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The importance and perspective of plant-based squalene in cosmetology

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Synopsis

Squalene is a highly unsaturated hydrocarbon from triterpenoid family, discovered as a major component of the liver oil of certain varieties of deep sea sharks. In the interest of protecting biodiversity, raw materials of animal origin must be replaced by alternative sources that respect our environment. Squalene is widely present as a component of the unsaponifiable fraction of vegetable oils (i.e., olive oil, amaranth oil). Amaranth oil seems to be the key source of squalene. Amaranth grains contains 7–7.7% lipids, and these lipids are extremely valuable because of the presence of ingredients like squalene, unsaturated fatty acids, vitamin E as tocopherols, tocotrienols, and phytosterols, which are not seen together in other common oils. In human skin physiology, squalene is not only used as an antioxidant, moisturizer, and material for topically applied vehicle, but is also used in treating skin disorders like seborrheic dermatitis, acne, psoriasis, or atopic dermatitis. Further studies on alternative sources are needed to explore the utility of squalene for treating skin.

INTRODUCTION

In Japan, a rich source of squalene is shark liver oil. Many Japanese people believe the shark liver oil contains powerful healing agents. Another rich source of herbal squalene is the amaranth plant, a type of grain plant that can survive in both scorching heat and extremely dry soil. It produces 6-foot stalks with feathery red or magenta plumes. The Greek word "amarantus" means "never withering." In India, the amaranth herb has been used for thousands of years. It is as rich in squalene and is as common in that region as the olive tree is in the Mediterranean basin. In the great epics of the ancient Indian cultures,

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this herb is believed to be empowered with immortal strength and fertility. The Sanskrit word for the plant "amaranth" means "King of Immortality." Aztec soldiers ate a very thick soup of this herb before going to war. The amaranth plant was outlawed by Spanish missionaries who were disturbed by its association with human sacrifice. In fact, they believed the key to the suppression of the Aztec culture was the annihilation of the plant. The Swedish Order of the Amaranth dates back to the 1653 reign of Queen Christina. The amaranth represented distinction and honor and was formed into the "Amaranthine Wreath," symbol of the Swedish order of the bond of fraternal friendship representing the strength and power of the plant (1).

Like the Aztecs who drank a soup of amaranth, ancient warriors of Japan and China—and even the Maoris of New Zealand—were known to drink shark liver oil before leaving for war. We can, therefore, trace cultural recognition of squalene-rich products with unique survival qualities to the Mediterranean region, Scandinavian, the Indian subcontinent, the Far East, and Central America (1).

Chinese healers were the first to conduct prescientific research into a rich natural source of squalene. In 1596, Lee Ji Chin, a Chinese healer of the Ming dynasty (1369–1644), composed a 52-volume compendium of some 2000 herbs, including the liver oil of the deep sea shark. Chinese traders brought the book to Japan, where it was known as Honzo Komoku. Samurai warriors used this oil to increase their strength. Villagers of Suruga Bay on the Izu Peninsula of Japan were accustomed to drinking the same oil. The local name of this special extract was "samedawa" or "cure all." In 1906, Dr. Mitsumaru Tsujimoto, a Japanese industrial engineer, discovered that samedawa contains extremely large quantities of an unsaturated hydrocarbon. He named the hydrocarbon squalene. Dr. Tsujimoto was presented the Imperial Award of the Japan Academy in honor of this achievement (1).

SQUALENE AND ITS SOURCES

Squalene is a highly unsaturated hydrocarbon from the triterpenoid family, consisting of six isolated double bonds (Figure 1). The molecule of squalene was discovered by a Japanese chemist Mitsumaru Tsujimoto as a major component of the liver oil of certain varieties of deep sea sharks. It is a low-viscous oil. When purified, it is very pale yellow to colorless with almost no odor. Shark liver oil is the general and most popular source of squalene (35–80%), which is also widely present as a component of vegetable oils (2–4). Oil of olive from *Olea europaea* contains 0.6–0.7% of squalene, whereas amaranth (*Amaranthus* sp.) oil contains appreciable quantities of squalene. Depending on the variety, the oil obtained from the amaranth seeds can contain up to 8% of squalene (2,3,5). The 13% of squalene has been found as a natural constituent of human skin lipids (6).

The best source of squalene is the liver of the deep sea shark (2,4,7). The last 40 years has brought a huge interest in squalene as a valuable compound in pharmacy or cosmetology; fishing on *Squalus* sp. went so far that some of them (i.e., *Squalus acanthias*, *Squalus albifrons*, *Squalus brevirostris*) are on the International Union for Conservation of Nature Red List of Threatened Species (8).

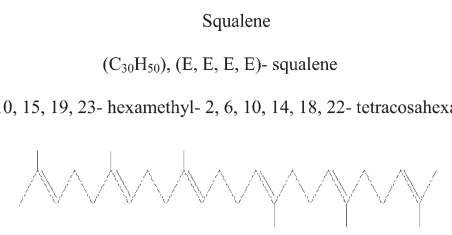

Figure 1. Squalene—structure and chemical nomenclature (2).

Oceana—the largest non-profit international ocean conservation and advocacy organization—has started the sharks' protection program in 2005. The campaign achievement was a declaration of some cosmetics companies to stop using shark liver oil in their products. L'Oreal (R)—as soon it was sure that there is applicable plant-based squalene supplies—had started switching to plant source as an ingredient of their creams, lotions, and glosses. Unilever® said it had stopped using shark oil in high-street brands such as Pond's® and Dove® some years ago and was now ensuring that beauty spas the firm owns in Spain did likewise. Boots®, Henkel®, Sisley®, Estée Lauder®, and Clarins® have either made similar decisions or never used shark sources in the first place (9).

In the interest of protecting biodiversity, raw materials of animal origin must be replaced by alternative sources that respect our environment. The main problem for the cosmetics industry is an authenticity of squalene source: is it shark oil or plant source? Isotope ratio mass spectrometry (IRMS) is a well-known and widely used technique to control authenticity (2). The method is based on the $^{13}C/^{12}C$ isotope ratio. The ratios of stable isotopes of a molecule are directly related to its origin (plant, animal, fossil, synthetic). Results indicated clear isotopic difference between the two principal sources of squalene (animal and vegetable) (2). The same authors' also proposed the gas chromatography combustion isotope ratio mass spectrometry (GC-C-IRMS), which allowed cosmetic manufacturers to be sure the squalene employed in their cosmetics is 100% plant origin. For consumers, this increases significantly the assurance that the products they use comply fully with the best industrial ethical and ecological standards (10).

Searching for an alternative source of squalene went to plant substitutes. Squalene is widely present as a component of the unsaponifiable fraction of vegetable oils. The very low level of squalene in vegetable oils does not represent a viable industrial source. Although olive oil (*O. europaea*) contains only 0.6–0.7% of squalene (2,5), large tonnages of oil are physically refined. During this process, the unsaponifiable fraction is concentrated in the refining condensate. This provides the bases of an industrial source of olive squalene. The industrial production of squalene from olive oil is considerably more complicated than the production from shark liver oil. This has a significant impact on the costs of olive squalene as compared to squalene obtained from shark liver oil (2).

The *Amaranth* sp. is a well-known plant because of its common use, followed by benefits from chemical structure of leafs and seeds. It was widely used by Mayas, Aztecs, and Incas (11,12). Amaranth contains appreciable quantities of squalene. Depending on the variety, the oil obtained from the seeds can contain up to 8% squalene. A lipid extract of the leaves has an average squalene concentration of 0.26%, which is clearly insufficient for industrial production (2). The studies on different transgenic crops

has shown that modifying the expression of certain genes improved crop plants and increased its resistance to low temperature and salinity of the soil (5,13). The amaranth genetic improvement should tend to increase the content of squalene and reduce raw fat (5).

The "Szarłat" (Łomża, Poland)—commercializing Amaranth-derived squalene company—has developed a technology called cold-pressing method. Cold-pressing method is a unique technology to produce oil from the seeds of amaranth without using organic solvents. By using this method, the contents and properties of squalene in amaranth oil remain unchanged. This method is extremely difficult in the extraction of small sized and low-fat content seeds. Developed methods allow the extraction of the native oil called virgin oil (virgine) (13–15).

PERSPECTIVE OF ADVANTAGES

Plant-based squalene may be used in cosmetics and personal care products according to the general provisions of the Cosmetics Directive of the European Union. The Cosmetic Ingredient Review Expert Panel noted that studies indicated that squalene was slowly absorbed through the skin. The toxicity of squalene by all routes was low. At 100% concentrations, the compound is a nonirritant to the skin and eyes. Products containing squalene were not dermal irritants or sensitizers (16).

Olive oil is graded as a healthy agent, not on the quality of the oil, but on the accompanying substances dissolved in it. Amaranth grains contain only 7–7.7% lipids, and these lipids are extremely valuable because of the presence of ingredients like squalene, unsaturated fatty acids, vitamin E as tocopherols, tocotrienols, and phytosterols, which are not seen together in other common oils (e.g., olive oil) (5,14,15). Amaranth squalene with accompanying compounds shows a high biological activity as is proven from the results of numerous researches (6). The advantages of amaranth oil in the near future can become one of the basic squalene sources (17) despite a bit higher price than other common oils.

Human skin, covering the entire outer surface of the body, is the largest organ and is constantly exposed to sunlight stress, including ultraviolet (UV) light irradiation. The skin tissue is rich in lipids, which are thought to be vulnerable to oxidative stress from sunlight. Squalene is a structurally unique triterpene compound, which is one of the main components (about 13%) of skin surface lipids (6,16). High content of squalene in our skin is good enough proof of its importance in this organ. Squalene helps the skin retain moisture, making it smooth and elastic. But its major role is to protect skin from UV radiation and thereby taking into account its anticancer and immuostimulating properties to protect from skin cancer. Undoubtedly, more extensive use of squalene in medicine and cosmetics is still ahead (17,18).

ROLES OF SQUALENE

Antioxidant. Squalene has been reported to possess antioxidant properties. *In vitro* experimental evidence indicates that squalene is a highly effective oxygen-scavenging agent. Subsequent to oxidative stress such as sunlight exposure, squalene functions as an efficient

quencher of singlet oxygen and prevents the corresponding lipid peroxidation at the human skin surface (19,20). Other studies have shown the effect of squalene on reducing superoxide anion. These results suggest the possible role in alleviating skin irritation (6,21). Topical application of antioxidants has been recently suggested as preventive therapy for skin photoaging and UV-induced cancer (20). It has been known that treating the skin with oils offers considerable protection from sunburn because of a strong absorption band in the erythemogenic region (21). Topical application of the cream containing antioxidants (vitamin E, CoQ_{10}, squalene) led to a significant increase in the sebum level (22). This could be a significant proof of the unique composition of amaranth oil.

Emollient. Amaranth oil has been reported to contain relatively large amount of squalene, which is used as an important ingredient in skin cosmetics and penetrants (13). Squalene as a natural emollient is quickly and efficiently absorbed deep into the skin, restoring healthy suppleness and flexibility without leaving an oily residue (6,23). When applied to washed or sun exposed skin and hair, squalene helps to restore the lost oils. It readily forms emulsions with fixed oils and lipophilic substances and does not oxidize nor turn rancid. Squalene has also been found to accelerate dye-dispersion in lipsticks, producing a high gloss and acts as a long-lasting fixative for perfumes. (21).

Skin hydration. Since squalene is a part of our skin's natural lubrication, it has a moisturizing effect on the skin. The ingredient not only provides hydration but also helps reduce the appearance of wrinkles and fine lines by filling the skin with water. On the basis of amaranth oil, characterized by a unique content of squalene, forms such as emulsion of "oleogel" can be used (17).

The vernix caseosa (VC) substitute based on squalene, which is composed of different lipid fractions mixed with squalene, triglycerides, cholesterol, ceramides, and fatty acids, mimic the lipid composition of VC. This substitute can be used as an innovative barrier cream for barrier-deficient skin (i.e., psoriasis) (6,24). There are no confirmed clinical studies for atopic dermatitis by using VC substitute.

Treating skin diseases. Seborrheic dermatitis and acne (following seborrheic dermatitis) are the most common skin disorders. It is important to keep the same level ingredients of sebum. The correct amount of free, unsaturated fatty acids (linoleic acid) ensures the proper density, fluidity, and viscosity of sebum. If the sebum is sufficiently liquid and not very viscous, it freely flows on the skin surface causing hyperkeratosis. Decisive is the percentage of triglycerides and squalene: the more the squalene, the less the acne changes. Squalene is often used in acne-against cosmetics that changes the composition of the lipid layer of the skin—when the content of squalene increases, fats are reduced appropriately (25).

The protective action of squalene and alkylglycerols from bacterial and fungal infection indicates that they could be recommended for patients suffering from atopic dermatitis. Patients with xerosis and disturbances of skin barrier are easily susceptible to atopic dermatitis (25). These studies were based on shark liver oil.

Analysis on the effects of cosmetics containing squalene have made several proposals. First, by improving the composition of sebum (i.e., by the enrichment of essential fatty acids and squalene), keratosis disorders are eliminated. Second, these cosmetics could prevent the skin irritations caused by product oxidation. Third, these act as bacteriostatic and antifungal agents to limit the number of microorganisms settled in the follicles (26).

Topically applied vehicles. Squalene is also used as a material or additive in topically applied vehicles such as lipid emulsions and nanostructured lipid carriers (NLCs). NLC is a novel type of lipid nanoparticle with a solid particle matrix possessing structural specialties and improvements, such as increased loading capacity, long-term physical and chemical stability, triggered release, and potentially supersaturated topical formulations (6). In many instances, it would be advantageous if the rate of penetration of medicaments through the epidermis could be increased. This would help in bringing about a more rapid and profound action of the locally applied compounds. If penetration (transfollicular, transepidermal, and into the horny layer) could be increased, many new compounds could be introduced into different therapies (27).

CONCLUSIONS

Because of the biological activity of squalene, which is widely used in cosmetics and pharmaceuticals, further tests are necessary for the verification of its use in preparations for skin. Since squalene is a part of our skin's natural ingredient, it has a moisturizing effect on the skin. The ingredient not only provides hydration but also helps reduce the appearance of wrinkles and fine lines by filling the skin with water. The VC substitute based on squalene brings the novel role in psoriasis and atopic dermatitis. The results of the studies on psoriasis are promising and possibly further clinical studies enable a wide application of squalene. There are no confirmed clinical studies for atopic dermatitis by using VC substitute, which seems to be taken. Bacterial and fungal protection was improved by squalene in atopic dermatitis, seborrhea, and acne. Also a combined role of squalene as an antioxidant, natural emollient, and skin hydration agent can be used to improve skin physiology problems. Squalene is also used as a material or additive in topically applied vehicles that may improve rapid and profound action of the locally applied compounds. Further studies on alternative sources are needed to explore the utility of squalene for treating skin, the high-quality amaranth oil seems to be the key of it. Several implications can be drawn from this review. Squalene shows several advantages for skin tissues. The present success of squalene shows the promise of further clinical trials for skin uses.

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