EUPATORIIN · C · H · O · H · N · O

THE ACTIVE PRINCIPLE OF EUPATORIUM PERFOLIATUM

THESIS
FOR DEGREE OF MASTER OF SCIENCE
in the
SCHOOL OF CHEMISTRY.

BY
CHARLES · H · SHAMEL · B · Sc.

Professor of Chemistry, Toxicology, and Microscopy;
College of Physicians and Surgeons.—Keokuk, Ia.

1891.
The Active Principles of Poisonous and Medicinal Plants

The fact that certain plants have marked medicinal and toxic effects has been known from antiquity. When chemists, in the first half of the present century, came to search for the reasons for these remarkable properties, it was found that the specific medicinal or toxic effect of any plant...
was due to the presence in the plant of organic substances of definite chemical composition. These substances were usually present in minute quantities and were extremely difficult to obtain in pure forms. When purified, however, they are usually crystalline and possess in an intense degree the specific properties of the plants from which they were derived.

After the methods of organic analysis had been perfected sufficiently to enable the chemist to determine the ultimate composition of these bodies with accuracy, it was found that there were two
general classes of these active principles; the first containing nitrogen while the other class had none.

The first class have received the name of vegetable alkaloids and comprise the greater number of vegetable poisons and medicines—especially the more potent and those most used in medicine. The composition as well as the physiological and chemical properties of the alkaloids have been very thoroughly investigated owing to their importance both from the medical and legal standpoint; the records of the researches forming a valuable literature which may be found in well digested form
in the materia medicas, dispensatories and toxicologies.

are corn. The second class, however, are comparatively little known but indeed of relatively inferior importance and have been used in medicine very little in the isolated state, though tinctures, decoctions, or of many of the plants containing such principles are often used, especially among the electers and in household practice.

Believing, nevertheless that their increasing number and application justify it, I have endeavored to collect some of the scattered facts, accessible which bear on this division of the active principles.
The Active Vegetable Principles which do not contain nitrogen.

In the present state of chemical knowledge concerning these substances, it is extremely difficult to give any classification of them or to definitely the other almost unnumbered bodies occurring naturally in plants which are not so far as medicinal or toxic properties are concerned. Since of all but a few of the best known the chemist knows nothing except the empirical formula and perhaps a few qualitative reactions.
while of the larger number his information does not extend even this far, it will be easily understood that a properly classification according to composition and structure is wholly impossible. It is probable, however, from indications derived from a study of a few of the active principles that the chemist will ultimately be able to ascertain the structural formula of these bodies. There very likely some common principles of structure or will be discovered which will render a scientific classification possible and also enable the chemist and physiologist to account for the peculiar properties of these bodies.
At present the active principles outside of the alkaloids may be classified as follows:

- Glucosides
- Bitter principles
- Coloring principles
- The Tannic Acids
- The Vegetable Acids

We shall consider here only those that come under the glucosides and under the bitter principles. The glucosides are so named because when treated with either, dilute acids or alkalies or with organized or unorganized fermenta or warm water at 100° they split up into glucose or some
allied compound, and another substance
or substances which usually retain the
specific properties of the glucoside.
A few of the glucosides contain
nitrogen, but, adhering to the
original purpose these will not be
considered here.

The general method of separ-
arating the off-glucosides is as follows:
Benz mostly soluble in water or alcohol,
the parts of the plant containing them
is extracted by either of these fluids and
the resulting solution is precipitated
by means of neutral lead acetate
which throws down acids, tannins,
In some cases the glucoside may
now be precipitated from the filtrate.
by the addition of lead sub-acetate. This precipitate is then filtered off, washed and dried and the glycoside extracted by boiling alcohol from which it crystallizes out on evaporation and cooling. In other cases the aqueous solution is freed from excess of lead acetate by H₂S, then concentrated sufficiently on the water-bath and the glycoside allowed to crystallize out. In both cases the crude glycoside is purified by recrystallization from water or alcohol.

The so-called bitter principles are some of them capable of being extracted by the general method for the alkaloids while others are
last obtained by a process similar to that described above for the glucosides.

In the following is given detailed descriptions in condensed form of the last known of these principles.

Perrotin

Derived from the fruit of Annuaraococcus. The powdered seeds are twice extracted with alcohol, the alcohol distilled off and the residue which contains much fatty matter is boiled with water. The watery solution is precipitated by basic lead acetate and the filtrate concentrated when the perrotin crystallizes out and is purified by recrystallization.
from water. The crystals are star-like groups of needles free from water. Picrotoxin is odorless, has a neutral reaction and a bitter taste. Sparingly soluble in water and ether; easily in alcohol, amyl alcohol and chloroform, also in the alkalies and ammonia.

*Formula C₃₀H₃₄O₁₃.*

Soluble in sulphuric acid with a yellow color, a trace of potassium dichromate causes it to turn violet then brown. Nitric acid afterward adding some sulphuric acid and then an excess of strong NaOH a brick-red color is developed. It produces in animals spasm of the epileptic kind, periodic cessation of the diaphragm and lengthening of
the heart beat. There is an alternation of klonic and tonic spasms affecting the motor and reflex centers. It is comparable in strength and effect to strychnine.

Digitolins

From digitalis purpurea.

Several glucosides are found in this plant; digitolins, digitaletins, digitalcerins and digitalcosmin.

The plant is extracted with water, alcohol and the residue that remains after evaporation of the alcohol is extracted with water as long as it imparts a bitter taste. The aqueous solution is precipitated by lead acetate and after
and after filtration and removal of excess of lead by \( \text{H}_2\text{S} \), ammonia is added to neutral reaction and after filtration is precipitated by tannic acid and this precipitate is washed, pressed and mixed with some lead oxide, then boiled with alcohol. Any lead dissolved is removed by \( \text{H}_2\text{S} \) and the alcohol mostly distilled off, then allowed to evaporate spontaneously. The result is a mixture of the alkaloids named above. Melt c. 81.85°C.

By treatment of this mass the digalpin may be separated and forms aggregates of needle-shaped crystals having a neutral reaction and bitter taste. Soluble in and sparingly in water; not soluble in ether or benzole.
It gives a yellowish green color with HCl and a brown rose color with concentrated sulphuric acid and a solution in dilute sulphuric acid gives a violet color with bromine water or bromine vapor.

Formula $\text{Cu}_9 \text{H}_{18} \text{O}_{30}$

Pigalaline is a powerful stimulant of the heart and circulatory system. In poisonous doses it produces in some cases hallucination diarrhea and loss of appetite lasting two or three days. In other cases it strongly affects the nervous system producing collapse.
Elaterin

From elaterium, a substance deposited from the juice of the fruit of echolium elaterium, or squinting cucumber.

It is obtained by exhausting the elaterium with chloroform, adding ether, and collecting the precipitated which is then washed with ether and recrystallized from chloroform.

It is soluble in melted carbolic acid which on the addition of sulphuric acid turns crimson.

Phosphoric reagent gives a green color passing into brown.

Vauth in sulphuric acid gives a blue color passing into green, then brown.

Elaterin is dissolved in cone. hydrochloric.
acid, evaporated on the water-bath, and the residue washed with boiling water gives on the addition of cone. sulphuric acid an amaranth red color.

Formula $Cr_2O_7^{2-}$

Elatine is a painful hydroagogue cathartic and in large doses excites nausea and vomiting.

Aloin

From Barbadoes aloe which contain

- Barbaloin: $C_{17}H_{20}O_7$
- Radaloin: $C_{16}H_{18}O$
- Sorealoin: $C_{15}H_{16}O_6$

To obtain barbaloin, Barbadoes aloe is macerated in hot water, some sulphuric or hydrochloric acid is added.
to separate amorphous substances and 18 after standing several days in the cold the liquid was drawn off pure and after evaporation to 1/3 volume outstanding several days crusts of berberovin appeared which was filtered by recrystallization from hot alcohol.

When taken internally it produces a strong cathartic effect.

\[
\text{Convallamina} \\
\text{Convallarin}
\]

From convallaria majalis

The whole plant at blooming time is dried, powdered and extracted with water and alcohol.

The aqueous extract contains convallamarin;
the alcoholic extract convallarin. 19

The aqueous extract is precipitated by lead acetate and in the filtrate, made alkaline with sodium carbonate, muriate acid is added. The washed and dried precipitate is extracted with alcohol and, on evaporation to dryness, the residue consisting of convallamarin is purified by treatment with ether.

The alcoholic extract is also precipitated by lead acetate, the excess of lead removed by H₂S and strongly concentrated. The residue is pumped by washing with ether.

Convallamarin forms a white powder mixed with small crystals, is
of a persistent bitter taste. It is soluble in water and alcohol, not in ether. It is precipitated by tannine acid. Nitric acid
cone sulphuric acid added to the aqueous solution gives a fine violet color.
Formula \( C_{46}H_{44}O_{12} \). It splits up into sugar and convallamarin. \( C_{40}H_{36}O_{16} \).

Convallarin crystallizes in rectangular prisms, has a harsh taste and is split up by acids into sugar and convallamarin.
Formula \( C_{36}H_{31}O_{7} \).

Convallarin \( C_{28}H_{26}O_{6} \).
The effect of the combined alkaloids is purgative in doses of 2-5 g. In smaller doses it acts as an emetic. Acts like digitaline on the heart.
Rosin

From gymea anthelmintic or Rosso.

It is obtained by treating Rosso with alcohol to which caustic soda has been added. The residue after the extraction is boiled with water and the alcoholic extract and the water mixed and distilled. The residue is treated with acetic acid which precipitates the Rosin as a white flocculent mass which becomes resin-like on drying and contains undissolved microscopic crystals. Soluble in alcohol, ether and alkaline solutions; not soluble in water. Melts at 142° to a resin-like mass which when touched with alcohol assumes the form of stellate crystals. Soluble in conc. sulphuric acid.
Rosin an anthelmintic been used for the expulsion of tapeworms

Formula C3, H38, 6

Catalpin

From catalpa lignonowodes, or common catalpa

The fruit deprived of their seed or the bark is extracted with alcohol, the alcohol evaporated and the residue dissolved in water, lead acetate is added to the filtrate which is mixed to bariam carbonate and evaporated to a thin syrup which is extracted with a mixture of ether and alcohol. The residue after the evaporation of the alcohol and ether is taken up
filtered and concentrated then boiled with animal charcoal and the charcoal extracted with boiling alcohol from which the bitter principle separates in radial crystals.

Soluble in water and alcohol, with difficulty in ether. It is a glucoside. It is precipitated by lead-subacetate, reduces gold chloride on standing several days, is soluble in nitric acid. The solution becoming reddish; cone sulphuric acid produces a reddish color.
Experimental Work

The experimental work was an attempt to isolate the active principles of *iris versicolor*, or common blueflag, and of *eupatorium perfoliatum*, commonly called "bouquet".

So far as the blueflag was concerned, the attempts gave no definite results. The method pursued was as follows: The washed and finely divided root was extracted with water acidulated with hydrochloric acid. The liquid was pressed out and neutralized with sodium carbonate and extracted with ether. In some cases the fluid was concen-
trated on the water-bath. In other cases this was not done. The ethereal extract on spontaneous evaporation left an oily residue which gave reactions with several alkaloid reagents. It was present in such minute quantity that little definite information could be gained concerning it.

The aqueous extract when applied to a rabbit killed it and also small mice producing in the latter very marked purgation before death. The attempt was then made to obtain the active principle by extraction with alcohol in a continuous extraction apparatus. In the receiver of this apparatus on cooling it was found that there were
numerous clusters of needle shaped crystals deposited on the sides. These I supposed was the active principle. However after several weeks work in separating and purifying a quantity of this substance it was found to be wholly inert. The gummy mass which remained after the evaporation of the alcoholic extract was also found to be inert as much as 20 cc being injected into a rabbit with killing it or apparently producing any effect except a local infiltration at the point of injection. The conclusion then is that the active principle must be volatile or one easily decomposed by the tead and evaporating and extracting.
This view is confirmed by the fact that when the finely divided root stock is kept in the air two or three weeks it loses its peculiar burning taste that it usually makes felt while on the fauces. I have not had time to investigate this plant further.

Active Principle of Eupatorium Perfoliatum

The dried plant, gathered at blooming time was extracted by hot alcohol in a continuous extraction apparatus for several hours. The excess of alcohol was then distilled off and the thick residue was treated with water and acetone.
hydrochloric acid. A black gummy mass separated which was removed by filtration and the filtrate of its neutralization by sodium carbonate was extracted with ether. On evaporation of the ether the active principle was deposited sometimes in the form of crystalline aggregates forming yellow grains and sometimes as a yellow resinous mass. The crystalline form was analyzed for bromine and found to contain none.

It was soluble in concentrated sulphuric and hydrochloric acids. Soluble in nitric acid with the production of a reddish-brown clear solution. The nitric acid solution when allowed
to evaporate spontaneously or in a vacuum over lime, crystallizes in beautiful prisms and six-sided plates. Some of the prisms were 3 in long.

The solution of the nitrate when injected into mice kill them in a few hours. The crystals when taken into the mouth first an acid taste from the nitric and they contain followed by a bitter taste. The solution of the nitrate has a very bitter taste.

Chemical Characteristics.
The crystallized principle itself is insoluble in concentrated sulphuric or hydrochloric acids.

Dissolves in nitric acid with a clear
reddish brown color. The solution becomes crystalline on evaporation.

These crystals are soluble in water and give the following reactions:

- With phospho-molybdate acid, a green color
- With picric acid, a few needle-shaped crystals
- Anise chloride is reduced slightly giving a light coloration
- With platinum chloride, no precipitate
- With mercuric chloride, no precipitate

Hydrochloric acid and no change
- Potassium dichromate, no change

The crystals melt at 100°-103°.

The principle itself does not melt and above 250° it blackens and suffers partial decomposition.

The principle is soluble in solutions.
of the alkalies. The solution in sodium hydroxide gives the following reactions, blank tests being made in each case with sodium hydroxide alone:

With gold chloride, a black flocculent precipitate forms and causes a deep reddish coloration. Phosphomolybdic acid produces an instantaneous green color which soon fades. The other alkaloid reagents do not give any characteristic reactions.

The material used in the work was five pounds of the leaves and blossoms very kindly furnished me by Mr. E. Davis to Detroit, Mich.
Analysis

The ultimate organic analysis gave the following results:

\[\begin{array}{ccc}
C & H & O \\
25.7\% & 3.1\% & 64.2\% \\
26.2\% & 2.7\% & 64.1\% \\
\{26.9\% & 1.5\% & \} \\
26.8\% & 2.5\% & 62.2\%
\end{array}\]

\[N\text{ }1.5\% = 6.97\% \text{ }\text{HNO}_3\]

\[N\text{ }1.6\% = 7.2\% \text{ }\]

These figures indicate code \(C_{20}H_{18}O_6\) as the empirical formula of the nitrate which would require the following theoretical percents:

\[\text{C }- 26.4\% , \text{ H }2.76\% , \text{ O }- 64.9\% , \text{ HNO}_3 69.6\% = 15.4\% \text{ N}.

\*

This may be an abnormal figure was most likely caused either by a drop of the \(8\% \text{ H}_{2} \text{SO}_4\) splashing into the molder connecting tube or by mistake in reading the weights.