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Research Paper

Scientific substantiation and the release of saponins from plant raw material for food and cosmetic cream technology

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Abstract

The article presents the results of author scientific research dealing with the use of saponins, biologically active substances in food and cosmetics technology. The problems of their formation in plants, the chemical nature and the features are considered, biological and physico-chemical properties of saponins are studied. By their nature, saponins are divided into steroid and triterpene, differing by glycoside part of a molecule, thus, with different biological and chemical properties, but they are all capable to develop foam in aqueous solutions, and this feature is the origin of their name. The name originated from the word «Sapo», which means soap. It should be noted that at present saponins are not studied fully as other biologically active substances (BAS), although they are of interest and relevance. In the course of scientific work, about 20 species of saponin-containing plant material, both steroid and triterpene ones, were selected and analyzed for the determination of saponins. The selection of raw materials with the highest content was performed for their further use in cosmetics and as the surfactants in the production of emulsion food products. The following research methods were used in the work: qualitative reactions to the presence of saponins, foaming, saponin extraction, the release from dry aqueous extract of both steroid and triterpin ones. The method and the scheme of their release and precipitation are presented in the article.
The work was carried out within the framework of the state assignment of FSBEI HE «Moscow State University of Food Production» No. 14.7404.2017/бч «Scientific and applied bases of application of traditional and nonconventional vegetable raw materials and secondary products of its processing (fruit and berry, grain, bean, oil, essential oil, herbs) in technology of specialized products of the food industry, cosmetology and pharmacy»

Keywords: *Vegetable Raw Materials; Biologically Active Additive; Steroid and Triterpin Saponins; Food Products; Cosmetics; Physicochemical and Biological Properties; Release; Determination; Research Methods.*

1. Introduction

Saponins are the heterosides of plant origin, the derivatives of triterpenes and steroids, which have hemolytic, surface activity and toxicity to cold-blooded animals. For the first time, Schreider identified the plants of the Saponaria (soap) genus from flowers in 1811, and Malone first proposed the name in 1919 from the word «SAPO» which means soap, for their property to form foam in aqueous solutions.
Among plants they are contained in cell juice of all plant organs: in grass (horsetail), in leaves (kidney tea), in seeds (chestnut), in underground organs (blue cyanosis, high litter, licorice nude and Ural, ginseng, aralia Manchu). Most saponins are found in underground organs.
All saponins take part in the biochemical processes of plants: in small concentrations they accelerate the germination of seeds, the growth and the development of plants, in large concentrations they inhibit, they are plant growth hormones, affect the permeability of plant cells due to surface activity.
Figure 1 shows the steroid saponins of «normal» and «iso» series.

«Normal» series of
Spirochetal ring «iso» series

Fig.1: Steroid Saponins: «Normal» and «Iso» Series

The carbohydrate portion is attached at the position 3 of aglycon, it can have 1 - 9 monosaccharides in the form of glucose, galactose, xylose, arabinose, galacturonic and glucuronic acids. Monosaccharides can form linear and branched chains.
Figure 2 shows the saponin «dioscin», the aglycon of which is joined by a branched triosa.

Fig.2: Steroid Saponin «Dioscin» From Japanese Dioscorei.

Triterpene saponins. General formula (C5H8),[6].
Depending on the number of rings in aglycone structure, they are divided into tetracyclic and pentacyclic ones.
Tetracyclic are represented by dammaran, its derivatives and the products of their oxidation, widely found in ginseng, devil's-club and birch.

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Fig. 3: Shows Tetracyclic Triterpene Saponins.

Dammaran
Panaxatriol Panaxadiol
fig.3: tetracyclic saponin «dammarane» and the products of its oxidation.
Pentacyclic triterpene saponins contain five rings, in their turn; they are divided into 4 groups: ursan derivatives (α-amirin); oleanan (ß-amirin); lupan (lupeol) and gopan.
From a medical point of view, the derivatives of α- and ß-amyrins, which differ from one another by the arrangement of (-CH3) group substituents, are of great importance.
An important representative is ursolic acid, found in foxberries фтв cranberries, can be found in the form of glycosides and free aglycone.
ß-amirin is found in the form of oleanolic acid, which is the aglucone of saponins-aralosides and is found in Manchurian aralia, blue cyanosis, horse chestnut, spring primrose, medicinal calendula and patricia medium.
Glycyrrhetinic acid is an aglycone of glycyrrhizic acid, which is found in common licorice and Ural licorice.
The main compound in the biosynthesis of saponins is squalene.
Saponins are widely distributed in the world. Nearly 70 families are known among triterpene aglycons. These include aral, phlox, legume, Asteraceae, Lamiaceae, polygalaceae, horse chestnut families. Discorey, liliaceous, brown, Zygophylloideae, amaryllis can be found with steroid aglycons.
It should be noted that triterpene saponins are found in the plants without steroid saponins.

2. Results

Saponins were widely known in ancient times as detergents due to their surface-active properties, which also have different biologically active properties.
The scientific research in the field of triterpene glycoside use in cosmetic hair care products claims that the inclusion of 0.01% to 5% of saponin by weight, or the corresponding sapogenin, or plant extract in which it is present (in particular, the roots or the leaves of alfalfa Medicago sativa) or in shampoos, promotes the renewal of epidermal cells, stimulates hair growth and prevents their loss. The recent developments in the field of cosmetology suggest the use of saponin complex with various phospholipids; such complexes are more active than the corresponding free forms. Saponin phospholipid complexes can be included in cosmetic compositions as microdispersions in the form of gels or emulsions. The saponins in the composition of detergents for hair give a stable foam, which avoids the use of synthetic stabilizers. Besides, saponins and saponin-containing extracts (horse chestnut) are used as the part of anti-cellulite and anti-fat deposits.
Although saponins are toxic to fish and other cold-blooded animals, they are virtually non-toxic to humans and can be used in medicines as well as in functional foods.
In medicine, saponin-containing extracts are used as anti-inflammatory, hepatoprotective, expectorant and healing agents.
From the therapeutic point of view, plant saponins are important mainly due to their venotonic, hemolytic, antidiabetogenic, immunomodulating and adaptogenic properties.
The ginseng of the Far East is used to produce saponins for medical purposes. To do this, they apply the method of tissue culture on certain nutrient media of medicinal plants raw material biomass. The obtained biomass was used for preparation of drugs.
As for food, saponins are traditionally included in the diet of people with such foods as beans, lentils or peas. For example, the daily diet of the UK residents contains about 15 mg/person, and the average daily dose of the vegetarian population is 110 mg/person.
In recent years, the introduction of new methods to determine the structure, the isolation and the separation of complex mixtures and the use of biological test systems can detect new biological activities of saponins.
One of the most studied properties of saponins is their hemolytic activity. They are able to dissolve the lipid part of the erythrocyte membrane. Thus, a semipermeable shell becomes a permeable one. Hemoglobin enters the blood plasma freely and dissolves in it. A red transparent solution – «lacquer blood» is developed. Only glycosides have hemolytic activity. In this regard, saponins are not used for intravenous administration as they cause anemia. The saponins lose their hemolytic activity when administered orally, after the hydrolysis in the gastrointestinal tract to aglycons. The hemolysis of erythrocytes is not caused by all saponins. Licorice saponins do not have such a property.
The hemolysis reaction with 2% suspension of erythrocytes in isotonic solution is based on the biological properties of saponins. An infusion of isotonic solution is prepared from the vegetable raw material to carry out this reaction.
As for the biological role of saponins in plants, they take part in all biochemical processes:
In small concentrations, they accelerate the germination of seeds, the growth and the development of plants, and in large concentrations, they inhibit these processes. Thus, saponins play the role of plant growth hormones
Saponins affect the permeability of plant cells, which is conditioned by their surface activity.
Saponins do not possess a hemolytic and surface activity in the composition of the same complexes.
The influence of environmental factors on the accumulation of saponins is strictly specific. It is difficult to reveal general patterns for all plants among them.
Six-flowered astragalus grows in the south of the European part of Russia in the steppe zone on dry meadows, along the slopes of the beams, and common licorice grows in the floodplains and the valleys of rivers, in steppe and semidesert regions of the European part of Russia and in Siberia.
Greek valerian polemonium grows along fringes and along forest roads in forest and forest-steppe zones of the European part of Russia and in Siberia.
The plants of the Araliai family grow in the Far East, where its own climatic and soil regime developed.
The accumulation of glycyrrhizic acid for licorice depends on soil salinity. The more salinity level, the less glycyrrhizic acid in licorice roots. The accumulation of glycyrrhizic acid is greater with soil moisture increase.
Ginseng, devil's-club, aralia and Dioscoréa nippónica occur in the forests of the Far East (Primorsky, Khabarovsk Territory).
Ginseng is cultivated in the Far East on certain nutrient media of medicinal plant raw material biomass. Later, this biomass is used to produce medicinal products.
Orthosiphon stamen (kidney tea) is imported from the countries of tropical Asia.
According to their physico-chemical properties, saponins are colorless or yellowish amorphous substances. Glycosides are isolated in the crystalline state with up to 4 monosaccharides in the carbohydrate chain.
Most saponins have optical activity, they are capable of lowering the surface tension, they are hydrolyzed by enzymes and acids easily. This surface activity is associated with the presence of saponins in the molecules of hydrophilic and lipophilic residues. In aqueous solutions, triterpene saponins give an acid reaction, and steroid saponins give a neutral reaction.
Glycosides are soluble in water and dilute (60-70%) alcohols, they dissolve in cold, in stronger (80-90%) alcohols - only with heating, and they precipitate during cooling. They are insoluble in organic solvents (acetone, chloroform, benzene, diethyl ether). The solubility increases with carbohydrate chain increase.
Free sapogenins do not dissolve in water, but are well soluble in organic solvents.
A specific property of saponins is their ability to reduce the surface tension of liquids (water) and give a strong, thick foam during shaking.
Depending on aglycone structure, the presence of individual functional groups, as well as the presence of a glycosidic bond, their

chemical properties change, they hydrolyze under the influence of enzymes and acids. The derivatives of mevalonic and glycyrrhitic acids are hydrolyzed by the action of alkalis.
Acid saponins form insoluble complexes with heavy metal (Ba, Pb) salts and form the complexes with proteins, sterols, lipids and phenolic compounds. They do not have hemolytic and surface activity in the composition of saponin complexes.
Saponins, which have a steroid nucleus in their basis, enter into a specific Lieberman-Burkhard reaction.
Thus, saponins are a complex group of compounds, steroid and triterpenoid derivatives, which have biological activity and toxicity to cold-blooded animals. Triterpene saponins are more widely distributed in the plant world, they are found in the plants of almost 70 families.
Steroid saponins often accompany cardiac glycosides in plants. The plants producing triterpene saponins do not contain steroid saponins and vice versa.
Currently, saponins are not studied as deeply as other biologically active substances (e.g., flavonoids, essential oils, vitamins, tannins, etc.). In this regard, the study of saponins is of undoubted interest and very relevant.
Saponins are widely used in the food industry - for the production of beer and effervescent beverages, for the production of halva, at home - to wash thin colored fabrics, to fix paints. Saponins make the part of fire-fighting mixtures and are the source of corticosteroid synthesis. In the cosmetic industry, the use of saponins is limited and not common, but they can be used in the production of shampoos and hair balms, creams and body and face scrubs and shaving gels.
In the course of scientific work, these types of raw materials were selected and analyzed for the determination of saponins: Caucasian yam (roots), lily of the valley (grass), Tribulus terrestris (grass), fenugreek (seeds), onion, green onions, garlic, white potatoes (Vivalldi species), yucca (grass), agave (stems), beetroot, common licorice (roots), ginseng (roots), kidney tea, horse chestnut (fruits), Greek valerian polemonium (roots), medicinal herb (roots), cabbage, primrose and patrinia (roots).
The purpose of this study is to isolate the biologically active substances of saponins that have foaming, emulsifying, stabilizing, antioxidant properties and have an antiseptic effect for the use in cosmetic and functional food products.

3. Discussion

The following research methods were used in the work: qualitative reactions to foaming; the extraction of saponins; the isolation of saponins from a dry aqueous extract
Qualitative reactions to saponins
A qualitative reaction to foaming provides an opportunity not only to verify the presence of saponins in the samples under study, but also to determine steroidal raw materials and triterpene saponins.
The reaction is based on the interaction of hydrochloric acid or sodium hydroxide with saponins. If there are triterpene saponins in the raw material, the foam forms in alkaline and acidic media. If the raw material contains saponins of the steroid group, a foam is formed in the alkali environment, several times larger in volume and stability.
In the course of qualitative precipitation of saponins, magnesium hydroxide and lead acetate were used from aqueous solutions. In this case, triterpene saponins are precipitated by medium lead acetate, and steroid ones - by magnesium hydroxide.
The results of the raw material research are presented in Table 1.

Table 1: Qualitative Foaming Reaction Indicators

Item №	Studied raw material	Foaming reaction		Precipitation reaction	
		Hydrochloric acid	Sodium hydroxide	Lead acetate	Magnesium hydroxide
Fruits and vegetables					
1	Bulb onions	-	+	-	+
2	Green onion	-	+	-	+
3	Garlic	-	+	-	+
4	White potatoes	-	+	-	+
5	Beetroot	+	-	+	-
6	Horse chestnut	+	-	+	-
7	Garden cabbage	-	-	-	-
Dry roots of herbs					
8	Caucasian yam	-	+	-	+
9	Common licorice	+	-	+	-
10	Ginseng	+	+	+	-
11	Greek valerian polemonium	+	+	+	-
12	London pride	+	+	+	-
13	Primrose	-	-	-	-
14	Patrinia	-	-	-	-
Dry leaves and stems					
15	Lily of the valley	-	+	-	+
16	Tribulus terrestris	-	+	-	+
17	Kidney tea	+	+	+	-
18	Yucca	-	-	-	-
19	Agave	-	-	-	-
Seeds					
20	Fenugreek	-	+	-	+

Foam and a small precipitate were observed in the studied samples No. [1-6], [8-12], [15-17].
No qualitative responses to saponins were obtained in the samples No. [7], [13], [14], [18], [19].
Steroid saponins were found in the samples [1-4], [7], [12-15], and triterpene saponins were found in the samples 5, 6, 8, 9, 10, 11, 14.
2. Saponin isolation method from a dry aqueous extract

Fig. 4: Saponine Isolation Scheme.

The scheme of saponin isolation is shown i\on Figure 4. The proposed scheme is based on the physicochemical properties of the substances contained in the extract.
The results of saponin release and their percentage in plant raw materials are presented on Fig. 5

Fig.5: Diagram - Saponin Content in Test Samples.

Having analyzed the vegetable raw materials, it was found that the highest content of steroid saponins is in beetroot, horse chestnut, common licorice, ginseng, carrion flower and partrinia. Saponins were also found in yam, burnut and fenugreek of triterpene nature.

4. Conclusion

Thus, there are the following results of the work:

1) It was shown that saponins are found almost in all the plants under study.
2) The scheme of saponin release from plant material was studied and carried out.
3) It was revealed that the greatest content of steroid saponins is found in yam, burnut and fenugreek, and triterpene saponins are found in beetroot and horse chestnut.

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