National University of Pharmacy Department of Chemistry of Natural Compounds and nutriciology

# The general characteristic of SAPONINS

# SAPONINS-containing Medicinal plants and herbal drugs

## Plan

- **1.** Give the definition of the terms "Triterpenoids", "Saponins", "Sapogenins", "Steroids", "Foaming index".
- 2. Classification of saponins with examples of structures.
- **3. Physical and chemical properties of saponins.**
- 4. Identification of saponins.
- 5. Quantitative analysis of saponins.
- **6. Biological properties of saponins.**
- 7. Medicinal plants, plant material containing saponins.

## Introduction

Saponins are naturally occurring plant glycosides; which is to say they are phytochemicals — chemicals found in plants. They possess soap-like qualities and produce a lather when mixed with water.

Saponins have a unique chemical structure that produces **foam** when mixed with water, just like a detergent.

And, also like detergent, saponins can bind with water as well as fats and oils. This means that, in the digestive tract, saponins produce an **emulsification** of fat-soluble molecules.

Specifically, saponins **bind to bile acids and help eliminate them from the body, preventing cholesterol from being reabsorbed**. You might even say saponins "wash away" various toxins.

The unique chemical structure of saponins allows them to offer a number of prospective health benefits.

It's believed saponins have a favorable effect on *cholesterol*, can help **boost the immune system, have an antioxidant effect, and may even support bone strength.** 

## Questions

 Give the definition of the term "Triterpenoids", "Saponins", "Sapogenins", "Steroids", "Foaming index".

2. Classification of saponins with examples of structures.

3. Physical and chemical properties of saponins.

4. Identification of saponins based on their physical, biological and chemical properties (reagent used – result obtained).

5. Quantitative analysis of saponins – methods and their descriptions.

6. Biological properties of saponins.

7. Structures of saponins:  $\alpha$ -amyrin,  $\beta$ -amyrin, lupeol, friedelin, dammarane, cycloartane, panaxadiol, panaxatriol, calendulosides, glycyrrhizinic acid, glycyrrhetic acid, aescin, diosgenin, cholesterol, cyclopentane perhydro phenanthrene.

8. Medicinal plants, plant material containing saponins.

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Saponins – a group of natural organic compounds, which due to their chemical structure appear to be glycosides, possessing a high surface and haemolytic activity, and are toxic to cold-blood animals (frogs, fish).

The aglycones of saponins are collectively referred to as *Sapogenins*. The more poisonous saponins are often called *Sapotoxins*. Glycosides = natural compounds which consists of sugar units(glycon) + aglycon (genin or nonsugar untins) which are connected by a glycosidic bond

**Saponins** = natural compounds which consists of sugar units(glycon) + aglycon or **sapogenin** which are connected by a glycosidic bond Plant materials containing saponins have long been used in many parts of the world for their detergent properties for example, in Europe, the root of Saponaria officinalis (Fam. Caryophyllaceae) and in South America, the bark of Quillaia saponaria (Fam. Rosaceae). Such plants contain a high percentage of the glycosides known as saponins (Latin Sapo, means Soap) which are characterized by their property of producing a frothing aqueous solution.

## **Properties:**

Saponins form colloidal solution in water (hydrophilic colloids) which froths upon shaking. These substances modify and lower the surface tension and therefore foam when shaken. This has led to their use to increase the foaming of beer.

Practical industrial applications of saponins include their use in cleaning industrial equipment and fine fabrics and as powerful emulsifiers of certain resins, fats and fixed oils.  In general, they have a bitter, acrid taste and drugs containing them are usually sternutatory (*causing or producing sneezing*) and **irritating to the mucous membranes of eyes and nose.**

- Characteristic for all saponins is their ability to cause haemolysis of red blood corpuscles and to destroy them. When injected into the blood stream, they are highly toxic.
- When taken by mouth, Saponins are comparatively harmless, being not absorbed from the intestinal tract. Sarsaparilla, for example, is rich in saponins but is widely used in the preparation of non-alcoholic beverages.

Saponins are toxic especially to cold-blooded animals e.g. frogs. Many are used as fishpoisons.

The actual cause of the haemolysis:

The red blood cells carry sterols in their **membranes**, and when brought into contact with saponins, the sterols of the RBCs are precipitated and the colloidal chemical properties of the membrane are so altered as to give hemoglobin passage to the surrounding medium.

Saponins have a high molecular weight and their isolation in a state of purity presents some difficulties.

# **Structure of Saponins** $(C_5H_8)_6$ :

- According to the structure of the aglycone or sapogenin, two kinds of saponin are recognized:
- 1. The steroidal type.
- 2. The triterpenoid type.
- Both of these have a glycosidal linkage at C-3 and have a common biosynthetic origin via mevalonic acid and isoprene units.

The steroidal type (commonly tetracyclic triterpenoids, C-27).

1. Spyrostanol type,



To compare saponine and steroidal alkaloid (pseudoalkaloid)



2. Furostanol type.

HO

OH

CH<sub>2</sub>OGlu

# The **triterpenoid** type (tetracyclic and pentacyclic triterpenoids, **C-30**). **1. Tetracyclic triterpenoids**

Dammaran

Cycloortan



## 2. Pentacyclic triterpenoids





.30





Fridelan



β-Amyrin (Oleanan)

# A. Steroidal saponins

The steroidal saponins are less widely distributed in nature than the pentacyclic triterpenoid type. Steroidal saponins are of great pharmaceutical importance because of their relationship to compounds such as the sex hormones, cortisone, diuretic steroids, vitamin D and the cardiac glycosides.

### **Quality determination of saponins**

Tests are based on physical properties:

a) They foam in aqueous solution.

b) Determination of chemical nature of saponines.

#### ✓ Tests are based on chemical properties :

a) Precipitation: barium water; lead acetate; 1% alcoholic solution of cholesterol.
b) Color reaction:

Reagent	Color			
$H_2SO_4$ , conc.	yellow → red-violet			
<i>Liebermann-Burhard</i> (acetaldehyde, $H_2SO_4$ conc., chloroform	red ring $\rightarrow$ violet $\rightarrow$ green (steroidal)			
Formaldehyde, $H_2SO_4$ conc.	Yellow → red			
Laphon ( $H_2SO_4$ conc., $Cu^{2+}$ , >t <sup>0</sup> C)	Dark blue-green			
Salcovskij ( $H_2SO_4$ conc., chloroform)	Orange lower layer			
Sb (III), Sb(V) cloride in chloroform	red → violet			
<i>Sanije</i> (vanillin, $H_2SO_4$ conc., >t <sup>0</sup> C)	teriterpenoid- red; Steroid - yellow			

✓ Tests are based on biological properties (hemolysis).

## **Chromatographic determination of saponins**



 Sistem:
 Chloroform:acetic acid:methanol:water (60:32:12:8)
 reagents:
 A. anise aldehide solution with ac.acid and methanol
 B. Haemolysis reaction

- 1. Aescin,
- 2. Horse Chestnut seed,
- 3. Senega root,
- 4. Sarsaparilla root,
- 5. Ginseng root

# Quantitative determination of saponins

*Biological method:* determination of haemolytical index
 *Haemolytical index* – the lowest concentration of saponins, which can cause total haemolysis of RBCs during 24 hours.

Gravimetric method : based on precipitation of saponins by reagent

*Physicochemical method* : determination of foaming index; spectrophotomety, gas-liquid chromatography

*Foaming index* - the lowest concentration of saponins, which can cause a persistent foam when an aqueous decoction is shaken.

## All in all

## **SAPONINS:**

produce high and stable foam

Have haemolytic properties

Very toxic to cold-blooded animals

Irritate mucose membranes

They are glycosides (aglycon+ sugar units), glycosides are soluble in water and not soluble in organic solvents, aglycone arenot soluble in water and soluble in organic solvents

20

GINSENG ROOT - Radix Ginseng Ginseng - Panax Ginseng C.A. Mey Fam - Araliaceae

Ginseng occurs in Russia and Eastern Asia (Korea, Chir etc). Cultivated Ginseng is produced in Korea, Japan, th Russia and the United States, primarily in Wisconsin. The roots are gathered from 3 to 6 year-old plant and carefully cleaned and dried at a temperature about 50-60°C.

**Descriptoin.** Ginseng root occurs in rachis-like root, to 25 cm long with 2-5 large branchings, rare without them. The body of the root is thickened, nearly cylindrical with clearly expressed ringed thickenings in the upper part. The outer surface is longitudinally, rare spirally wrinkled. Their colour is yellowish-white. The upper part of the root narrows into transverse wrinkled rhizome-root-collar. The rhizome is short with several scars of fallen-off stems. The rhizome is represented by extended traces of the stem and topical bud. The odour is specific, the taste is sweet, burning, then bitterish.



#### Chemical constituents.

Ginseng contains a complex mixture of triterpenoidal saponins. These glysosides have been categorized into 3 series, the panaxosides, OH the ginsenosides and the chikusetsusaponins. Some 13 ginsenosides (e.g. ginsenosides  $R_0$ ,  $R_a$ ,  $R_b$ , etc.) have been described. The panaxosides are termed panaxoside A, B, C etc. Ginseng root contains a mixture of both steroidal and pentacyclic triterpenoidal saponins. Other compounds of the root having therapeutic activity are high-molecular-weight polysaccharides; these are known as panaxans and they have been shown to have hypoglycaemic activity. Other constituents isolated include sterols, vitamins of the B groups, volatile oil, fixed oil, ferments.





**Panaxadiol** 

OH



Ginsenoside,  $R_1$ =D-glucosa-glucosa,  $R_2$  = arabinosa - D glucosa

**Uses. It is classified as an ADAPTOGEN**. Ginseng exerts tonic and stimulant actions. The drug is used for treatment of anemia, diabetes, gastritis, sexual impotence, sluggishness, exhausting, nervous tension, hypotension. The drug is administered in such forms as powders, extracts and teas.

LOCOWEED HERB - Herba Astragali dasyanthi Locoweed - Astragalus dasyanthus Pall. Family - Fabaceae

**Distribution.** Southern Ukraine, Moldova, Southern Russia.

**Plant.** A perrenial herbs. Its aerial portion consists with a numerous slender hairy stem attaining a height of 30 to 40 cm bearing alternate imparipinnate leaves with long petioled, pale green, ovate, entire leaflets and axillaries spikes of yellow-colored papilionaceous flowers The fruit is a compressed legume containing kidney-shaped seeds.

**Description.** The dried herb, harvesting during flowering- Odor slight, peculiar, taste sweetish.



#### **Constituents.**

Herbs containing flavonoids (mainly quercetin); treterpenic glycosides ; tannins.



Dasyanthogenin

#### Uses.

Water infusion uses for treatment hypertensia I-II stage, cardiovascular insufficiency, chronical nephritis.



MARIGOLD FLOWER - Calendulae Flores Marigold-Calendula officinalis L.

Fam. Asteraceae

Saponins -2-10%Flavonoids – 0,8%

Flavonoids from flowers demonstrated positive antimicrobial activity against Staphylococcus aureus. Klebsiella pneumoniae, Candida monosa.

RO

faradiol-3-O-laurate

Calendulosides

faradiol-3-O-myristate

faradiol-3-O-palmitate

Saponins

laurate myristate

R

laurate myristate palmitate maniladiol-3-O-laurate maniladiol-3-O-myristate





Organic extracts of the dried flowers of topical application of Calendula has been shown to enhance the granulation and epithelialization of damaged skin. Liquid extract, Calendula Tincture, Ointment





HORSE CHESTNUT SEED- Semen Hippocastani Horse chestnut - Aesculus hippocastanum Fam.- Hippocastanaceae

**Triterpene saponins** (aescin) 3-5%, Hydroxycoumarins: chief component is aesculin, fraxin and scopolin; Flavonoids: including rutin, quercitrin, and isoquercitrin; Tannins : Condensed tannins (only in the seed-coat), oligomeric proanthocyanidins;

**Polysaccharides:** starch 50%;

fatty oil 2-3%.





## **Effects of horse chestnut**

As found in different animal tests and preclincal investigations, the principal ingredient of Horse Chestnut seed extract, triterpene glycoside mixture (aescin), has an anti-exudative, vascular tightening effect, and reduction of vascular permeability which result in an antiedemic effect.

Treatment of symptoms found in pathological conditions of the veins of the legs (chronic venous insufficiency), for example pain and a sensation of heaviness in the legs, cramps, pruritis and swelling of the legs. *Venostasin, Retardkapseln, Noricaven, Rexiluven, Aescusan, Hoevenol*, etc

### Liquorice root - *Radix Glycyrrhyzae* Liquorice - *Glycyrrhyza glabra L*. Fam. - *Fabaceae*

This plant is grown in Spain, Italy, England, France, Germany and the U.S.A., but the Russian and Persian drugs are obtained from wild plants.

**Description.** It consists generally of roots in nearly cylindrical pieces, up to 1 m long and 5-20 mm in diameter; externally, the bark is brownish grey to dark brown, longitudinally wrinkled, occasionally bearing small dark buds in rhizomes or small circular or transverse rootlet-scars in roots. The peeled root is yellow, smooth, fibrous, finely striated; fracture, fibrous, in the bark and splintery in the wood; internally, bright yellow. A distinct cambium ring separates the yellowish grey bark from the finely radiate yellow wood;

The fracture of roots is fibrous, yellow in colour. The odour is absent, the **taste is sickly-sweet**.



#### Chemical constituents.

The major constituents are triterpene saponins. Glycyrrhizin (glycyrrhizinic acid) is the major component (2-9%); Glycyrrhizin occurs as a mixture of potassium and calcium salts. It on hydrolysis releases two molecules of D-glucuronic acid and the

aglycone glycyrrhetic (glycyrrhetinic) acid.

Glycyrrhizin is generally regarded as the active principle of Radix of Glycyrrhizae and is responsible for its sweetness, which is 50 times that of sucrose.

Flavonoid constituents include liquiritigenin and isoliquiritigenin. Uses. As an expectorant in the treatment of coughs and bronchial catarrh. Also in the prophylaxis and treatment of gastric and duodenal ulcers, and dyspepsia. As an anti-inflammatory agent in the treatment of allergic reactions, rheumatism and arthritis, to prevent liver toxicity, and to treat tuberculosis and adrenocorticoid insufficiency

**Contraindications.** Radix Glycyrrhizae is contraindicated in patients with hypertension, cholestatic disorders or cirrhosis of the liver, hypokalaemia, or chronic renal insufficiency, and during pregnancy. *Syrupus "Liquriton", "Flacarbin"*.





Isoliquiritigenin

OH,

#### GREEK VALERIAN RHIZOME WITH ROOTS - Rhizomata cum radicibus Polemonii Greek valerian, Jacob's ledder - Polemonium caeruleum Fam - Polemoniaceae

#### **Chemical constituents.**

The drug contains triterpenic pentacyclic saponins- derivatives of  $\beta$ -amirin. Aglycones of these compounds are ethers of tritertenic alcohols –longispigenol, barrigenol, et so on.



	K	I.I.	112	13	14
Лонгиспиогенол	Н	Н	ОН	Н	Н
AR 1-баригенол	ОН	ОН	ОН	Н	Н
R 1-баригенол	ОН	ОН	ОН	ОН	Н
Камелиагенин Е	ОН	ОН	ОН	Н	СНО



#### Uses.

As a demulcent in the treatment of sore throats, and as an expectorant in the treatment of coughs and bronchial catarrh.

ARALIA ROOT - Radix Araliae elatae Aralia (spikenard) - Aralia mandshurica (A. elata), A.racemosa Fam. - Araliaceae

*Act. const*  **- Saponins : β-Amyrine:** Araloside A, B, C.

alkaloids: aralin,
Essential oil,

✓ resine,



Use Tincture, "Saparal" – tonic, adaptogen



Pine grass, Field horsetail EQUISETUM STEM - Herba Equiseti Equisetum arvense L. Fam. Equisetaceae

- **Flavonoids**: 0.6 to 0-9%: apigenins, luteolins,
- quercetins glucosides;
- Acids: caffeic acid ester up to 1%;
- Silicic acid 5 to 7.7%: to some extent water-solub
- Pyridine alkaloids: nicotine (traces), palustrine,
- Saponins triterpenic: equisitonin.

#### Uses

Horsetail has a mild **diuretic and spasmolytic action in** animal tests. The flavonoids and silicic acid contribute to the astringent effect.

It is used for infections of the urinary tract, kidney and bladder stones, wounds and burns.

## ENGLISH IVY LEAVES - Hederae helicis Folia English Ivy - Hedera helix L., Fam. Araliaceae

English Ivy is indigenous to the temperate regions of Europe, and also north and central Asia

**Triterpene saponins**: aglycone hederagenin, oleanolic acid, bayogenin, chief components hederosaponin C, additionally hederosaponin B;



- **Volatile oils steroids**: sterols, including βsitosterol, campesterol;
  - Flavonoids: including rutin

#### Uses

English Ivy is a respiratory catarrh used for the symptomatic treatment of chronic inflammatory bronchial conditions.

*Dry extracts* of Ivy Leaf are used in monopreparations and also in combination products.

Prospan, combination product Bronchipret

GOTU KOLA LEAF - Centellae asiaticae Folia Gotu Kola - Centella asiatica (L.) Urb.,

Fam. Apiaceae.

The plant is indigenous to southeast Asia, India,
Sri Lanka, parts of China, the western South Sea Islands
Madagascar

Triterpene saponins: including asiatic acid, madecassic acid (6-hydroxy asiatic acid), terminolic acid; including asiaticoside, asiaticoside A, asiaticoside B; volatile oil 0.1%.





#### Uses

The plan is used internally for rheumatism and skin diseases. Externally, the drug is used for poorly healing wounds, leprosy sores. In Asia, the drug is used to enhance urination, for physical and mental exhaustion, diarrhea, eye diseases SOAPWORT ROOT – SAPONARIAE RADICES Soapwort - Saponaria officinalis L.,

Fam. Caryophyllaceae

The plant is indigenous to the temperate regions of North America, Asia, and Europe

**Triterpene saponins** up 2 to 8%: aglycones quillaic acid, gypsogenic acid.



## Uses

As an expectorant for cough and other diseases of the respiratory tract, folk medicine internal uses also gastrointestinal disorders, liver and kidney disorders, rheumatic gout, neurasthenia. External folk medicine indications include skin rashes, eczema and as a gargle for tonsillitis

#### Orthosiphon leaf - Foium Orthosiphonis Java tea, Orthosiphon - Orthosiphon stamineus Benth. Family - Lamiaceae (Labiatae)

Orthosiphon, is a ligneous plant coming from Indonesia. It grows in India, Australia and tropical America, in damp places, rivers and marshes. They are harvested before flowering time and the natives of Java prepare them like tea.



The leaves may reach 7.5 cm in length and 2.5 cm in width. The petiole is short. The lamina is oval to lanceolate, the apex acuminate. The lower surface of the leaves is light greyish-green and the upper surface is dark green to brownish-green. The venation is pinnate with few secondary veins. Examined under a lens (103), the secondary veins, after running parallel to the midrib, diverge at an acute angle. The margin is irregularly and roughly dentate. The petioles are thin, up to 8 mm long and usually violet-coloured. Occasionally, inflorescences in clusters of bluish-white to violet flowers, not yet opened, are found.



#### Chem const.

The leaves contain terpenoids: diterpenes, triterpenes: **saponins** (ursolic acid), particularly αamyrin;

steroids (beta-sitosterol);

betaine, choline;

**lipids** - 3 %;

organic acids: benzoic acid, glycolic acid; phenolic compounds : chlorogenic acid, rosmarinic acid;



α-amyrin

**flavonoids**: flavones (apigenin, eupatorin, scutellarin); 5 to 6 % tannins; traces of **essential oil** consisted in terpenoids, notably, monoterpenes and sesquiterpenes: β-elemene, β-caryophyllene, β-selinene, γ-cadinene, humulene.

#### Uses.

Orthosiphon leaves have **powerfully diuretic functions and ease the elimination of chlorides and nitrogenous waste.** It is mainly due to their content in saponins, essential oil and potassium salts.

Orthosiphon also presents choleretic, anti-inflammatory, hypotensive and cholesterollowering virtues, it has hypoglycaemic activities. Yam rhizome with roots - *Rhizoma cum radicibus Dioscoreae* Plant Yam - *Dioscorea nipponica Makino* Family - *Dioscoreaceae* 

**Distribution. Collection.** Both wild and cultivated plants are used. Dioscorea nipponica are found in China and Russia. Dioscorea is collected in autumn, washed and dried at a temperature up to 60°C.

**Description.** The raw material of yam is represented by pieces of rhizomes up to 30 cm long and to 2 cm in diameter. The rhizomes are cylindical, slightly curved, unbranched, longitudinally wrinkled. They are covered by thin layer of cork, which usually easily breaks off. The upper surface of rhizomes bears leaf-scars. Thin unbranched adventitious roots stem from rhizomes; they are up to 40 cm long and about 1 mm in diameter. The outer surface of rhizome is lightbrown or yellowish. The fracture is even, white. The odour is weak, specific. The taste is bitter, slightly burning.

ioscorea caucasica Thomas Schoepke



R = Glu-O-Rha-O-Rha - DioscinR = Rha-O-Glu-O-Glu - Gracillin

**Chemical constituents.** Rhizome with roots contain steroidal saponin: dioscin, whose sapogenin is diosgenin.

#### Uses.

Yam rhizome with roots exert **antisclerotic** and diuretic effects. Diosgenin, obtained upon hydrolysis of dioscin, is now the **major source of the synthesis of glucocorticosteroids**, which are prepared by processes that involve microbial transformation.

#### Sarsaparilla root – *Radix Sarsaparillae* Smilax – *Smilax officinalis*, Fam. Liliaceae

The species is indigenous to tropical and subtropical regions of America, eastern Asia and India.

The species are **evergreen shrubs** with climbing branches and stipular tendrils. They have a short, creeping or ascending rhizome with numerous long roots. The branched, thorny, nodular stem is yellowishgreen. The leaves are in 2 rows. They are alternate, simple and often hardy, with 3, joined main ribs. The leaves are ovate and cordate, and petiolate. They turn into climbing tendrils above and break off at this point when they die. The flowers are white or pale green, yellow or brown. They are usually in axillary cymes or racemes, and contain 6 petals in 2 circles.

In long roots up to 6 mm, in diameter; light grayish-brown, longitudinally ridged; with few fibrous rootlets; fracture of cortex brittle, central cylinder fibrous. It is pale orange, horny cortex, a yellow band porous woody zone and a lighter colored central pith.



**Steroid saponins** 0.5-3%: chief components are sarsaparilloside, parillin, aglycones sarsapogenin;

other constituents: caffeoylshikimic acid, ferulic acid, shikimic acid, kaempferol, quercetin; resin; starch; volatile oil.



OH

OH

CH-OH

Sarsaparilla root

Uses

The steroid saponins in the drug are responsible for its irritating effect on the skin and the strong diuretic and diaphoretic effect in high doses, as well as its effect as an emulsifier and foam stabilizer. Preparations of Sarsaparilla root are used for skin diseases, psoriasis, rheumatic complaints, kidney diseases, and as a diuretic and diaphoretic.

BHC 199, BHP 1996, Martindale 35th edition

Adam's Needle leaves – Folia Yuccae filamentosae Adam's Needle - Yucca filamentosa L., Fam. Agavaceae

**Steroid saponins: protoyuccoside C**, yuccoside B, **yuccoside E**, yuccoside C, **aglycones including sarsapogenin, tigogenin.** 

> The plant is used for liver and gallbladder disorders and as raw material for semi synthesis of steroidal hormones.



#### American aloe leaves – Radix Agavae American aloe, Agava americana L. Family - Agavaceae

It is naturalized in Florida. All agaves are natives of Mexico and are cultivated for centuries. It blossoms only once in a hundred years.

The leaves of the American aloe are long, lanceolate, thick, succulent, and curved or reflected backward.

The Agava are employed in Mexico in the manufacture of a spiritous beverage known as *pulqué*. This is prepared by fermentation of the saccharine liquid, known as honey water, which exudes from the leaves and root when cut.





HO

**Chem. const.** Steroidal saponins including hecogenin, manogenin, hitogenin.

**Uses.** Steroidal saponines are the a source of **synthesis hormoff**.