glabra*	Glycyrrhizic acid, liquiritigenin, isoliquiritigenin, and their glycosides	Glabridin, glabrene

have been reported to remove reactive oxygen species and have oxidative stress defense,[24] and are studied for antiviral actions against HIV[25] and hepatitis C virus.[26] There also reports on the effectiveness against burn wound opportunistic infection.[27] Glycyrrhiza is widely used in cosmetics mainly for medicinal effects such as antiinflammatory action and antiallergy action.

Glycyrrhetic acid, an aglycone of glycyrrhizic acid, has been reported to show not only antiinflammatory and antiallergy actions, but also antibacterial action.[28] Along with glycyrrhizic acid analogues, flavonoid glycosides such as liquiritin or isoliquiritin, and specimen-specific flavonoids such as glabridin, glycycoumarin, and licochalcone A are found in licorice (Table 20.1). Approximately 150 substances of lipophilic flavonoid alone have been isolated and/or identified.[29]

The structures are diverse as well as their effects. As an example in cosmetics, glabridin (Fig. 20.3) is used for effects such as antioxidative effects,[30] estrogen-like activities,[31] and has been reported to have a, inhibition effect for melanogenesis.[32] Additionally, liquiritin (Fig. 20.4) shows whitening effects,[33] liquiritigenin shows antifungal action,[34] isoliquiritigenin shows effects such as antioxidative effects[35] and histamine H2 blocking,[36] and licochalcone (Fig. 20.5) A shows antiinflammatory action.[37] These flavonoid glycosides have been identified as flavone glycoside, chalcone glycoside, or isoflavone glycoside, and have been reported to show many pharmacologic actions such as antioxidative, antiinflammatory, antitumor, or anticancer actions.[38–42]

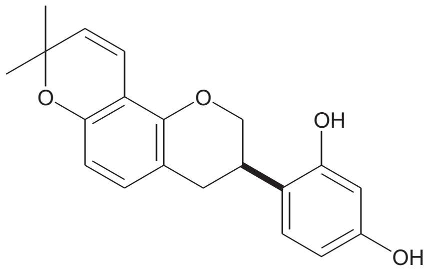


FIGURE 20.3 Structure of glabridin.

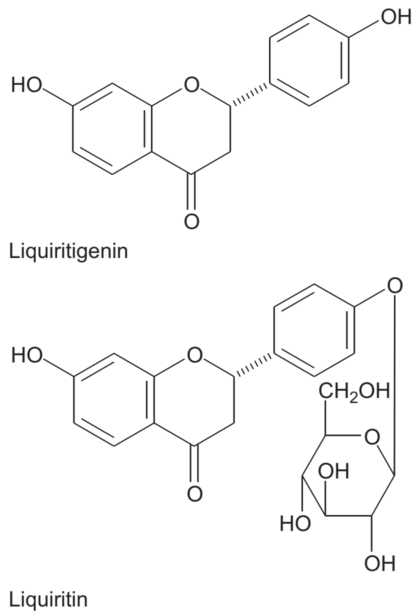


FIGURE 20.4 Structure of liquiritin and liquiritigenin.

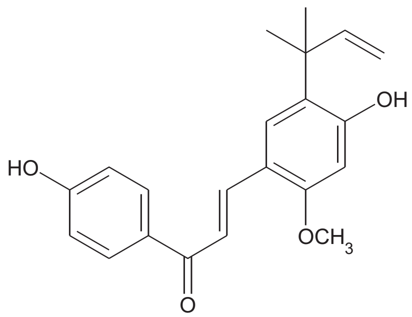


FIGURE 20.5 Structure of licochalcone A.

Overall, the flavonoid components of licorice show many effects and indicate that glycyrrhizic acid, the main component of licorice, is not the only effective component and suggests the wide applications of licorice (Table 20.2).

Many other botanical extracts are used or researched in cosmetics, and here we will see some examples of extracts where the active substance has been identified. Note that not all of these examples are applied in actual products and are examples of research in botanical extracts.

*20.5.1.2 Amla (*Emblica officinalis*)*

Amla is a woody plant in the euphorbiaceae family found in India. In Indian traditional medicine, known as Ayurveda, amla is listed as of the most important herbs and is known to have effects such as tonic and rejuvenation.

TABLE 20.2 Use of Licorice

Industry Type	Effect	Use
Medicine	Anti-inflammatory effect, anti-allergic effect, fibroblast proliferation effect, granuloma formation action, antibody forming inhibitory action, stress reaction inhibitory action, anti-ulcer effect, detoxification	Gastritis, gastric ulcer, duodenal ulcer, stomachodynia, gastric hyperacidity, hangover, autointoxication, dermatitis, urticaria, eczema, drug intoxication, hepatitis, jaundice, various pain, moist cough, conjunctivitis
Cosmetics		Cream, lotion, hair tonic, soap, bath agent, toothpaste
Food	Salt familiar, umami, flavoring, sweetness, flavor enhancement	Soy sauce, miso, pickled vegetables, food boiled down in soy sauce, soup, sauce, dainty, fish paste products, drink, frozen dessert, confectionery, candy, cocoa, dairy products

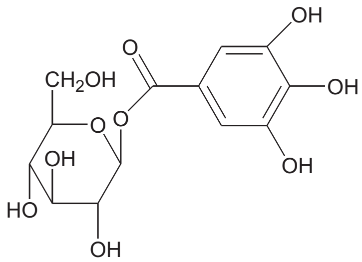


FIGURE 20.6 Structure of β-glucogallin.

The oil of amla is one of the world's oldest hair conditioners, but in recent research it has been reported to have fibroblast proliferation stimulation and procollagen biosynthesis stimulation action.[43] Furthermore, recent studies revealed that it also has whitening action, strong antioxidative effects, and MMP-1 blocking action, and its strong antioxidative effect was found to be from β-glucogallin (Fig. 20.6).[44]

20.5.1.3 Ashwagandha (Withania somnifera)

Ashwagandha is one of the most commonly used herbal medicines in the Ayurveda, and has been used for purposes such as antiinflammatory action, antistress, revitalization, and memory enhancement.[45] It is also said to moisturize the skin, and is used as an external agent for preventing freckles and wrinkles. Recent research reports that the concentration of intracellular reduced glutathione is maintained with induced expression of glucose-6-phosphate dehydrogenase, showing reactive oxygen species removal activity, thus ashwagandha is a botanical material that is expected to prevent cell death and aging caused by UV.[46] The active components of ashwagandha have been identified as steroid lactones, namely withaferin-A (Fig. 20.7), withanolide-A, withanoside, and withanone.[47–50]

20.5.1.4 Duabanga grandiflora

Duabanga grandiflora is a tree in the lythraceae family, found in the Malay Peninsula, the Yunnan Prefecture in China, and other subtropical rain forests and highlands, and its seed decoct has been used in folk medicine for food poisoning, stomachache, peptic ulcer, and other purposes. Recent studies show possibilities with cosmetic application with its effect of promoting production of type-III collagen that decreases with age,[51] and the effective component has been identified as a hydrolytic tannin, eugeniin (Fig. 20.8).[52]

20.5.1.5 Acerola (Malpighia emarginata)

Acerola is an evergreen shrub with edible bright red fruit found in the West Indies and from Northern South America to Central America. The extract of acerola contains large amounts of polyphenols, and among these polyphenols, quercitrin (Fig. 20.9) is a characteristic flavonol glycoside that is reported to have advanced glycation endproduct inhibiting action[53] and antiinflammatory action.[54]

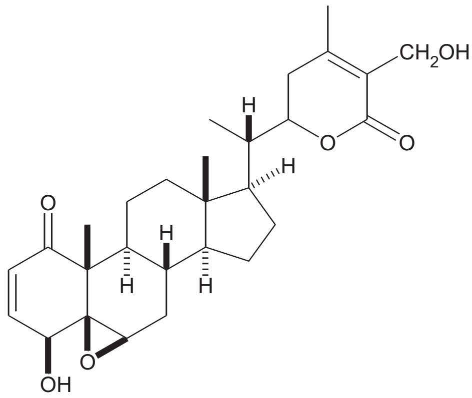


FIGURE 20.7 Structure of withaferin A.

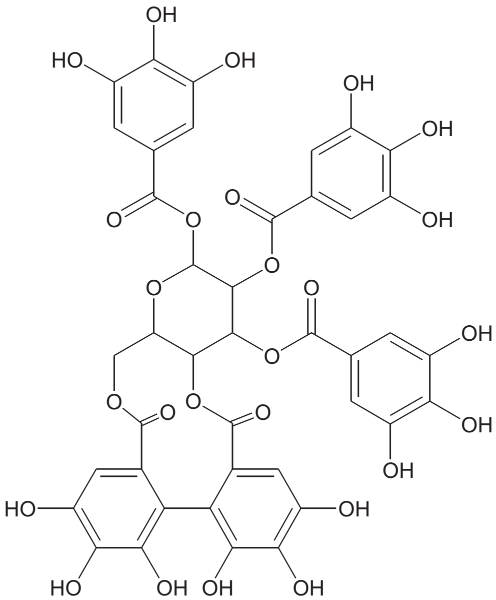


FIGURE 20.8 Structure of eugeniin.

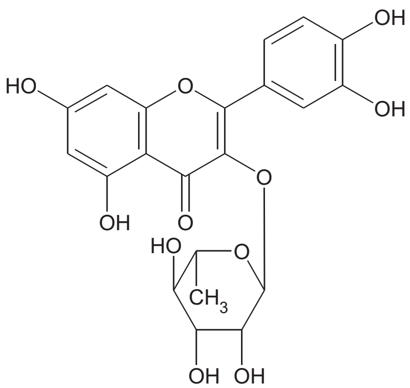


FIGURE 20.9 Structure of quercitrin.

20.5.1.6 Anastatica hierochuntica

Anastatica hierochuntica is an annual plant in the Brassicaceae family, found in the Sahara Desert and that grows by spreading on the ground. It has been an amulet for women in Ancient Egypt, and is believed to be effective against female disorder, and many active substances have been identified.[55,56] In cosmetics, many active substances have been identified from this plant that inhibit melanogenesis, and the active systems has been reported as tyrosinase inhibiting activity, or TRP-2 mRNA expression inhibition.[57]

20.5.1.7 Ginkgo (Ginkgo biloba*)*

Ginkgo leaf extract is known to accelerate thioredoxin reductas-1-gene expression, and the active substance kaempferol has been found to inhibit epidermal intracellular oxidation and it has been reported that it has potential for antiaging action of the skin.[58]

20.5.1.8 Green Perilla (Perilla frutescens *Var.* crispa *f.* viridis*)*

Chalcone is an aromatic enone in the flavonoid family and is a precursor of flavone. Many chalcones have been found from plants,[59] and have been reported to show antiinflammatory action and/or antioxidative effects.[60] DDC (Fig. 20.10) is a chemical compound that has chalcone as its main structure and is found in Green perilla. The DDC activates in the organism's antioxidative system through Nrf2-ARE pathway and induces the expression of antioxidative enzymes, and is reported to show cellular defense action against in vivo oxidative stress.[61]

20.5.1.9 Veratrum album *Subsp.* oxypetalum

In 1939, a polyphenol *trans*-resveratrol (Fig. 20.11) was found from *Veratrum album*, a plant in the liliaceae family. In 1988, this substance was reported to be in grape skin, and in 1992 the French Paradox was reported, where the rate of ischemic heart disease patients was lower with subjects who reported higher wine consumption.[62] In cosmetics, they are reported to have antiinflammatory action by inhibiting NF-κB functions that cause inflammation.[63] Furthermore, they are reported to inhibit the expression of MSH receptors, melanocortin 1 receptors, and block the downstream signals that synthesize them.[64]

20.5.1.10 Citrus unshiu

β-cryptoxanthin (β-CPX) was identified as the active component of *Citrus unshiu*. β-CPX is a carotenoid that has been reported to improve osteoporosis and obesity. It is reported to inhibit the expression of ET-1 receptors endothelin A and endothelin B receptors (EDNRA, EDNRB) under UVB stimulation.[65]

20.5.1.11 Artemisia (Artemisia indica *Var.* maximowiczii*)*

Artemisia is a perennial herb in the Artemisia genus of the Asteraceae family, and is found in hills and fields around the world. The chemical substances of Artemisia consist of essential oil, flavonoids, and polyphenols (Fig. 20.12),[66–70] and are reported to show many antioxidative effects.[71–73]

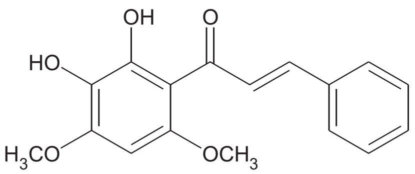


FIGURE 20.10 Structure of DDC (2′,3′-dihydroxy-4′,6′-dimethoxychalcone).

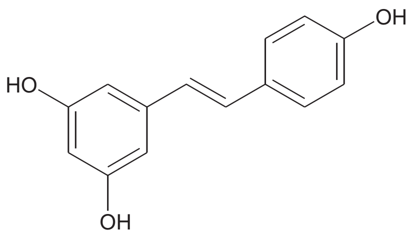
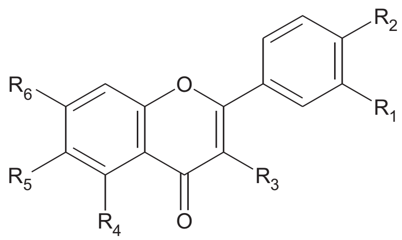


FIGURE 20.11 Structure of *trans*-resveratrol.



R_1	R_2	R_3	R_4	R_5	R_6	Name
-H	-OH	$-OCH_3$	-OH	-H	$-OCH_3$	kaempferol; 3,7-dimethyl ether
-OH	-OH	-OH	-OH	-H	-OH	quercetin
$-OCH_3$	-OH	$-OCH_3$	-OH	-H	-OH	quercetin 3,3'-dimethyl ether
-OH	-OH	$-OCH_3$	-OH	-H	$-OCH_3$	quercetin 3,7-dimethyl ether
$-OCH_3$	-OH	$-OCH_3$	-OH	-H	$-OCH_3$	quercetin 3,7,3'-dimethyl ether
$-OCH_3$	-OH	$-OCH_3$	-OH	$-OCH_3$	$-OCH_3$	quercetagenin 3,6,7,3'-tetramethyl ether
$-OCH_3$	$-OCH_3$	$-OCH_3$	-OH	$-OCH_3$	$-OCH_3$	quercetagenin 3,6,7,3',4'-pentamethyl ether

FIGURE 20.12 Structure of the flavonoid aglycones found in *Artemisia vulgaris*.

20.5.1.12 Xylocarpus granatum

Xylocarpus granatum is a plant in the mahogany family originating in Bangladesh, and its secondary metabolite has been reported to have a unique chemical structure and interesting biological effects.[74] For example, the active component of methanol extract of the leaf has been identified as a new chemical compound through isolation and analysis using Wnt signal blocking as an indicator.[75] On the other hand, the Wnt signal has been reported to control pigment stem cells' differentiation to skin melanocyte,[76] and has been a subject of interest in whitening.

20.5.2 Effectiveness of Botanical Aroma Compounds

Plants are a reservoir of aroma compounds. The biosynthesized aroma compounds often have structures similar to neurotransmitters or hormones, and show various medicinal effects. Here we will focus on the medicinal effects of the aroma compounds that work directly on cells or biomolecules instead of the effects caused by their smell or flavor. Tea tree oil (*Melaleuca alternifolia*) and terpinen-4-ol have been found effective to inhibit human M14 melanoma cell growth,[77] and peppermint oil has been reported to show antivirus action.[78] Furthermore, some mixed essential oils have shown synergistic effects,[79,80] and some have been reported to enhance skin permeability.[81,82] Other effects are also known, such as the free radical scavenging action of citrus essential oils,[83] or inhibition action of reactive oxygen species increase and cell death caused by UV and nitrogen dioxide with rosemary oil and its main component, 1,8-cineol.[84] Rosemary and other essential oils have been reported to have antibacterial action against bacteria such as *Propionibacterium acnes*,[85,86] and many essential oils have been reported to show antiinflammatory action with arachidonic acid cascade—related enzyme blocking.[87,88] It is also known that TRP receptors that react to temperature stimulation and pain are activated with essential oil components, and menthol has the effect of creating a cool feeling by raising the threshold temperature of cold receptor TRPM8.[89,90] Furthermore, camphor activates heat receptor TRPV3.[91] Grapefruit oil has been shown to inhibit neutral fat accumulation by inhibiting expression of genes that relate to neutral fat synthesis with glycerol-3-phosphate dehydrogenase.[92] Even when focusing on the medicinal effect of aroma compounds, you will understand how they have potential for many effects.

20.6 THE FUTURE AND CHALLENGES IN BOTANICAL SUBSTANCE DEVELOPMENT

20.6.1 Safety of Botanical Substances

Allergic contact dermatitis is caused when allergen substances penetrate into the skin and cause a delayed cellular immunity. Once a foreign substance that causes dermatitis is recognized as a skin sensitization allergen (sensitization), it causes a memory effect in the immune system and the allergen causes allergic response for the rest of one's life. As such, it is vital to confirm if the ingredients can cause skin sensitization. Safety should be the first and foremost priority in cosmetics, and cosmetics distributors owe the responsibility for their products and the responsibility to guarantee safety for their consumers.

However, banning animal testing in cosmetics started in the EU from 2004 and became effective in 2013,[93] and this trend is spreading globally. Although the trend against animal testing is spreading, in vitro testing methods for tests such as toxicokinetics, repeated dose toxicity, carcinogenicity, skin sensitization, or genotoxicity are yet to be established.[94] Furthermore, databases of in silico testing and structure-activity relationship are generally utilized as one of the few alternatives to animal testing, but the data is usually on high-purity single substances and is difficult to apply to multicomponent systems such as plant extracts. With food, experience from consumption is a strong basis to avoid allergy, but an incident occurred recently in Japan with hydrolyzed wheat protein (Glupal-19S) in soap, causing acute wheat allergy response, and this became a strong public incident. This is a notorious case that proved food allergy could be caused transdermally and not only by consumption.[95−98] Though this is a very rare case, it shows that such reactions can occur and they must be considered in development, and indicates that materials that have been proven safe from a long history as herbal medicines as well as for consumption still have advantage in development. Furthermore, there are expectations for plants that have been thoroughly studied in ethnobotanic research and the results have reported not only on the effects and activity but also on safety.[99] No matter how they are chosen, it is important to limit the target ingredients to those with abundant information on their safety, for example, plants with histories as herbal medicines. An alternative direction of botanical substance research that is actively being studied is using ingredients that have been proven safe for dermatological use, and studying their mechanisms to reveal new function systems.

20.6.2 Development by Fermentation of Botanical Substance

New methods are being studied to produce active components of plants without relying on extraction. Let's take glycyrrhiza as an example.

China is a main supplier of glycyrrhiza, but the habitat of glycyrrhiza is being destroyed and environmental damage is increasing due to the growing demand in China and overgathering of wild glycyrrhiza. With this change, gathering and export restriction of rare plants such as glycyrrhiza is being considered like that of rare earths, and there is a growing concern on price raise or stable supply. Decrease of the supply of high-grade glycyrrhiza with high glycyrrhizic acid content has also become a problem, and factory harvesting examinations and other methods of breeding high glycyrrhizic acid content glycyrrhiza breeds are being actively studied.[100] In the future, molecular breeding of high glycyrrhizic acid content will become more important, but there are also new endeavors using fermentation engineering with microbes to produce glycyrrhizic acids. To achieve this method, it is vital to identify the biosynthesis mechanism and synthesis enzymes of glycyrrhizic acids. We will take a look at the current progress of this fermentation method.

The carbon structure of glycyrrhizic acid is β-amyrin, a triterpene commonly found in many plants. β-amyrin synthase closes the rings of 2,3-oxidosqualene.[101] cDNA encoding β-amyrin synthase has been isolated from many species including *G. glabra*. Glycyrrhizic acid is biosynthesized with the oxidation of the C-11 and C-30 carbon and the glycosylation reaction of the third hydroxyl group of β-amyrin. These steps were predicted to be caused by two oxidation enzymes and two glycosylation enzymes. In 2008, Muranaka et al. identified the enzyme (CYP88D6) that changes 11-oxo-β-amyrin, a biosynthesized intermediate of glycyrrhizic acid by using a two-step hydroxylation reaction at the 11th position of β-amyrin as a catalytic reaction.[102] Furthermore, their approach successfully identified a second enzyme (CYP72A154), which is responsible for C-30 oxidation in the glycyrrhizin pathway. The CYP72A154 catalyzes three sequential oxidation steps at C-30 of 11-oxo-β-amyrin, and changes 11-oxo-β-amyrin to glycyrrhizic acid, which corresponds to the aglycon of glycyrrhetic acid[103] (Fig. 20.13). These biosynthesis genes expressed in an engineered yeast strain and glycyrrhetic acid was successfully synthesized. However, the yield of glycyrrhetic acid using transgenic yeast is still low and still needs further improvement to get enough yield for actual application. Methods for higher productivity must be studied, such as molecular breeding to optimize triterpenoid production by improving the mevalonate pathway or controlling the sterol pathway after 2,3-oxidosqualene. Some glycosylation enzymes of glycyrrhizic acid have also been identified, and industrial fermentation methods for glycyrrhizic acid production from glycyrrhetic acid is steadily advancing.[104]

20.7 CLOSING REMARKS

The history of the relationship between humans and plants is extremely long, and humans have used plants in every aspect of life, not only in cosmetics but for food and medicine as well. We have always relied on plants, when developing botanical materials for cosmetics it is needless to say that botanical substances are natural

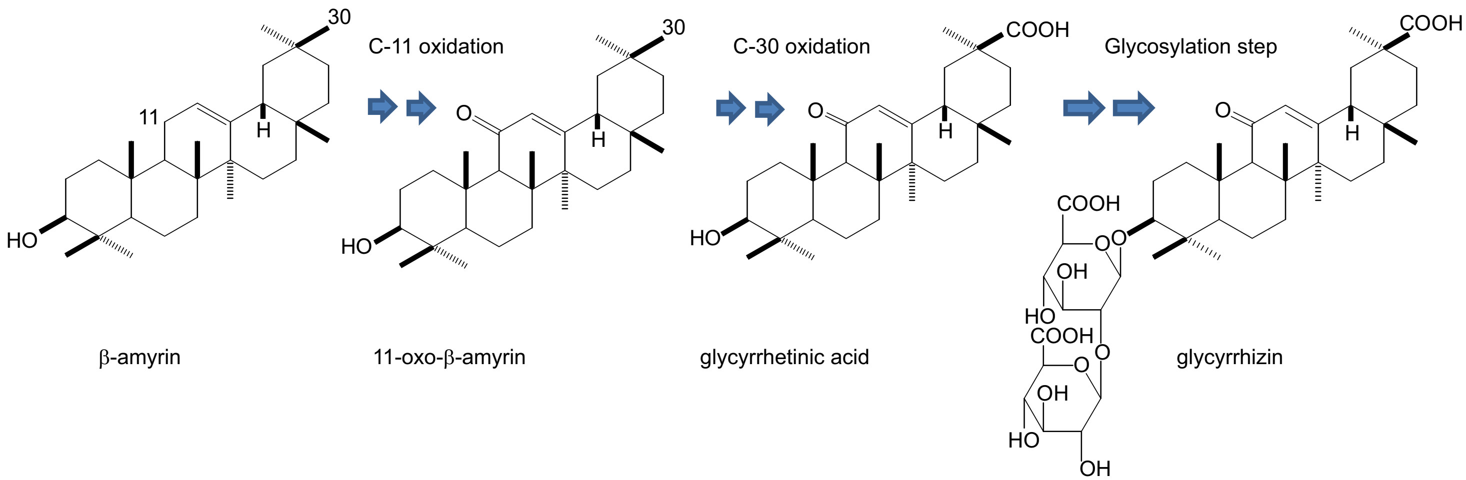


FIGURE 20.13 Proposed pathway for biosynthesis of Glycyrrhizin.

substances that must be treated differently from other chemicals. Safety assurance must be the foundation, but many specific areas must also be pursued, such as securing resources, being compliant with the Convention on Biological Diversity, standardization of materials, and unification of compositions. As fundamental research advances and the mechanisms of the skin and body are revealed, development of botanical substances will also advance to make more effective cosmetic ingredients and medicinal substances. Strong evidence will become required in cosmetics as well as a scientific basis of the effectiveness, and the demand for evidenced-based cosmetics is stronger. When looking at the future of cosmetics, the hopes for botanical materials with infinite potential seem even more promising.

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