



3 1151 02690 5848





SOME CONSTITUENTS OF THE POISON IVY PLANT.

(RHUS TOXICODENDRON)

---CCO---

D I S S E R T A T I O N

---CCO---

Submitted to the Board of University Studies of the
Johns Hopkins University in Conformity with the Re-
quirements for the Degree of Doctor of Philosophy.

By

William Anderson Syme.

1906.

C O N T E N T S .

	Page
Acknowledgments-----	3
Literature-----	4
Introduction-----	5
Work of Khittel-----	11
Work of Maisch-----	12
Work of Pfaff-----	14
Experimental:-----	
Gallic Acid-----	24
Fisetin-----	28
Rhamnose-----	32
The Poison-----	40
Potassium Permanganate as a Remedy for Rhus Poisoning-----	53
Biography-----	55

A C K N O W L E D G M E N T S .

----0000----

The author desires to avail himself of this opportunity to tender his thanks to those under whose guidance he has worked while a student at the Johns Hopkins University, namely to Professors Remsen, Morse, Jones, Andrews, and to Doctors Tingle and Acree for instruction in lecture room and laboratory.

He is especially indebted to Dr. S. F. Acree, at whose suggestion this research work was undertaken, for counsel and assistance in its prosecution.

He would also thank Messrs. Parke, Davis and Co., of Detroit, Mich., for the preparation of the crude material used in this investigation, and the U. S. Department of Agriculture, Washington, D. C. for electrotypes of figures 17, 18, and 19 in Bulletin No. 20, Division of Botany.

L I T E R A T U R E .

---000---

- Acides Gummiques, Garros (Dissertation) 1895.
American Chemical Journal.
American Journal of the Medical Sciences.
American Journal of Pharmacy.
Annalen der Chemie und der Pharmacie (Liebig).
Annales de Chimie et de Physique.
Berichte der deutschen chemischen Gesellschaft.
Biochemie der Pflanzen (Czapek) 1905.
Brooklyn Medical Journal.
Bulletin de la Societe Chimique.
Bulletins 20 and 26 U. S. Department of Agriculture,
Division of Botany.
Chemie der Zuckerarten, Von Lippmann, 1905.
Chemiker-Zeitung.
Comptes rendus.
Industries of Japan, J. J. Rein.
Journal of the Chemical Society.
Journal of Eperimental Medicine.
Les Sucres, Maquenne, 1900.
Manual of Botany, 6th Edition, Gray.
Medical and Surgical Reporter.
New York Medical Record.
Proceedings of the American Pharmaceutical Association.
Treatise on Chemistry, Roscoe and Schorlemmer.
Ueber Mategerbstoff, Reuchlin, (Dissertation) 1904.

I N T R O D U C T I O N .

---COC---

Plants belonging to the natural order Anacardiaceae (Cashew ^{family} or Sumach family) are found in all the temperate climates of the world and quite frequently in semi-tropical climates. Many of these plants play important parts in economic botany, yielding dye-stuffs, tanning material, wax, varnish, and drugs. Several species are poisonous. At least three poisonous species of the genus RHUS are found in the United States. These three are all common and well known plants, but confusion frequently arises concerning them on account of the different names by which they are known in different localities. For example, poison ivy (Rhus toxicodendron or Rhus radicans) probably the best known poisonous plant in America, being found in all the States except those in the extreme West, is often confounded with and popularly called "poison oak". The true poison oak is the Rhus diversiloba of the Western States.¹ The third and most poisonous species of this plant is Rhus venerata or Rhus vernix; it is the Rhus vernicifera of Japan, from which Japanese lac is obtained. It is popularly known in the United States as "poison elder". It grows in swamps from Canada to Florida.

1. Chesnut, Bull. No. 20, U. S. Dept. of Agr., Div. of Botany.



Fig 1.



Fig 2.

As the poison ivy is by far the most common of these plants in the Eastern States, a brief description of it is given here: ¹ A shrub climbing by rootlets over rocks, etc., ~~xx~~ or ascending trees, or sometimes low and erect; leaflets 3, rhombic-ovate, mostly pointed, and rather downy beneath, variously notched, sinuate, or cut-labeled - high climbing plants (R. radicans) having usually more entire leaves. It is found in thickets, low grounds, etc., Greenish flowers appear in June.

(Space for cuts)

Fig. 1. Poison Ivy, Rhus radicans or Rhus toxicodendron.
a, spray showing aerial rootlets and leaves
b, fruit. both one-fourth natural size.

Fig. 2. Poison Oak, Rhus diversiloba, showing leaves, flowers, and fruit, one-third natural size.

(Crescent Bulletin No. 20, Division of Botany, U. S. Department of Agriculture)

1. Gray, Manual of Botany, 6th Edition p. 119.



Fig. 3

Fig. 3. Poison sumach (*Rhus vernix*) showing
leaves, fruit, and leaf-scars, one-fourth
natural size.

(Chestnut Bulletin No. 20. Division of Botany,
U. S. Department of Agriculture)

In the general description of the order Anacardiaceae,
Gray¹ says: "Juice or exhalations often poisonous." Whether it is contact with some part of the plant, or with the exhalation from the plant, that causes the well known skin eruption has been a topic for discussion ever since its source was known. On account of its intangible nature there has been more speculation than experimental evidence bearing on this question, although a few investigations have been made with the object of isolating the poison. It is most generally believed that the exhalations are poisonous. Dr. J. H. Hunt² states that the

1. Gray, Manual of Botany, 6th Edition, p. 119.

2. Brook. Med. Jour. June 1897.



exhalations have been collected in a jar and found to be capable of inflaming and blistering the skin of an arm plunged into it.

Prof. J. J. Rein¹, in his treatise on Lacquer Work, describes the poison of the Japanese lac tree, Rhus vernicifera, as being volatile, as do also the Japanese chemist Yoshida² and the French chemist Bertrand³. Recent work by Prof. A. B. Stevens⁴, however, seems to show that this poison is not volatile, and is similar to, if not identical with that obtained by Pfaff⁵ from Rhus toxicodendron and Rhus venenata.

Not many cases of internal poisoning by Rhus toxicodendron are on record in medical literature. Two cases of poisoning from eating the fruit of this plant have been described⁶. The subjects of these cases were two children who had eaten nearly a pint of the fruit. The symptoms are described in detail, being in general, similar to those of alkaloidal poisoning. Warm water was given to promote emesis; afterwards large quantities of carbonate

-
1. Rein, The Ind. of Jap., p. 338, et seq.
 2. H. Yoshida on Urushi Lacquer, Jour. Chem. Soc., 1883, p. 472.
 3. Ann. de Chem. et de Phys., Series VII, Vol. 12, p 125, 1897.
 4. Amer. Jour. Pharm. 78, p. 53, Feb. 1906.
 5. An account of Pfaff's work will be found in another part of this paper.
 6. Amer. Jour. Med. Sci. 51 (1866), p. 560.

of soda were given in solution under the belief that it was an antidote to the poison. Otherwise they were treated on general principles. Both children recovered.

Another case of internal poisoning is the following:¹
Three children drank an infusion of the root of poison ivy thinking it was sassafras tea. The first of these cases was diagnosed as measles, but on the appearance of similar symptoms in the sisters of the first patient, the cause of the trouble was found. All recovered.

Dr. Pfaff² explains the few fatal cases that have followed Rhus poisoning on the assumption that enough of the poison was absorbed through the skin to cause renal complications in persons having chronic kidney trouble. He showed that the poison, when given internally, produces a marked effect on the kidneys, causing nephritis and fatty degeneration of this organ.

The irritating action of poison ivy has been attributed at different times to the "exhalation", to a volatile alkaloid,³ to a volatile acid, and to a non-volatile oil. Pfaff³, who made the most recent investigation of this poison, obtained from the plant a non-volatile oil having the same action on the skin as the plant itself. He found this oil in all parts of the plant and concluded that it was the active

-
1. Med. and Surg. Rep. 17, Nov. 1867.
 2. Jour. Exp. Med. 2, (1897), p. 181.
 3. Loc. cit.

principle, and that one could be poisoned only by actual contact with some part of the plant. He assumed minute quantities of pollen dust to be in the air to account for the cases of "action at a distance" so frequently quoted. Pfaff says: "In my opinion, it is more than doubtful if ever a case of ivy poisoning has occurred without direct contact with the plant or with some article that has been in contact with the plant. The long latent period of the eruption in some cases may obviously render mistakes extremely easy as to the occasion when contact with the plant really occurred." Granting, however, that the active principle is practically non-volatile when isolated from the plant, we cannot say positively that it is not volatile in the juices of the plant, or under the influence of vital forces. It is quite conceivable that the water transpired by the leaves of the plant may carry with it a quantity of the poison sufficient to produce the dermatitis on a person very susceptible to its action. It is also conceivable that a volatile poison manufactured by a living plant could become non-volatile by changes in it consequent upon the death of the plant.

Up to the present time, only three important chemical investigations of the active principle of Rhus toxicodendron have appeared in medical and chemical literature, these being the researches of Dr. J. Khittel, J. M. Maisch, a pharmacist, and Dr. Franz Pfaff, of the Harvard University Medical School, to whose work reference has been frequently

made. The chemical work of these investigators and their conclusions are given here in some detail for the sake of completeness.

Khittel's Investigation.

The first attempt to find the poisonous constituent was made by Khittel in 1857. His work was published in Wittstein's Vierteljahrresschrift fur praktische Pharmacie, VII, 348-359¹. Khittel obtained 37-1/2 oz. of fresh leaves of poison ivy from the botanical garden in Munich, dried them, and got a residue of 9-1/2 oz. which he analyzed. Not detecting anything to which the poisonous qualities of the plant could be attributed, he made another series of experiments which, as he thought, showed that a volatile alkaloid is the poisonous constituent. It was obtained by the following process: "3 oz. of the powdered leaves were infused with hot distilled water, after three days strained, expressed, the liquid evaporated to 3 oz., and with the addition of potassa, carefully distilled to one-half. The clear, colorless distillate had an alkaline reaction, and an odor resembling henbane or hemlock. It was saturated with sulphuric acid, evaporated and treated with a mixture of equal quantities of alcohol and ether which left sulphate of ammonia behind, the solution was evaporated spontaneous-

1. A free translation of this paper is given in Amer. Jour. Pharm. for 1858, p. 542.

ly, distilled with potassa, the alkaline distillate neutralized with hydrochloric acid, and a precipitate could now be obtained with chloride of platinum. Want of material prevented further experiments."

The editor of the American Journal of Pharmacy inserts the following note: "It would have been more satisfactory if the author had given some physiological evidence of the poisonous nature of the alkaloid substance obtained. It is quite interesting to hear that the hitherto intangible venom of this plant has at last been detected."

1
Work of Maisch.

The next investigation of this plant was made by Maisch in 1864. He criticizes Khittel's experiments as follows: "It is well known that the exhalations of Rhus toxicodendron exert a poisonous influence on the human body; the poisonous principle must, therefore, be volatile and, at the same time, be naturally in such a loose state of combination as to be continually eliminated and separated with the usual products of vegetable exhalations. It is natural to suppose that during the process of drying, the greatest portion of the poisonous principle should be lost. The loss must be still greater if the dried leaves are powdered, a hot infusion prepared from them, and this infusion evaporated

1. Proc. Amer. Pharm. Assn. 1865, p. 166, and Amer. Jour. Pharm. 1866, p.4.

down to the original weight of the dried leaves. It is obvious that Dr. Khittel could not have selected a better method for obtaining the least possible quantity of the poisonous principle, if, indeed, it could be obtained by this process at all."

Maisch then worked up 8-3/4 oz. of the leaves of the plant in a way to get the alkaloid, making some improvements on Khittel's method, but failed to find it. Believing that the poison was a volatile acid, he enclosed some fresh leaves of the plant in a tin box with several test papers. The blue litmus paper became red showing the presence of an acid. He concluded from this experiment that the exhalations of the leaves contained a volatile organic acid which he thought was the poisonous substance. To determine this point, he prepared the acid in larger quantity by macerating the leaves with water, expressing and distilling the expressed juice. He was poisoned in doing this work although he had not been affected by handling the living plant and had considered himself immune. He obtained an acid which investigation showed to be somewhat like formic acid, more like acetic acid, but having some reactions different from both. "Taking all the reactions together, it is unquestionably a new organic acid for which I propose the name of Toxicodendric Acid," writes Maisch. He further says: "That it is the principle to which poison oak owes its effects on the human system

was proved to my entire satisfaction by the copious eruption and formation of numerous vesicles on the back of my hand, on the fingers, wrists, and bare arms while I was distilling and operating with it. Several persons coming into the room while I was engaged in with it were more or less poisoned by the vapours diffused in the room; and I even transferred the poisonous effects to some persons, merely by shaking hands with them.

"The dilute acid, as obtained by me, and stronger solutions of its salts, were applied to several persons, and eruptions were produced in several instances, probably by the former, though not always, which was most likely owing to the dilute state of the acid. Whenever this was boiled, I always felt the same itching sensation in the face, and on the bare arms, which I experience on continual exposure of my hands to the juice of the plant."

PEAFF'S WORK.

By far the most valuable work on Rhus toxicodendron is that of Pfaff. From a clinical study of Rhus poisoning, Pfaff came to the conclusion that the poison must be a non-volatile skin irritant. The more volatile the irritant, the quicker is its action on the skin. Formic acid acts very quickly; acetic acid, less volatile than formic, acts more slowly, but still much more quickly than poison ivy, the latent period of which is usually from two to five days. Pfaff thought that the volatile acid obtained by Maisch

might have contained some of the poisonous principle as an impurity, but that it would not produce the dermatitis if prepared in a pure state. He therefore prepared a quantity of the acid by distilling the finely divided fresh plant with steam. The yield was increased by acidulating the mixture with sulphuric acid before the distillation. The acid distillate so obtained was freed from a non-poisonous oily substance by shaking the solution with ether. Barium and sodium salts were made by neutralizing the acid, and were purified by crystallization. Analysis showed them to be salts of acetic acid, and they gave the characteristic tests for this acid. The toxicodendric acid of Maisch was thus shown to be acetic acid, and was therefore not the poisonous principle of the plant.

Pfaff obtained the active principle by the following process: The plant was extracted with alcohol, the alcohol was distilled off, and the residue was taken up in ether. The ether solution was washed with water and dilute sodium carbonate solution, and the ether was evaporated. An oily, black, poisonous substance partly soluble in alcohol was obtained. To get the active principle in a pure state, this residue was extracted with alcohol and filtered and the filtrate was precipitated fractionally by lead acetate. The final precipitates consisted of the lead compound of the poison in a pure state. On decomposing the lead com-

pounds with ammonium sulphide, shaking out with ether, and letting the ether evaporate spontaneously, a non-volatile oil was obtained which gave the characteristic skin eruptions. The pure lead compounds made in different preparations were analyzed and assigned the formula $C_{21}H_{30}O_4Pb$. The oil itself was not analyzed. Pfaff proposed the name Toxicodendrol for the oil. He found that it was not volatile, was decomposed by heat, was soluble in alcohol, ether, chloroform, benzene, etc., but insoluble in water. Its effects upon the human skin were studied in many experiments upon himself and others. It was shown that an exceedingly minute quantity of the poison will produce the dermatitis, even 1/1000 mg. applied in olive oil being active. The oil was given internally to rabbits, its effects being most marked on the kidneys.

The oil obtained by Pfaff from Rhus venenata seemed to be identical with that from Rhus toxicodendron.

EXPERIMENTAL.

The writer's investigation was undertaken with the object of attempting to throw more light on the chemical nature of the poisonous substance found in Rhus toxicodendron. Soon after commencing work, however, it became apparent that the poison could be more intelligently studied if the sub-

stances associated with it in the plant were first identified; the scope of the work was therefore extended to an investigation of the other constituents of the plant, and it was hoped that a knowledge of the properties of these constituents would suggest a more economical way of getting the poison than the method of fractional precipitation.

The crude material for this work was prepared by Messrs. Parke, Davis and Co., of Detroit, Mich. according to special instructions submitted to them: 67-1/2 lbs. of fresh leaves and flowers of poison ivy were collected near Detroit and carefully inspected by a competent botanist. This material was thoroughly macerated and put into ten-liter bottles with ether. The mass was thoroughly shaken, water being added to make it more mobile. The ether was then separated off and the extraction was repeated three times in the same way to insure complete removal of the toxicodendrol. The ether extracts were combined, thoroughly dried with anhydrous sodium sulphate, and the ether was distilled off, the temperature being kept below 40°C during the entire distillation. The residue after the removal of the ether was a thick, black, tar-like mass, weighing 3 lbs. 11 oz. In extracting the plant, about twenty-four gallons of ether were used. It is a significant fact in regard to the volatility of the poison that during the process of preparing this material none of the employees engaged in the work were in any way affected,

since proper precautions were taken and the utensils were handled with rubber gloves.

The crude ether extract¹ was shipped to Baltimore in August and was kept in a cool place until November when the investigation was begun. When the bottle was opened, there seemed to be an escape of a vapor and a nauseating odor suggesting crushed green leaves pervaded the atmosphere. Some days later, irregular red patches appeared on the face though a mask of cotton cloth was worn during the work, and the hands were protected by rubber gloves.

Assuming from Pfaff's work that this original material contained the non-volatile oil toxicodendrol, the first experiment was to ^{try to} distil it out under diminished pressure. For this purpose, an Anschutz distilling bulb containing ten grams of the tar was connected with a vacuum pump. After a pressure of 2 mm. had been established the bulb was gradually heated in a bath of Wood's metal. Nothing distilled over. The material began to carbonize at a temperature of 140°-150°

It was then thought that perhaps the oil could be converted into an ester which might be more volatile and could be distilled out. 20 grams of the original material were dissolved in 100c.c. of absolute alcohol containing 3 gm. of hydrochloric acid gas, and the mixture was heated 10 hours on a water-bath under a return condenser. After the heating, the

1. This will be designated as the original material.

mixture had a delightful ethereal odor. The flask was corked and left standing several weeks while other work was in progress. The ester solution was then put in a vacuum desiccator over sulphuric acid and the alcohol evaporated. A black tarry solid mass was left having the ester odor. It was extracted with warm water and filtered from insoluble tar. The filtrate had a green color and the ethereal odor. It was shaken out with ether; the ether layer had a blood-red color while the water layer was deep green. The extraction with ether was continued until the water layer was no longer green. The combined ether extracts were evaporated ~~in a~~ ~~vacuum~~ desiccator without heat. A black tar-like solid was left very much like the original material but it had the ester odor. It was partly soluble in water and readily soluble in alcohol. The alcoholic solution was tested on the skin and found to be not poisonous. The ester, or mixture of esters, was not investigated further in this connection, but was later shown to give the reactions for gallic acid and methyl furfurol. These reactions will be referred to in connection with other experiments.

After a few other preliminary experiments, it became evident that the original material was a complex mixture of substances and that it would have to be fractionated by some means and the fractions studied separately.

A portion of the original substance was treated with 50 per cent alcohol and was found to be partly soluble in

this medium. The solution was filtered from insoluble tar. A portion of the yellow filtrate gave a reddish yellow precipitate with lead acetate. The alcoholic solution was distilled in an Anschütze flask under diminished pressure; a yellow liquid condensed in the arm of the flask while most of the alcohol was collected in a bottle connected with the arm. The yellow liquid was acid to litmus. Water was added, the solution was shaken out with ether and the ether was evaporated. When the small residue was completely dry it was a yellow solid soluble in dilute alcohol and acid to litmus. The substance was not volatile enough to justify the use of this method for getting it.

Chlorophyll could not be removed from the original substance because the solvents for Chlorophyll such as alcohol, ether, fats, petroleum, and carbon bisulphide dissolve large quantities of the mixture.

A precipitate obtained by adding lead acetate to a filtered solution of the original substance in 50% alcohol was suspended in water, decomposed by hydrogen sulphide, shaken out with ether and the ether evaporated. The residue appeared at first to be a yellow oil, but on complete evaporation of the ether in a desiccator, a yellow solid was obtained apparently the same as that obtained by vacuum distillation.

A solution of the original material in 50% alcohol was filtered through bone-black and the filtrate was colorless. Examination showed that everything had been removed by the

bone-black and the filtrate was apparently pure alcohol and water.

In precipitating an alcoholic solution of the crude material with a solution of lead acetate, it was noticed that at least two kinds of precipitates were formed. The part that went down first was darker in color than that ~~thrown~~ down later. Pfaff used the last fractions in obtaining his oil and stated that these precipitates consisted of the lead compound of the oil in a pure state. It was found by experiment that the darker part was soluble in ether while the lighter part was not. This indicated that the darker part consisted of tarry matter which was brought down mechanically or separated out on the dilution of the alcoholic solution by the lead acetate solution, or was perhaps a lead compound soluble in ether. To test this point an experiment was made as follows: Some of the crude material was thoroughly extracted with 50% alcohol. The tar insoluble in 50% alcohol was then treated with 95% alcohol; most of it dissolved; the solution was filtered and lead acetate in 50% alcohol was added. A greenish colored precipitate was formed which was filtered off and found to be completely soluble in ether and soluble to a considerable extent in strong alcohol. These experiments suggested that the light colored lead compound which was thought to contain the poison could be purified by extraction with ether in a Soxhlet apparatus more conveniently than by the tedious process of

of fractional precipitation. Further preliminary experiments showed that 50% alcohol extracted from the original material all of the substance or substances which gave the light colored precipitate and dissolved only a small amount of the tar.

288 gm. of the crude material were then extracted several times with 50% alcohol and filtered; the insoluble tar was washed and saved for examination. To the filtrate was added an excess of a solution of lead acetate in 50% alcohol. The large precipitate, which will be designated as "precipitate A", was filtered and drained by suction in a Büchner funnel. The alcoholic "filtrate A" was saved. Precipitate A was extracted with ether in Soxhlet extractors until the ether came over practically colorless, the operation being interrupted from time to time to stir up the precipitate in the thimble. The green colored ether solution was saved for future examination. The lead precipitate, after extraction with ether and drying, weighed about 116 grams. A portion of this lead compound was decomposed by hydrogen sulphide in a mixture of water and ether which was well shaken during the operation. The ether was separated, filtered, and evaporated under diminished pressure without heat, and there remained a yellow oily looking residue having a pleasant odor. When the ether and water were completely removed in a vacuum desiccator a hard, brittle, yellow resin weighing about 16 grams was

obtained. It was soluble in alcohol, had a strong acid¹ reaction and was free from nitrogen and sulphur. The nitrogen tests were made by the Lassaigh and soda lime² methods, and the sulphur test was made with sodium nitroprusside after fusing the residue with sodium. The main portion of the lead compound was decomposed under alcohol by hydrogen sulphide, filtered, and the alcoholic filtrate evaporated in vacuo. The same yellow acid resin was obtained. Experiments continuing through several weeks were made in applying solution of this ~~kin~~ resin to rats, rabbits and guinea pigs. Finding it to be without effect upon these animals it was tried on the writer and found to be not poisonous³. In the meantime the resin was being studied in the laboratory.

1. Nitrogen was found very readily by the soda lime test in the ~~tar~~ left after extracting the original material with 50% alcohol, but was not found by the Lassaigh test.
2. Stevens. Amer. Jour. Pharm. 77, 255, June 1905.
3. Whenever it is stated in this paper that a solution was poisonous or not poisonous, the test was made by the writer upon himself.

GALLIC ACID.

An alcoholic solution of the resin was just neutralized with potassium hydroxide. During the titration, the solution rapidly became dark brown. After neutralization it was shaken with ether; the water solution remained brown while the ether layer was nearly colorless and contained practically no dissolved substance. A portion of the water solution of the potassium salt on being acidified with sulphuric acid and standing over night, deposited a slight precipitate. The solution of the potassium salt gave a heavy precipitate with lead acetate somewhat similar to the original lead precipitate A, and also slight precipitates with salts of zinc, mercury, copper, and ~~silver~~ silver (with reduction). It gave a bluish-black color with impure ferrous sulphate and a dark color with ferric chloride. It reduced ammoniacal silver nitrate and Fehling solution. These experiments indicated the presence of a tannin compound. An alcoholic solution of the resin gave the same color reactions with iron salts as did the potassium salt. To determine which one of the tannin compounds was present was a matter of some difficulty since the di- and tri-hydroxybenzoic acids have in general the same color reactions. The presence of other plant substances in the solution also interferes with the color tests, and finally, a substance which gives a blue color with iron salts and one giving a green color may be found

together in the same plant.¹ Further tests with a solution of the resin in dilute alcohol, and with a water solution of the acid precipitated by adding sulphuric acid to a solution of the resin in potassium hydroxide, led to the conclusion that the acid is gallic acid. These tests were the following: (1) Boiling with an excess of potassium hydroxide gave a black substance (tauromelanic acid). (2) The acid was not precipitated by gelatin. (3) On addition of potassium cyanide a transitory red color appeared which reappeared on shaking with air. Gallic acid is distinguished from tannic acid by tests (2) and (3). At later stages in the work the potassium, barium, and sodium salts of gallic acid were obtained, and finally the pure acid was made by decomposing the sodium salt with sulphuric acid and **cr**ystallizing from water. A portion of the acid so obtained was further purified by dissolving in absolute alcohol and pouring into absolute ether.² The melting point behavior of the acid corresponds with that of gallic acid; it melted with decomposition at about 230°. For further identification, some of the acid was converted into an ester by the following process: It was dissolved in 80% alcohol, hydrochloric acid gas was passed in, and the solution was heated an hour on the water bath. It

1. Liebig's Annalen, CXI, p. 215.

2. Uber Mategerbstoff, p. 20.

was then evaporated to a small bulk, neutralized with barium carbonate and extracted with ether. The ether, on evaporation, left the ester which was crystallized from water and dried in a desiccator over sulphuric acid. The anhydrous ester agreed in melting point (156° - 159°) and other properties with the ester of gallic acid described by Grimaux¹. For the sake of comparison, an ester was made from gallic acid obtained from another source and the two agreed in properties. A mixture of the two esters melted within the limits given for the ester of gallic acid.

While the tests leading to the identification of gallic acid were being made, another series of experiments was in progress. Eleven and one-half grams of the resin obtained from lead precipitate A by decomposition with hydrogen sulphide were treated with 0.1 n. potassium hydroxide added from a burette until the acid was exactly neutralized. All went into solution. On shaking with ether some of the potassium salt separated out and was saved for examination. The solution became brown on exposure to air and got darker as the work proceeded. The acid in solution as a potassium salt was precipitated out in four fractions by adding for each fraction one-fourth the amount of ^{0.1n} 0.1n sulphuric acid required to neutralize the potassium hydroxide used.

1. Bull. Soc. Chim. (II) Vol. 2, 95 (1864).

The precipitates were filtered off and examined. The first was small in amount, gummy and hard to filter. The solution was shaken with ether after each precipitate had been filtered off. The succeeding precipitates were in better condition, but were not pure. All appeared to be impure gallic acid which had become brown by absorption of oxygen. They were saved however to be tested for poison. After the last fraction had separated, the filtrate was shaken several times with ether and saved for further examination, which will be described under "Rhamnose". This filtrate is designated as B.

At this stage of the work a portion of the resin obtained from lead precipitate A was tested and found to be not poisonous as already mentioned. By this test, all the substances contained in the lead precipitate A after its extraction with ether in the Soxhlet apparatus, were eliminated from the possible poisonous substances. The poison must therefore have been extracted by the ether.

A fresh portion of the original poisonous material was treated with 50% alcohol and filtered from insoluble tar. The filtrate was precipitated in six fractions by lead acetate. The last fractions were lighter in color and apparently much purer than the first. The sixth lead precipitate was decomposed by hydrogen sulphide, the light-yellow water solution was tested and found to be not poisonous. It gave

the characteristic reactions for gallic acid. The poison, if precipitated at all by lead acetate, must have gone down in one of the preceding fractions. Later experiments showed that it is brought down partly mechanically and partly as a lead compound in the first precipitates.

FISETIN.

Having identified gallic acid, and not finding any other phenol derivative in the lead precipitate, some of the original material was extracted with hot water to remove gallic acid and filtered from tar while hot. The filtrate had a deep yellow color. On cooling over night, an olive green precipitate separated out which was dried and found to be a light powder. It was practically insoluble in cold water, soluble with great difficulty in boiling water from which it separated in yellow flakes, slightly soluble in ether and in acetic acid, but readily soluble in alcohol. The solutions were not acid to litmus, gave a dark color with ferric chloride, an orange-red precipitate with lead acetate which was easily soluble in acetic acid, and an orange yellow precipitate with stannous chloride. These properties and reactions indicated that the substance was the dye-stuff fisetin and that it occurs in the free state in this plant though it is usually found as a glucoside of fisetin combined with tannic acid. A compound of this kind was found in

Rhus cotinus and named "fustin-tannide" by Schmid¹. He showed that the fustin-tannide could be decomposed by acetic acid into tannic acid and a glucocide, fustin $C_{46}H_{42}O_{21}$. Fustin, on heating with dilute sulphuric acid, gave fisetin and a sugar supposed to be rhamnose. Fisetin was also found² as a glucoside compound in Rhus rhodanthema by Perkin.

The yellow substance which separated from the boiling water solution was further purified by dissolving in a small quantity of hot alcohol and adding hot water. On cooling, the yellow substance separated out in a flocculent condition. Examined under the microscope, the flakes appeared to be made up of masses of fine crystals.

An alcoholic solution of the substance gave a black color with ammonia which became red on addition of more ammonia. Concentrated acids intensified the yellow color of the alcoholic solution. Fehling solution and ammoniacal silver nitrate were reduced by it. Potassium hydroxide added to an alcoholic solution gave at first a deep red color accompanied by a green fluorescence which disappeared leaving a yellow liquid. With an excess of caustic potash, the red color returned and was permanent. These reactions are

-
1. Berichte 19, 1735 (1866).
 2. Jour. Chem. Soc. 71, 1194 (1897).

characteristic for fisetin.¹

Furthermore, fisetin should give protocatechuic acid and phloroglucinol by fusion with caustic potash under proper conditions.² The experiment was carried out as follows: 2 grams of fisetin ~~was~~^{was} gently heated in a nickel crucible with 6 grams of caustic potash dissolved in 6 c.c. water. An inflammable gas, apparently hydrogen, was evolved during the fusion. The pasty mass was dissolved in water, acidified with sulphuric acid, and filtered. The filtrate was shaken out with ether containing one fourth its volume of alcohol. The ether was evaporated and the residue was extracted with warm water and filtered. Lead acetate was added to the filtrate to precipitate protocatechuic acid, while phloroglucinol remained in the filtrate from this precipitate. The lead precipitate was suspended in water, decomposed by hydrogen sulphide, filtered, and evaporated to obtain protocatechuic acid. That the substance obtained was protocatechuic acid was shown by the following characteristic tests: (1) It gave a greenish brown color with ferric chloride; on addition of one drop of a dilute solution of sodium carbonate, the color became dark blue; on adding more sodium carbonate the

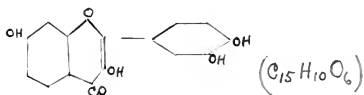
1. Berichte 19, 1740.

2. Ibid. 1747; Annalen, 112, 97.

color became red. (2) A violet color was obtained when a solution of the acid was treated with a drop of sodium carbonate solution and then with a drop of ferrous sulphate. (3) It reduced ammoniacal silver nitrate. (4) It did not reduce Fehling solution.

The filtrate supposed to contain phloroglucinol was treated with hydrogen sulphide to remove lead, filtered, and shaken with ether. The residue left on evaporating the ether was taken up in water. This solution gave the following reactions characteristic for phloroglucinol: (1) It reduced both silver nitrate and Fehling solution. (2) It colored pine wood moistened with hydrochloric acid red. (3) It gave a beautiful red color with vanillin and hydrochloric acid, and (4) a deeper red color with oil of cloves and hydrochloric acid, becoming purple on standing. (5) It gave a violet color with ferric chloride.

The substance is then, without doubt, fisetin. The formula¹ of fisetin is supposed to be



1. Biochem. Pflanz. II, 521.

RHAMNOSE.

It was stated above that Semid obtained a sugar solution by the decomposition of a fisetin-glucoside from Rhus cotinus, and Perkin obtained the same from a glucoside in Rhus rhodantha. These investigators thought that the sugar was isodulcitol or rhamnose but they did not isolate it on account of the small quantities of material at their disposal. Moreover, the sugar is very hard to crystallize in the presence of other soluble substances and is not found in large quantity in plants. Maquenne¹ could obtain only 15^{to}/₂₀ gm. of rhamnose by working up 1 kg. of the berries of Rhamnus infectorius. Assuming that the free fisetin found in poison ivy leaves had its origin in the decomposition of a fisetin-glucoside by natural processes, it was reasonable to suppose that the sugar would also be found in the free state although according to Roscoe and Scharlemmer:² "Isodulcitol does not occur in the free state in nature, but is found as a peculiar ethereal salt belonging to the class of glucosides. On boiling with dilute sulphuric acid, this splits up into isodulcitol and other bodies" The more recent works on the sugars³ and on plant chemistry mention the occurrence of rhamnose only

1. Ann. de Chim. et de Phys., 6th Series, XXII, 76 (1891).
2. Treatise on Chem. Vol III, Pt. III, 492.
3. Les Sucres; Chem. der Zuck.; Biochem. der Pflanz.

in the glucoside form, with one possible exception. The exception referred to is the occurrence of a free sugar, supposed to be rhamnose, in a certain palm-wine¹. Czapek says:² "The well known methyl pentoses do not occur in the free state in plant organisms so far as we know."

Since rhamnose forms a lead compound, the sugar, if present, should be found in the first lead precipitate, A, and also in filtrate A in case it is not completely precipitated in the presence of acetic acid and alcohol.

The filtrate A (about two liters) was examined first. It had a light yellow color, contained an excess of lead acetate, and was acid from the acetic acid liberated in the precipitation of the lead compound A³. This filtrate was evaporated by dryness under diminished pressure to remove alcohol, water, and acetic acid. The clear distillate had a peculiar odor suggesting both tea and amy^ml formate. It was saved for examination and was found to be not poisonous. The residue in the dish after evaporation was a tough reddish brown, gummy mass which could be drawn out into fine threads.

1. Chem. Zeit. 23, Rep. 177.
2. Loc. cit. 1, 209.
3. On standing several weeks, a small quantity of tar separated out on the walls of the vessel, also a brown precipitate which was filtered off, suspended in water, and hydrogen sulphide was being passed in when an accident occurred and it was lost.

It had a pleasant sweet odor. It was extracted several times with hot water each portion being filtered. A yellow brown powder remained undissolved and was saved. The combined filtrates deposited more of the yellow solid on standing. This powder will be referred to later as "P". The filtered liquid was freed from lead by hydrogen sulphide. The solution then had a lemon yellow color, a sweet odor and was acid from acetic acid. On concentrating the solution by evaporation and making a small portion of it alkaline with sodium hydroxide, the yellow color came out very intense¹. The alkaline solution reduced Fehling solution and ammoniacal silver nitrate indicating the presence of a sugar. Another portion of the solution gave a slight precipitate with phenyl hydrazine in the cold. The remainder of the solution was evaporated to dryness, extracted with water, filtered, and again evaporated. A dark sticky syrup was left which was only partly soluble in water. This was treated with water, the filtrate filtered, and evaporated, the water being replaced from time to time to remove acetic acid. Finally the liquid gave the following tests for rhamnose, besides those already mentioned:

- (1) With a-naphthol² and sulphuric acid, a purple violet color.
-

1. "By warming with alkalis or barium hydroxide, rhamnose is colored yellow"-
Von Lippmann, Chem. der Zucker. I, 177.
2. Chem. der Zuek.

- (2) With thymol¹ and sulphuric acid, a red color.
- (3) With resorcinol² and sulphuric acid, red color.
- (4) With orcinol³ and hydrochloric acid, red color.
- (5) With ammonium picrate and sodium picrate, yellowish red color.
- (6) With phloroglucinol and hydrochloric acid, red color.
- (7) It decolorized an alkaline solution of potassium fericyanide.
- (8) It gave a white precipitate with lead acetate.

The filtrate B (p. 27 of this manuscript) from which gallic acid was precipitated by sulphuric acid in four fractions was saved to examine for sugar. To remove gallic acid completely, and other vegetable matter, it was shaken out several times with ether, and was kept at a low temperature with salt and ice for a long time. It was left standing for several weeks during which time more brown matter separated out and was filtered off. The filtrate was evaporated to a small bulk, cooled and filtered from crystals of potassium sulphate. The filtrate was evaporated to dryness, the residue taken up in water and filtered through boneblack. Addition of alcohol caused complete precipitation of potassium sulphate. The solution then gave the above mentioned characteristic ^{taste} for rhamnose.

-
1. Chem. der Zuck.
 2. Rayman, Sur L'Isodulcite, Bull. Soc. Chim. 47, 668 (1887).
 3. Acides Gummiques.

All attempts to get the osazone of the sugar by the method of Fischer¹ failed, probably on account of the small quantity of the sugar present. The plant, it will be remembered, was originally extracted with ether in which rhamnose is practically insoluble. The above described tests, however, can leave no doubt as to the identity of the sugar.

Additional evidence that the sugar is rhamnose was obtained² by a method described by Maquenne² as follows:

"The production of methyl furfural in the dehydration of isodulcitol furnishes a very simple means of characterizing this sugar in mixtures which contain it; it is sufficient, for example, to distil 50gm. of quercitron wood with as much sulphuric acid and about 150gm. of water, then to rectify the liquid obtained in order to get several drops of the crude furfural, which on addition of alcohol and concentrated sulphuric acid gives immediately the green coloration characteristic of methyl furfural. This procedure is applicable to extracts as well as to entire plants, and has the advantage that it does not require the separation of isodulcitol, the crystallization of which is often very slow and at times impossible when it is mixed with other very soluble substances."

The experiment was tried with the crude ether extract of the plant according to the directions of Maquenne, and the green color with alcohol and sulphuric acid was obtained from the thicker oily portion of the distillate. This test can³ be made with hydrochloric acid³ as well as with sulphuric.

1. Berichte XX, pp. 1089, 1091, 1188, 2566.
2. Ann. de Chim. et de Phys. (6) XXII, 93 (1891).
3. Biochem. der Pflanz. I, 210.

Therefore the color test was tried with the ester mixture prepared in one of the early experiments by boiling the original plant material with hydrochloric acid and alcohol. Methyl furfural was found here also, this method indeed giving better results than that of Maquenne. § The presence of free ^l rhamnose has thus been shown in the original material, in the first precipitate by lead acetate, and in the filtrate from this precipitate. Experiments to be described under "The Poison" showed that the ether extract from the Soxhlet apparatus contained a substance which yielded rhamnose when hydrolyzed by dilute sulphuric acid.

The presence of free gallic acid, fisetin, and rhamnose in the plant can be readily explained by a series of assumptions for which there is a considerable amount of experimental evidence. There is reason to believe that tannin-like bodies are formed at the expense of chlorophyll¹, that complex tannin bodies can be broken down by acetic acid (also found in Rhus toxicodendron) into a tannic acid and a glucoside (for example, the "fustin-tannide" mentioned above yields tannic acid and fisetin-glucoside); and finally that the glucoside can be hydrolyzed by acids or enzymes giving, in the sumach plants, fisetin and rhamnose.

Nitrogenous ferments which can effect the hydrolysis of glucosides and give rise to sugars are frequently found in

1. Comptes rendus CXV, 892.

plants, for example, emulsin in almonds, myrosin in mustard, and erythrozym in madder. Acree and Hinkins¹ found that diastase, pancreatin, and a number of other enzymes hydrolyze with the formation of glucose and acetic acid. Stevens² obtained a nitrogenous oxidizing enzyme from Rhus vernicifera. The close relationship between the poisonous species of Rhus would lead us to suppose that the same soluble ferment exists in poison ivy, though it was not detected in the original material used in these experiments, probably because the plant was extracted with ether in which the enzyme is insoluble. The existence of such a soluble ferment would explain the presence of free sugar and free fisetin.

EVIDENCE OF THE PRESENCE OF A FATTY ACID IN FILTRATE A.
-----OCCO-----

The brown substance P, obtained from filtrate A by evaporation and extracting the residue with hot water, was suspended in warm water and dilute sulphuric was added. A white precipitate was formed and a strong fatty acid odor was developed. After the mixture had been heated for some hours on the water bath a small portion was made alkaline and it reduced Fehling

-
1. Amer. Chem. Jour. 28, 370.
 2. Amer. Jour. Pharm. 77, 255 (June 1905); 78, 53 (Feb. 1906).

solution. The main solution was filtered and the precipitate supposed to be a fatty acid was saved. The filtrate was neutralized with barium carbonate, filtered, evaporated, freed from caramel, and the solution then gave the tests mentioned above for rhamnose.

A portion of the precipitate supposed to be a fatty acid was ignited in a porcelain spoon. It fused, carbonized, and burned. The remainder was heated with alcoholis potash and reprecipitated with hydrochloric acid. The precipitate was washed and heated with alcohol. Part of it dissolved. The insoluble part was found to be a lead compound. On boiling it with hydrochloric acid and cooling, lead chloride crystallized out. This was confirmed by dissolving the lead chloride in hot water and precipitating as lead sulphide. These experiments were not carried farther on account of the small quantity of material, but they show that the gummy substance obtained from filtrate A contained rhamnose (either as a lead compound of free sugar or as a lead compound of a rhamnoside), and also, most probably, the lead compound of an organic acid.¹

THE FRAGRANT DISTILLATE.

Several times in the course of this work, extracts of the original plant material in alcohol and in water were distilled under diminished pressure for the purpose of concentrat-

1. A wax obtained from Rhus succedanea was shown by Stahmer to contain palmitic acid and glycerol in the form of glyceryl palmitate. Annalen 43, 343, (1842).

ing the solutions. The distillate, in every case, had an ethereal odor suggesting amyl formate in very dilute solution, but was more fragrant. The distillate from a water extract was examined. It was a clear liquid, a little darker than pure water, was not poisonous, was neutral to litmus paper, gave no color with ferric chloride, reduced ammoniacal silver nitrate but not Fehling solution, and gave a faint red color with dilute ammonium hydroxide and with sodium carbonate.

A small quantity of a finely divided black precipitate separated out from the water distillate on standing.

The substance with the fragrant odor was extracted by shaking the distillate with ether and letting the ether evaporate spontaneously. A very small quantity of a yellow solid was deposited on the sides of the dish. This substance had a strong and persistent odor, so sweet as to be almost nauseating. Not enough was obtained for examination or analysis. This fragrant residue was difficultly soluble in water and the solution reduced silver nitrate in ammonia. A steam distillate of the original plant material had the same fragrant odor as the distillate from a water extract.

THE POISON.

288 gm. of the original poisonous material were extracted with 50% alcohol, and this alcoholic solution was precipitated with lead acetate in the manner already described (p. 22)

of this manuscript). The lead precipitate so obtained was extracted with ether in Soxhlet extractors and after the extraction was found by test to be free from poison. Therefore the poison, if precipitated by the lead acetate, must have been extracted by the ether. This ether solution had a dark green color, and was acid from acetic acid brought down in the lead precipitate. The ether was evaporated in a vacuum desiccator without heat and there remained a small quantity of an acid mixture of water and a soft tar; the watery part was colored green showing that the tar was soluble to some extent in dilute acetic acid. The mixture had the peculiar odor of the original material. A small drop of the green watery part was applied to the wrist, allowed to remain a few minutes and was then removed by absorbent paper, but the spot was not washed. Itching and reddening of the skin commenced within twenty-four hours. At the end of forty-eight hours, there was a well developed case of poisoning. How this was cured will be described in another place.

A small portion of the poisonous mixture was dissolved in alcohol, and this solution was divided into three parts. The first part was treated with ferric chloride but it gave no color reaction. Another portion of the alcoholic solution was diluted with water. It became turbid. The third portion gave a dirty-green precipitate with lead acetate, which seemed to come down more readily when the solution was diluted with

water. The main portion of the poisonous mixture was then dissolved in 95% alcohol and lead acetate in 50% alcohol was added. The precipitate was filtered, washed, decomposed by hydrogen sulphide in a mixture of water and ether. The ether solution was filtered and evaporated. The residue was a tar which on standing in a desiccator for some time became dry enough to break into sticky lumps. An alcoholic solution of this substance gave a dark color with ferric chloride and a light colored precipitate with lead acetate.

To get more of the poisonous tar for study, 233 gm. of original material were extracted with 95% alcohol. Strong alcohol was used in order to dissolve as much of the tar as possible. The solution had a dark greenish color but was somewhat yellow in thin layers. The undissolved tar was filtered off and extracted twice again in the same way. The tar left after the third extraction was only slightly soluble in alcohol, and its solution was not poisonous. The three filtrates from these three extractions were precipitated separately by lead acetate in 50% alcohol. The first precipitate was larger, darker in color, and carried down more tarry matter. The second was not as large as the first and was light green. The third was quite small, was black, and was not a lead compound at all but some of the tar which separated out on diluting the strong alcohol with the weaker

grade containing lead acetate. It was soluble in ether and less soluble in alcohol. The alcoholic solution of this third lot gave no precipitate with hydrogen sulphide. The first and second lead precipitates were filtered by suction and washed with water. They were kept a day or two in a desiccator over sulphuric acid but did not become completely dry. The weight of these two moist precipitates together was 172gm. They were combined and extracted with ether in Soxhlet extractors which were kept in operation during work hours for three days.

In the meantime the alcoholic filtrates from these lead precipitates were combined and concentrated on the water-bath by distilling off two liters of alcohol. The alcohol obtained had the peculiar odor of the original material, but was not poisonous.

After a long extraction of the lead precipitate in the Soxhlet extractors, the green ether solutions were combined and washed by shaking them with water to remove lead acetate and acetic acid in case any should have been held in the lead precipitate. The ether was distilled off at a low temperature and there remained a soft tar, a portion of which was not completely soluble in 95% alcohol. The alcoholic solution had a greenish yellow color and was poisonous. The tar was also partly soluble in acetic acid, and this solution was found to

contain lead. Thinking that the lead acetate had not been completely washed out, the main part of the tar was dissolved in ether and shaken with water. The wash water continued to give a test for lead as long as the washing was continued. This indicated probably the hydrolysis of an unstable lead compound. Hydrogen sulphide was passed into the ether solution mixed with water to remove the lead. Lead sulphide was filtered off, and the ether was evaporated. A small portion of the tar residue in alcoholic solution gave a color reaction with ferric chloride. As this may have been due to traces of lead gallate dissolved in the extraction with ether and afterwards decomposed by hydrogen sulphide, the main portion of the tar was redissolved in ether and shaken with water until it no longer reacted with ferric chloride. The ether was then evaporated and a soft, black, poisonous tar or gum of uniform consistency was left which was shown by tests to be free from gallic acid and lead. These experiments showed that some of the poison was precipitated as a lead compound soluble in ether and some was brought down mechanically in the free state. To see if the extraction with ether in the Soxhlet apparatus was complete, the residue from in the thimbles was decomposed by hydrogen sulphide and shaken with ether. The dark colored ether solution was freed from gallic acid by shaking with water and dilute sodium carbonate solution, and was evaporated. A small

quantity of tar was obtained which was added to the main portion.

A solution of the poisonous tar in 95% alcohol did not reduce Fehling solution and did not give a precipitate with lead acetate except the separation of a small quantity of tar, which was not a lead compound. The lead compound of the poison was apparently soluble in 95% alcohol as well as in ether, for it would not precipitate in this medium, although it was found in the original precipitate by lead acetate. The alcoholic solution of the tar became turbid on diluting with water.

In order to see if the poison is volatile with vapor of acetic acid, since this acid is found in the plant and it is thought by some that the poison is volatile, a portion of the tar was distilled under diminished pressure with acetic acid. It was soluble to some extent in the acid. The temperature did not go higher than 55°C. during the distillation. A tube containing cotton wet with sweet oil was placed between the receiver and the water suction so that the uncondensed vapors would have to pass through the cotton. This cotton was rubbed on the skin and was not poisonous. The yellow distillate collected in the receiver was also tested and was not poisonous.

HYDROLYSIS OF THE TAR. About 5gm. of the tar free from gallic acid and sugar was dissolved in alcohol and dilute (2%) sulphuric acid was added. Some of the tar separated out on diluting the alcohol with the acid. The mixture was heated on a water-bath during work hours for four days. A purple

and green fluorescent solution was formed, though much tar was left apparently unchanged. The alcohol was evaporated off and the solution was filtered from tar. The fluorescent filtrate was shaken with ether, by which the green substance was removed, leaving the solution purple. The ether left, on evaporation, a small quantity of a green substance having a pleasant ester odor. It was not examined. ^{further} A portion of the purple solution was exactly neutralized with sodium carbonate. This solution gave a blue-black color with ferric chloride which became red on addition of another drop of sodium carbonate, indicating gallic acid. It also reduced Fehling solution.

Another portion of the purple solution was made alkaline with sodium carbonate. A reddish-brown flocculent precipitate was formed and was filtered off. The filtrate did not give any color with ferric chloride, but it reduced Fehling solution. It also gave the test for rhamnose with d-naphthol.

The main portion of the purple solution was made alkaline with sodium carbonate; the precipitate was filtered off and dissolved in acetic acid. This solution was yellow and gave a reaction with ferric chloride similar to that of gallic acid. The filtrate from the precipitate by sodium carbonate was concentrated by evaporation until sodium sulphate began to crystallize out. Alcohol was added to precipitate the sodium sulphate completely, the mixture was heated and filtered.

The alcoholic filtrate was concentrated to a syrup which reduced Fehling solution and gave the characteristic tests for rhamnose already described. By this hydrolysis, the tar was split up into rhamnose and some form of gallic acid which could be precipitated by sodium carbonate. This compound, whose acetic acid solution was yellow, probably contained fisetin also. The reason for this last statement will appear from the following experiment.

Decomposition of the Tar with Acetic Acid.

A portion of the poisonous tar was heated in an open dish with strong acetic acid. The tar seemed to be decomposed to some extent giving a yellow substance. Acetic acid was added from time to time as it evaporated. After several evaporations, water was added, the mixture was heated to boiling and filtered. This filtrate No. 1 will be mentioned later. The residue in the dish consisted of undecomposed tar and an olive-green flaky substance. This substance was heated with a fresh portion of glacial acetic acid. Water was added, and the mixture was boiled and filtered. The filtrate had a deep yellow color suggesting fisetin. It was shaken out with ethyl acetate which became colored yellow. A portion of the ethyl acetate solution gave an orange red precipitate with lead acetate showing the presence of fisetin. The ethyl acetate was removed from the remainder of the solution by evaporation and the yellow residue was taken up in

alcohol. This alcoholic solution gave the characteristic reactions for fisetin with stannous chloride, with potassium hydroxide, with ferric chloride and with Fehling solution.

Filtrate No. 1 obtained by heating the poisonous tar with acetic acid and hot water as described above was investigated as follows: A portion of it gave a reddish colored precipitate with sodium carbonate as in the case when the tar was hydrolyzed with sulphuric acid. The remainder was nearly neutralized with sodium carbonate and lead acetate was added in excess to remove gallic acid. The excess of lead was removed by sulphuric acid, and the sulphuric acid ^{was removed} by barium carbonate. The solution on evaporation reduced Fehling solution to some extent but a white precipitate was also formed.

In this experiment, gallic acid and fisetin and probably sugar were formed by decomposition of the poisonous gum with acetic acid, the acid found in the plant by Pfaff. The presence of free gallic acid, fisetin and rhamnose in the plant can be explained by the natural hydrolysis of a complex gum or tar or a constituent thereof.

The poisonous property is lost in the general rearrangement which takes place during hydrolysis.

The poisonous tar was not hydrolyzed by boiling with a dilute solution of sodium carbonate.

It was found, as has been stated elsewhere, that the lead compound of the poison could not be precipitated in 95% alcohol.

Further experiments, however, showed that on extracting the poisonous gum with 50% alcohol, a portion of it dissolved, and this solution gave a precipitate with lead acetate which was a true lead compound. The remainder of the purified tar (about 10gm.) was treated with 50% alcohol and filtered. Very little dissolved in alcohol of this strength, but on addition of lead acetate in 50% alcohol to the solution, a light colored precipitate was formed, which became dark on standing. It was filtered off, washed free from lead acetate, decomposed by hydrogen sulphide, and shaken out with ether. The ether left, on evaporation, a yellow resinous substance having a faint odor like garlic. By drying in a desiccator, a small quantity of a solid yellow resin was obtained which was completely soluble in alcohol. A very small drop of this solution applied to the skin on the end of a glass rod which had been drawn out to a point caused an eruption in about thirty-six hours. Following the nomenclature used by Maisch and Pfaff, this substance will be designated as Toxicodendrin, the ending "in" indicating its glucoside nature.

The filtrate from the lead precipitate just described was freed from the excess of lead acetate by hydrogen sulphide, was tested for poison, and was found to be poisonous, showing that the precipitation by lead acetate was not complete even in 50% alcohol. On spontaneous evaporation of the solution,

a yellow, sweet smelling resin was left.

A portion of the alcohol solution of the toxicodendrin gave a dark coloration with ferric chloride, did not reduce Fehling solution and was slightly acid to litmus.

To see whether the toxicodendrin could be hydrolyzed, the remainder was dissolved in alcohol and dilute sulphuric acid was added. A fine, white precipitate was formed at once which rose to the surface on standing as a light flocculent substance. The mixture was heated for several days on a water-bath, filtered from unhydrolyzed resin and the filtrate was neutralized and concentrated in the way already described. The solution obtained reduced Fehling solution. Not enough was obtained for further sugar tests, but all the hydrolysis experiments point to the conclusion that the poisonous substance is a rhamnoside, and is the source of the sugar in the plant.

The reaction with ferric chloride observed whenever a lead compound of the poison is decomposed by hydrogen sulphide may be explained by the formation of traces of gallic acid or fisetin through the action of the weak acids present.

The supply of purified poisonous tar having been exhausted in the preceding experiments, further study of the active principle is postponed until more can be prepared. It is highly desirable to investigate the white precipitate formed by addition of sulphuric acid to an alcoholic solution of the

toxicodendrin.

OXIDATION OF THE PURIFIED TAR WITH NITRIC ACID.

When the purified poisonous material (p. _____) was extracted with 50% alcohol, only a small quantity was dissolved as was stated above. The insoluble residue was treated with fuming nitric acid. Violent reaction took place at once with copious evolution of red fumes and heat. When the reaction was over, a sticky red gummy mass was left which was slightly soluble in warm alcohol. The water extract was yellow, and the alcoholic solution was red. That the water extract contained picric acid was shown by the following experiments:

- (1) A portion was gently warmed with a few drops of a strong solution of potassium cyanide and two drops of sodium hydroxide. The red color of potassium isopurpurate was formed.
- (2) A portion of the water solution was heated with glucose and a few drops of sodium hydroxide. The deep red color of picraminic acid was produced.
- (3) A few drops of an ammoniacal solution of copper sulphate was added to the water extract. A yellow-green precipitate was formed.
- (4) The water extract dyed silk, but did not dye cotton cloth.

DISTILLATION OF THE TAR WITH SODA LIME.

About 25gm. of the tar left after extracting the original material with hot water was dissolved in ether and poured into a glass retort containing soda-lime. The ether was ~~then~~ distilled out leaving the tar intimately mixed with the soda-lime. The retort was then gradually heated. Vapors and liquid were given off, both of which turned red litmus blue and had a strong odor like tobacco smoke. No odor of ammonia was detected¹. At the high temperature of the triple burner, a semi-solid, red, greasy substance collected in and closed the condenser tube. This substance had the same powerful odor as the liquid portion of the distillate. The clear, watery portion of the distillate was separated from the thicker parts, and was found to contain pyrrol and pyridine derivatives by the following characteristic tests:

- (1) Wood moistened by hydrochloric acid was turned red by it.
- (2) Colorless fumes were formed when brought near hydrochloric acid; ~~when~~ mixed with hydrochloric acid, a red insoluble substance was formed.
- (3) It precipitated the hydroxides of iron salts, gave a light blue precipitate with copper sulphate, and a white precipitate with mercuric chloride.

The greasy, semi-solid mass was extracted with 10% hydrochloric acid and filtered. On addition of a solution of

1. See Amer. Jour. Pharm. 77, 256.

mercuric chloride to the red filtrate, a brown flocculent precipitate was formed. It was filtered off and distilled with caustic soda but the distillate did not contain pyridine.

POTASSIUM PERMANGANATE AS A REMEDY FOR RHUS POISONING¹.

In the early stages of this work some experiments were made to see if potassium permanganate could be used to purify the lead precipitate by oxidizing the tar brought down in precipitation. It was found that the permanganate attacked the lead precipitate as well as the other organic matter in the vessel. This fact and the well known value of permanganate in treating skin diseases, its use as an antidote for some kinds of alkaloid poisoning², as an antidote given to cattle poisoned by plants, and as an antidote for snake bites³, suggested its use as a remedy for Rhus poisoning⁴. Maisch⁵ mentioned that he had used it with success, but it never came into general use, probably on account of its staining the skin and clothing. In carrying out this work abundant opportunities for testing its value as a remedy for the dermatitis caused

-
1. This section is added in the hope that it may be of use to others who are subject to this form of poisoning.
 2. Moor, N. Y. Med. Rec. 45 (1894), 200.
 3. Bull. No. 26, U. S. Dpt. Agr., Div. of Bot. 47.
 4. Lacerda, Comptes rendus 93 (1881), 466-469.
 5. Amer. Jour. Med. Sci. 52, (1866), 285.

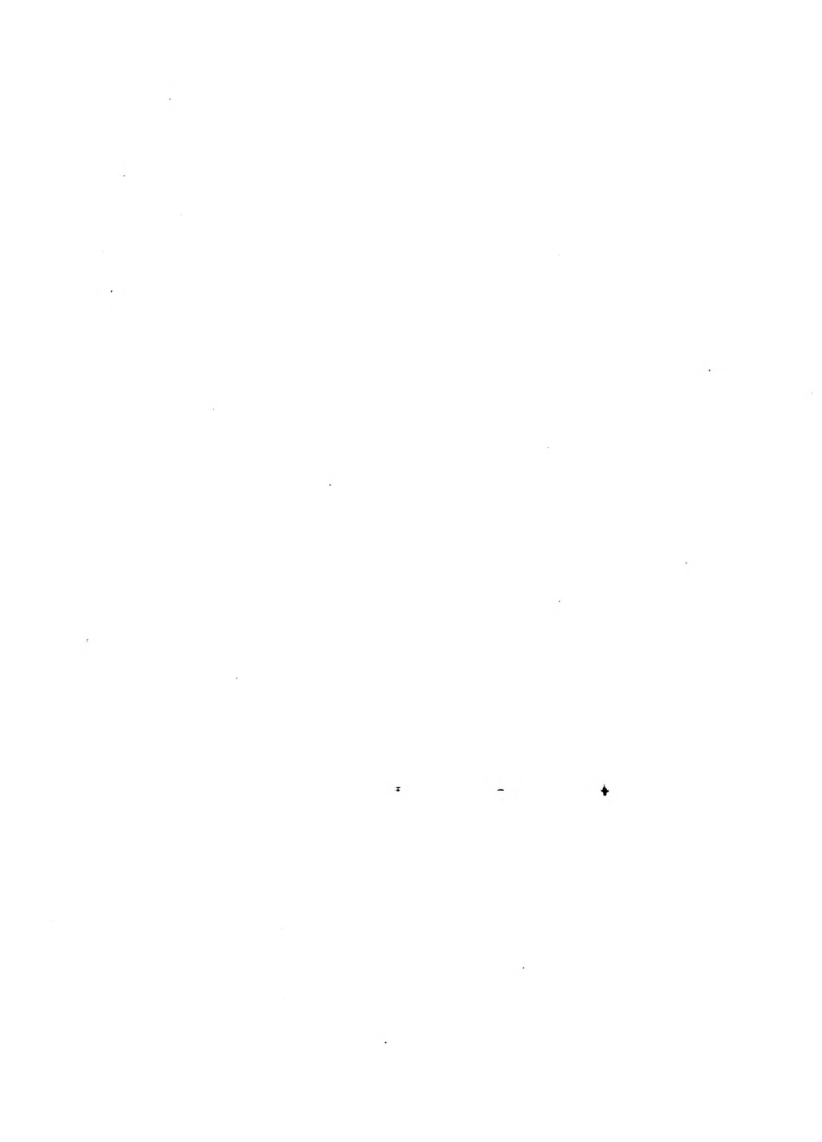
by poison ivy were afforded by many cases of accident^{al} and intentional poisoning. The best example of the latter was obtained with the ether solution from the extraction of the lead precipitate in the Soxhlet apparatus (page 41 of this manuscript). After removing the ether, a small drop of the residue was applied to the wrist as described. An itching red spot about the size of a dime was noticed in thirty-six hours which steadily increased in size. Nearly two days after the application of the poison, a dilute solution of potassium permanganate containing a little caustic potash was rubbed into the spot until the pimples were destroyed. A little black spot was left wherever there had been a pimple showing that the permanganate had been reduced to oxide in the skin. The place was washed and nothing more was thought of it until the morning following when it was noticed that the wrist had commenced to swell during the night, and the characteristic watery secretion was running from the poisoned spot. More permanganate solution was applied without potash and the wrist was bandaged thinking that this would prevent the spreading of the eruption, but it really facilitated spreading by becoming saturated with the poisonous fluid and keeping it in contact with a larger surface of skin. In the meantime the swelling and inflammation had extended nearly to the elbow. The arm now had the appearance of having been bitten by a snake.

To reduce the swelling it was immersed in hot water. This seemed to bring out the eruption very quickly and the blisters were treated with permanganate as fast as they appeared. The swelling was reduced but returned during the night. On the evening following, the forearm was immersed in a bowl of hot permanganate solution containing a little caustic potash. The solution was kept as hot as could be borne for about half an hour. After this bath, the poison was completely oxidized, for the swelling was reduced and did not return, nor was there any fresh eruption. What appeared to be a severe case of poisoning was thus cured very quickly. The use of hot water not only reduces the swelling but also helps to destroy the poison. The action of permanganate is also more rapid at high temperatures.

The oxidizing power of permanganate, as is well known, is greater in acid solution than in alkaline, five atoms of oxygen being available in the former and three in the latter according to these equations:



Permanganate was used as a remedy in some cases mixed with dilute sulphuric acid, and in others, with zinc sulphate; also with lime water. It was found to be satisfactory whether used alone or with any of the substances mentioned, provided it was well rubbed into the skin. The concentration of the



solution used was varied according to the location and condition of the eruption. Where the skin was thin or already broken, dilute solutions (one per cent) were used. In one case, the eruption appeared in the palm of the hand where the skin was so thick that it was necessary to open it before the remedies could reach the poison. The difficulty of getting the remedy in contact with the poison in the skin is the reason why the eruption is hard to cure.

The remedy most commonly used for this eruption is an alcoholic solution of lead acetate. This remedy is unsatisfactory for the reason that its action consists in depositing an unstable lead compound of the poison in the skin where the conditions of moisture and temperature are favorable for its decomposition, liberating the poison with all its irritant properties. Moreover, alcoholic preparations should not be used because the alcohol dissolves the poison and on evaporation, leaves it spread over a larger surface like a varnish. Potassium permanganate, however, oxidizes the poison completely. The only objection to the use of permanganate of which the writer is aware is that it stains the skin. The stain can be removed by vigorous scrubbing with soap, or it will wear off gradually in a few days. It can be removed at once by certain acids, but these should not be used by persons not familiar with their action.

With the knowledge of the facts mentioned, many solutions

were tested for poison by applying them to the skin, and when an eruption appeared, it was cured quickly and permanently by rubbing in a permanganate solution, usually mixed with dilute sulphuric acid.

SUMMARY.

Leaves and flowers of the poison ivy plant were extracted with ether and the ether was removed by evaporation. In the residue, the following substances were found and studied: gallic acid, fisetin, the sugar rhamnose, and a poisonous tar, gum or wax.

The lead compound of the poison was soluble in ether; this fact gave a means of separating the poisonous substance from the non-poisonous matter in one operation.

The poison was not volatile with vapor of acetic acid, or with vapor of alcohol.

The poisonous tar or wax was decomposed by acids and yielded gallic acid, fisetin, and rhamnose, showing the probable source of these compounds in the plants, and indicating that the poison is a complex substance of a glucoside nature.

It was found that a portion of the poisonous substance could be precipitated by lead acetate from a solution of the tar in 50% alcohol.

All cases of poisoning developed on the writer were easily cured with potassium permanganate.

The following method is suggested for obtaining the poisonous substance from the plant: Extract the plant with alcohol, filter, and precipitate at once with lead acetate. Wash the precipitate, dry, and extract with ether in Soxhlet extractors (loosely filled). Combine the ether extracts, mix with water, and pass in hydrogen sulphide. Separate the water and the ether solution, and filter the latter. Wash the ether solution thoroughly by shaking with water, and then evaporate at a low temperature.

BIOGRAPHY.

W. A. Syme was born in Raleigh, N. C., on July 11, 1879. He was prepared for college at the Raleigh Male Academy, entered the North Carolina College of Agriculture and Mechanic Arts in 1896, and was graduated in 1899 with the degree B. S. He was an Instructor in Chemistry at the same College from January 1st, 1900, until June, 1903, when he received the degree M. S. for graduate work. In October following, he entered Johns Hopkins University as a graduate student in Chemistry, and was awarded one of the North Carolina Scholarships. His minor subjects are Physical Chemistry and Biology.

Syme, William Anderson
SOME COMMITMENTS OF THE
POISON IVY PLANT
Pub. Diss. 1906
127,823

ISSUED TO

Syme, William Anderson
SOME COMMITMENTS OF THE
Pub. Diss. 1 06
127,823

POISON IVY PL

