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The Glycosides of Asclepias Cornuti or the Common Milkweed*

By A. E. Rihn† and H. G. DeKay‡

The common milkweed has been studied for its caoutchouc content (1), fibers, seed hairs, seed oil (2) and constituents in its lactiferous sap (3). A number of papers on these topics have been published since 1910.

The common milkweed was official in the U. S. P. from 1820 to 1864. The related species, *A. tuberosa* was official in the U. S. P. until 1916 when it was placed in the N. F. In 1936, it was deleted from the N. F.

Both species are members of the genus, *Asclepias*, and belong to the *Asclepiadaceæ* family. The common species has two botanical names, *Asclepias syriaca* (Linné, 1753) and *Asclepias Cornuti* (Decaisne, 1844). It may be differentiated from other species by its erect follicles which are covered with short soft processes and mounted on recurved pedicles. The leaf is broad and rounded at the apex, and different in other minor respects (4).

The sugars of the plant were studied by Gerhardt (6).

The related species native to Europe had been studied as far back as 1825 (5). Harnack (7), Gram (8), Tanret (9), Kubler (10) studied the glycoside from *Vincetoxicum officinale*. Gram also studied the glycosides from *A. curassavica* and *A. tuberosa* and found that they possessed a similar pharmacological activity.

Kubler assigned the formula, $C_{46}H_{70}(OCH_3)_4O_{16}$, to the glycoside obtained from *V. officinale*. It has since been determined that two forms of the compound exist, one water-soluble and the other water-insoluble.

Masson (11) isolated a saponid from *V. officinale* and found that it resembled the glycoside, vincetoxin. Van Rijn (12) states that vincetoxin is apparently identical with asclepiadin, the compound obtained from *A. tuberosa* and *A. curassavica* by Gram. A number of bulletins of the United States Department of Agriculture report the isolation of toxic gycosides from species of *Asclepias* native to the West.

Asclepias Cornuti has been used in dropsy, and as an emetic and cathartic. Gram's study on the pharmacological action of the glycoside showed that it affected the respiration, causing the heart to stop in diastole. No report of a thorough study on the glycosides of *A. Cornuti* was found in the literature.

EXPERIMENTAL

STUDIES ON ASCLEPIAS CORNUTI

The plants were gathered in Tippecanoe County,

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Indiana, during August and September 1936. They were removed from the ground with rhizome and stem intact, and dried in the open air. After separation, the rhizome and stems were separately ground to a coarse powder, and when needed for treatment, were reduced to a finer powder in a hand mill. The powdered drug was first extracted with benzin to remove caoutchouc and other fatty material.

As a result of handling the fresh plants, an urticaria appeared. This affection was characterized by vesiculation and swelling of the hands, and persisted for approximately three weeks. The intense burning itch was alleviated by immersion in hot water, followed by application of Calamine Lotion.

In the processes of purification of the plant extracts, all evaporations were carried out *in vacuo*. The containing vessel was immersed in a water-bath at temperatures below 35° to 40° C. to hasten evaporation under reduced pressure.

The aqueous and alcoholic extracts of the plant left thick syrupy residues when evaporated to low volumes. Sucrose and glucose were identified in these residues. The timed osazone reaction given in Mulliken Volume I (13) was employed. It was found difficult to remove the glycoside with solvents in the presence of the hydrated sugars, since the sugars tended to form colloidal solutions in chloroform and alcohol.

STUDIES ON THE GLYCOSIDES

Part 1. Leaves and Stems

Experiment I.—The drug was percolated with lime water, the percolate acidified with acetic acid and filtered. The filtrate was treated with fuller's earth, which was collected on a filter paper, washed, dried and extracted with absolute alcohol. The bitter substance obtained did not show the properties of a glycoside.

Experiment II.—The drug was extracted with alcohol by percolation. The percolate was filtered, concentrated *in vacuo*, and allowed to evaporate spontaneously in the air until a heavy green precipitate had formed. The mixture was filtered, and the filtrate shaken out with ether. The other layer was evaporated and the residue dissolved in water and alcohol, and purified by treatment with lead acetate. The purified solution left a bitter residue which was non-glycosidal, and showed no properties of an alkaloid or tannin.

Experiments III and IV.—The drug was percolated with alcohol, and the extract mixed with magnesium oxide and evaporated to dryness. The residue was treated with absolute alcohol, and the solution purified by the lead acetate method of Rosenthaler. The filtrate obtained in this procedure was evaporated to dryness, redissolved in alcohol, and one volume of ether added to precipitate the sugars. After filtration, the filtrate was evaporated to dryness, and the residue extracted with warm distilled water. The mixture was filtered through talc on a Buchner funnel. The filtrate was colloidal and left a brown-colored residue which had the following properties:

1. It dissolved in water, giving a bitter, nauseating solution.
2. It was rendered insoluble by acids, but redissolved on addition of alkali.
3. It had no reducing power.
4. It gave negative tests with Mayer's reagent and tannic acid solution.
5. It was toxic to the common sparrow when injected into the abdominal cavity.

The residue which was not dissolved by warm water was removed from the talc by treatment with absolute alcohol. It possessed similar properties, but was non-toxic to the common sparrow.

The common sparrow was used as the test specimen in these experiments for two reasons: *first*, the isolation of the toxic substance was not accomplished until late in the year and it was impossible to obtain frogs and other accepted animals before completing the work; and *second*, we desired some subject that had similar ability of emesis as that of the human.

Inasmuch as this is the first time, so far as we know, that the sparrow has served for experimental purposes, the results may be open to question.

The observations upon the test bird were compared in each case with a control. The symptoms and results led us to conclude that the substance isolated was toxic.

Part 2. Glycoside of the Rhizome

Experiments I to IV.—As an introductory study, these experiments were conducted on a commercial preparation of *A. tuberosa*, "Asclepidin" (Parke, Davis Co., Detroit). Several glycosidal preparations were obtained, using the general method employed in the treatment of *Asclepias Cornuti* (cf. Exp. V). These products were similar in properties to asclepiadin, but varied in their toxicity when tested on the common sparrow.

The yield of the impure glycoside was approximately 0.001%.

Experiment V.—In this experiment, a glycoside was obtained from the rhizome of *Asclepias Cornuti*. The powdered drug was percolated with alcohol, and the percolate evaporated to low volume, filtered, and then evaporated to dryness in presence of magnesium oxide. The dried residue was extracted with alcohol, and the solution obtained filtered with the aid of talc. Sufficient water was added to the filtrate to make an 80% alcoholic solution, and this was purified by the lead acetate process. The purified solution was evaporated to drynes and extracted with absolute alcohol. The alcoholic solution was evaporated and the residue again extracted with absolute alcohol. Chloroform was added and the mixture filtered with the aid of talc. One volume of ether was added to three volumes of the filtrate, the mixture filtered, and the filtrate evaporated to dryness. The residue was dissolved in

chloroform, treated with activated charcoal and filtered through a charcoal mat on a Buchner funnel. The filtrate was evaporated to dryness and extracted with ether to remove impurities. The residue not dissolved by the ether was taken up in chloroform and the solution washed with several small portions of water. The chloroform was removed *in vacuo*, the residue obtained was dissolved in a known quantity of alcohol. Two volumes of ether were then added, the mixture shaken out with water and the aqueous layer filtered into a small dish. This was placed in a vacuum desiccator and evaporated to complete dryness. The transparent residue possessed the following properties:

1. It was tan in color.
2. It dissolved readily in water, alcohol and chloroform, but not in ether.
3. When hydrolyzed by boiling with dilute sulfuric acid, it reduced Fehling's solution. The untreated residue had no reducing power.
4. The solid and its solution possessed a bitter taste.
5. Mayer's reagent gave a negative text.
6. Tannic acid produced a copious white precipitate.
7. It showed the following actions on being heated:

Softening	94–100° C.
Fusion	107–108° C.
Swelled	120° C.
Darkened	160–180° C.
Reddish brown	203–205° C.

The heated mass was dissolved in water, and the solution tested. The substance no longer showed the properties of a carbohydrate.

8. After standing for eight days in a stoppered vial, it exhibited the same properties as before and was found to be toxic to sparrows.

The yield of the glycoside was approximately 0.002%.

In several other experiments, the glycoside was precipitated by addition of tannic acid solution to a purified extract of the rhizome. The precipitate was collected and treated with litharge. The dried mass was then extracted with absolute alcohol but no glycoside was obtained. Hence, it is apparent that this method should be used with caution.

CONCLUSIONS

1. Sucrose and glucose were present in the aqueous and alcoholic extracts obtained from parts of *A. Cornuti*.
2. The leaves and stems contain a bitter principle which may be divided into a toxic and a non-toxic fraction.
3. The leaves and stems examined did not contain any significant amount of glycosides.
4. A toxic glycoside which resembled asclepiadin in properties was isolated from the rhizome of *A. Cornuti*, apparently in impure form.
5. The overground portions of *A. Cornuti* contained a principle which was irritating to the dermis.

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Studies on Cantharides. I. The Titration of Cantharidin*,†

By Benjamin P. Hecht and Lloyd M. Parks‡

INTRODUCTION

For many centuries the drug Cantharides, commonly referred to as Spanish or Russian Flies, has been used in medicine. Its history dates at least as far back as the time of the early Greeks, as shown in the writings of Hippocrates.

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