(10) M. M. Knude and T. Proud, "The Ineffectiveness of Vaginal Smears in Predicting the Oestrus Cycle in the Rabbit," *Am. J. Physiol.*, 88 (1929), 446.

(11) A. Prybill and K. Maurer, "Versuche über Werthestimmung und Altern von Mulerkornzubereitungen," *Arch. der. Pharm.*, 266 (1928), 464.

(12) F. Wokes and G. K. Elphick, "The Preparation of Liquid Extract of Ergot," *Quart. J. Pharm. Pharmacol.*, 2 (1929), 539; 3 (1930), 59.

(13) F. Wokes, "The Stability of Extracts of Ergot," *Ibid.*, 2 (1929), 386.

(14) F. Wokes and G. K. Elphick, "The Extraction of Ergot by the Methods of D. A. B. VI and U. S. P. X," *Ibid.*, 3 (1930), 599.

(15) M. I. Smith, "A Quantitative Colorimetric Reaction for the Ergot Alkaloids and Its Application in the Chemical Standardization of Ergot Preparations," *Public Health Report*, 45 (1930), 1466; M. I. Smith and E. F. Stohlman, "Standardization of Ergot, Comparative Study of the Chemical and Biological Methods of Ergot Assay," *J. Pharmacol.*, 40 (1930), 77.

(16) K. Spiro and A. Stoll, "Active Substances in Ergot," *Schweiz. med. Wochschr.*, 51 (1921), 529.

(17) A. W. Forst and H. Weese, "Über die uleruswirksamen Substanzen im Mutterkorn," *Arch. exptl. Path. Pharmakol.*, 117 (1926), 232.

(18) A. Stoll and E. Rothlin, "Ueber Mutterkornpräparate," *Schweiz. med. Wochschr.*, 57 (1927), 106.

A PHARMACOLOGICAL NOTE ON CIMICIFUGA.

BY DAVID I. MACHT AND HELEN M. COOK.

INTRODUCTORY.

Cimicifuga, popularly known as Black Cohosh, Black Snakeroot, Rattleroot, Rattlesnake Root, Bugbane, Bugwort, Squaw Root and Macrotys, is the dried rhizome and root of *Cimicifuga racemosa*, of the family of the *Ranunculaceæ*. This drug has enjoyed considerable vogue in the United States, and it was official in the British Pharmacopœia but dismissed from it in 1914. A number of its fluid preparations are still recognized in America and the resin obtained from cimicifuga, known as cimicifugin, or macrotin, can also be found on the market. In addition to the resin, which can be obtained in impure form by precipitating a saturated tincture of the root with water, cimicifuga contains a volatile oil and a bitter neutral substance. This drug was introduced into medical practice a hundred years ago by Young (1). In addition to Young's paper on cimicifuga, there is another early reference to this drug by N. S. Davies (2).

Cimicifuga preparations have been recommended for a number of different conditions. Sir Lauder Brunton recommends it as a stomachic, cardiac tonic and expectorant (3). The eighteenth edition of Hare's "Practical Therapeutics," published in 1922, cites as indications for its use the following clinical conditions: rheumatism, chronic bronchitis, chorea, amenorrhea and subinvolution and tenderness of the womb (4).

Even the earlier editions of Osler's "Principles and Practice of Medicine" recommended the use of cimicifuga in St. Vitus' dance, or chorea (5). In spite of these various indications to be found in books on therapeutics, there is no scientific experimental work on record concerning the pharmacology of Snakeroot. In connection with a study of various drugs, old and new, purported to exert an effect on uterine muscle, the present author has made an experimental investigation of the fluidextract of cimicifuga and of cimicifugin; and the results obtained are briefly recorded in this place.

Inasmuch as cimicifuga has been principally recommended as a stomachic, a specific for chorea and as an "uterine tonic," the author in performing his experiments was chiefly concerned with: 1, the effects of cimicifuga on cerebral convulsions and on neuromuscular coördination; 2, the effects of the drug on the intestinal movements; and 3, its action on the uterus.

EFFECTS ON THE BRAIN AND NEUROMUSCULAR APPARATUS.

Since cimicifuga has been especially recommended in chorea, or St. Vitus' dance, two series of experiments were undertaken to ascertain its possible effects on the brain and on neuromuscular coördination, on the one hand, and convulsions, on the other. In order to study the effects of cimicifuga on the brain, psychopharmacological experiments were made with white rats. The method used has been described by the writer in various papers. Albino rats are trained to run in the circular maze and reach its center in the shortest time possible without an error. White rats of good pedigree can be trained to solve the maze problem in two weeks and, when they have done so, are ready for the study of pharmacological agents. The running time and behavior of an individual rat are determined before administration of the drug. The animal is then given the drug to be studied, and at intervals of fifteen minutes thereafter is tested to ascertain its psychological activity, muscular coördination, etc. Since fluidextract of cimicifuga could not be given by injection, it was necessary to administer the drug by a specially constructed "stomach tube" devised by the author. With such an instrument in expert hands, the drug can be introduced with impunity into the stomach of a rat without the use of an anæsthetic. With a fine tuberculin syringe the solution is then injected into the stomach. In the present research, trained rats were selected and given 0.1 cc. of the fluidextract of cimicifuga diluted with 0.9 cc. of water. Control experiments were made with similar quantities of 95 per cent ethyl alcohol diluted with ten times its volume of water. The results obtained were very interesting. When rats weighing about 300 Gm. were given 0.1 cc. of 95 per cent alcohol in 0.9 cc. of water by stomach tube, no effect whatever was noted on their psychological activity. Neither the brain nor the neuromuscular apparatus was depressed after such doses of alcohol. When 0.2 cc. of a 95 per cent solution of ethyl alcohol was so administered, a mildly depressant effect was noted. When 0.1 cc. of fluidextract of cimicifuga was diluted with water and introduced in the rat's stomach in the same way, no sign of either depression or excitement was observed. However, a distinctly depressant effect, which lasted about two hours, was produced by doses of 0.2 cc. of the fluidextract. The following protocols illustrate the psychopharmacological experiments described above.

PSYCHOPHARMACOLOGICAL EXPERIMENTS (FEBRUARY 25, 1932).

Rat *No.* 1—weight, 250 grams: normal running time, 5.3 seconds; no errors.
Given 0.1 cc. of fluidextract of cimicifuga by stomach tube.
Running time 1/2 hour later, 5.6 seconds; no errors.
Running time 1 1/2 hours later, 6.0 seconds; no errors.

Rat *No.* 2—weight, 250 grams: normal running time, 15.3 seconds; no errors.
Given 0.2 cc. of fluidextract of cimicifuga by stomach tube.
Running time 1/2 hour later, 26.6 seconds; one error.
Running time 1 1/2 hours later, 26 seconds; no errors.

Albino rats were also used to ascertain whether or not cimicifuga preparations have an inhibitory effect on convulsions. It is well known that camphor, when injected in sufficient quantities in various animals, particularly rats and guinea pigs, produces tonic and clonic convulsions of epileptiform character. Advantage was taken of this convulsant action of camphor in the present problem. Adult rats weighing 300 Gm. or more were given as much as 0.5 cc. of fluidextract of cimicifuga diluted with water by stomach tube. From fifteen to thirty minutes later such rats were injected with camphorated oil in doses of 0.2 cc. (40 mg.) per hundred Gm. weight. As control animals, other rats to which cimicifuga had not been administered were injected with equivalent doses of camphorated oil. It was found that the onset of convulsions, their violence and duration, in no way differed in the two sets of rats. The same convulsions took place in the animals treated with cimicifuga as occurred in the normal rats injected with camphor. In other words, even large doses of cimicifuga extract, 0.5 cc., were not sufficient to inhibit the camphor convulsions, as bromides are known to do, for instance (6). The following protocols will serve as illustrations.

CONVULSANT EXPERIMENTS (FEBRUARY 19, 1932).

Rat *A*—weight, 350 grams.

Given 1.0 cc. (200 mg.) of camphorated oil by intraperitoneal injection.

Four minutes later violent clonic and tonic convulsions commence and continue a long time.

Rat *B*—weight, 380 grams.

Given by stomach tube 0.5 cc. (100 mg.) of fluidextract of cimicifuga diluted with water.

Fifteen minutes later given 1.0 cc. (200 mg.) of camphorated oil by intraperitoneal injection.

Four minutes thereafter violent clonic and tonic convulsions commence and continue a long time.

EFFECT OF CIMICIFUGA ON INTESTINES.

Studies were made of the action of fluidextract of cimicifuga and of cimicifugin on the isolated intestines of rat, guinea pig, rabbit and especially the cat. The method employed was the usual one. Segments of the small intestine, taken either from the ilium or jejunum, were suspended in oxygen and Locke's solution, kept at a constant temperature of 38° C.; and the normal peristaltic contractions were recorded on a slowly moving drum. Various quantities of cimicifuga preparations were then introduced into the chamber of Locke's solution in which the segments were suspended, and the effect on the contractions and the movements of the intestinal muscle was observed. Controls with equivalent doses of alcohol were also made. The results obtained are well illustrated in Figs. 1, 2 and 3. In Fig. 1, a strip of the intestine of the cat was kept alive in oxygenated Locke's solution; and its movements were recorded on a slowly moving kymograph. A suspension of 5 cc. of cimicifugin in physiological saline was introduced into the Locke's solution. It will be noticed that the resin suspension produced no effect whatever on the intestinal contractions. The same preparation was then treated with a 0.5 cc. of fluidextract of cimicifuga introduced directly into the chamber containing 50 cc. of Locke's solution. It will be noted that this produced a primary stimulation of the intestinal contractions. Such a primary stimulation of the intestinal contractions, however, is usually transient. When large doses of the fluidextract are employed, such as are shown in Fig. 2, the intestinal muscle is relaxed and paralyzed. It is

still capable, however, of responding with a contraction to a large dose of pilocarpine. In Fig. 2 is shown the effect of 1.0 cc. of ethyl alcohol introduced into 50 cc. of Locke's solution. It will be seen that no change in the intestinal movements was produced. This was followed by 1.0 cc. of fluidextract of cimicifuga to produce relaxation of the intestinal segment, but not complete paralysis, as shown by its contraction to 4.0 mg. of pilocarpine. Such a depressant effect occurred even after very small doses of cimicifuga. In Fig. 3 is shown the effect commonly produced by fluidextract of cimicifuga on isolated intestines. Here 0.1 cc. of fluidextract produced a distinct relaxation; 0.2 cc. more produced further relaxation and, finally, paralysis and death of the preparation, as indicated by its failure to respond even to 5 mg. of pilocarpine hydrochloride. Such repeated experiments have demonstrated that fluidextract of cimicifuga, studied on the isolated surviving intestines, may

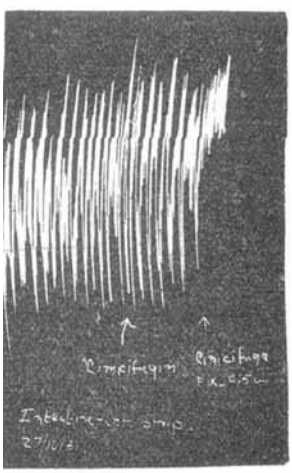

Fig. 1.—Effect of cimicifugin and of fluidextract cimicifugæ on intestine of cat.

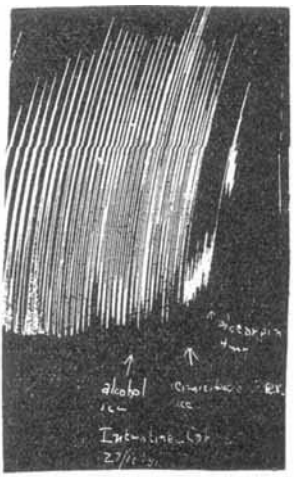

Fig. 2.—Effect of larger doses of fluidextract cimicifugæ on intestine of cat.

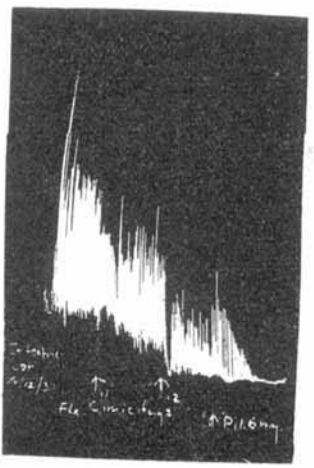

Fig. 3.—Paralyzing effect of repeated small doses of fluidextract cimicifugæ on intestine of cat.

produce a primary transient stimulation of contractions, and this is followed by a relaxation and, finally, paralysis of the intestinal muscle. This is undoubtedly due to the volatile oil contained in the fluidextract, because when similar experiments are made with macrotin or cimicifugin alone no effect of any kind is produced.

EFFECT ON THE UTERUS.

The action of cimicifuga on the uterus is very similar to that on the isolated intestine. Experiments were made on the surviving uteri of the guinea pig and cat. It was found that cimicifugin produced no effect at all on the uterine preparation. Ethyl alcohol alone, given in small quantities, also failed to produce either stimulation or relaxation of the uterine contractions. Fluidextract of cimicifuga, however, produced a primary contraction, followed immediately by relaxation and death of the uterine muscle. Figure 4 illustrates the results obtained. Here the horn of the guinea pig's uterus was first treated with 0.1 cc. of ethyl alcohol introduced in

50 cc. of Locke's solution with no change in the contraction of the uterus. After ten minutes this was followed by 0.1 cc. of fluidextract of cimicifuga, which produced a marked primary contraction of the uterus followed by rapid relaxation and paralysis. After this paralysis the uterine segment failed to respond even to such a powerful stimulant as 1 mg. of ergotoxin.

EFFECT ON CIRCULATION AND RESPIRATION.

In order to complete the pharmacological study of cimicifuga a number of experiments were made to ascertain its effects on the vital organs, the circulation, the respiration and the kidney function. The effect of administration of fluidextract of cimicifuga by mouth to rabbits on the kidney function was negative. Even considerable doses failed to impair the kidney function of rabbits as revealed

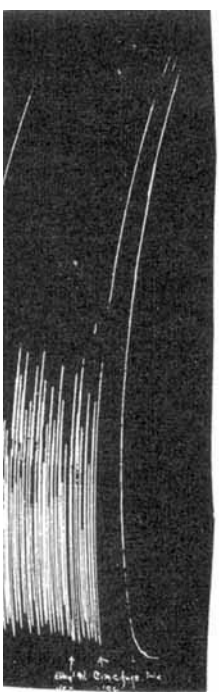

Fig. 4.—Effect of fluidextract cimicifugæ on uterus of guinea pig.

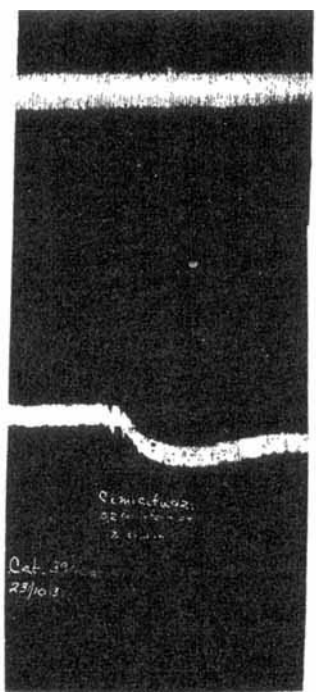

Fig. 5.—Effect of small doses cimicifugæ on respiration and circulation of cat.

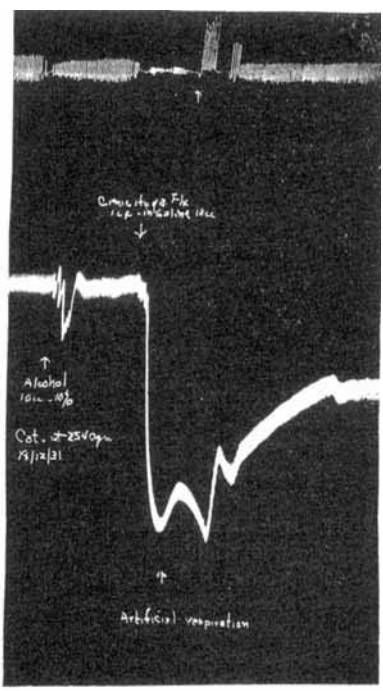

Fig. 6.—Effect of large doses cimicifugæ on respiration and circulation of cat.

by the phenolsulphonphthalein test. The effects on the blood pressure and respiration are illustrated in Figs. 5 and 6. In Fig. 5 are shown the blood pressure and respiratory curves of a cat weighing 3.9 kilo under ether-anesthesia. Fluidextract, 0.2 cc., was injected slowly into the femoral vein. A distinct and prolonged fall in the blood pressure was noted. The respiration, however, was not affected. In Fig. 6 are seen the effects of a larger dose of cimicifuga given by intravenous injection. Here 1.0 cc. of cimicifuga in saline solution was injected. This was immediately followed by a fall of the blood pressure to almost dead level

and paralysis of the respiration. The animal was saved by instituting artificial respiration and an injection of adrenalin. The fall in blood pressure and the effect on the respiration in this case cannot be explained by the amount of alcohol contained in the fluidextract because injection of even larger doses of alcohol alone in such animals produced only a very slight transient fall in blood pressure, if any at all, from which the animal rapidly recovered.

DISCUSSION.

The experiments described above clearly reveal that in the first place the fluidextract of cimicifuga is not an innocuous drug when administered in sufficient quantities and especially by injection. It produces a toxic effect on the circulation and respiration. Studies on the intestines and uterus indicate that cimicifuga extracts exert a depressant and paralyzing effect on isolated organs. Whether the same effect is produced in the living animal by administration of small doses of cimicifuga by mouth is very doubtful. It probably has no effect on either the intestines or uterus in such cases. Very large doses of cimicifuga preparations certainly have no specific effect on the uterus except indirectly through general poisoning of the animal. In this respect cimicifuga may be compared with the so-called emmenagogue oils, studied by the author some years ago (7). It was found that such drugs had no specific effect on the uterus and that when they did exert any action on that organ, it was only one phase of the general poisoning of the victim. As regards the action on the central nervous system, the psychopharmacological (8) experiments recorded above, and also the experiments with camphor, cast great doubt on the usefulness of cimicifuga as a sedative in chorea or similar conditions.

The writer has had occasion to make a survey of about one hundred thousand prescriptions, covering the last thirty years (1900–1931), in the files of the retail department of Hynson, Westcott & Dunning, Inc. These prescriptions were written, for the most part, by prominent medical practitioners in the City of Baltimore. Among the whole number there were certainly not more than a dozen prescriptions calling for cimicifuga or any of its derivatives. It is evident that modern clinical experience furnishes no favorable support for the therapeutic claims of cimicifuga in the earlier literature and agrees with the experimental results of the present investigation.

SUMMARY.

1. Fluidextract of cimicifuga and cimicifugin were studied in regard to their effects on the brain and neuromuscular apparatus, the intestines, the uterus, the circulation, the respiration and kidney function.

2. Even when administered in large doses, cimicifuga exerts very little sedative effect on the brain and neuromuscular apparatus of albino rats, as indicated by their behavior in the circular maze.

3. Large doses of cimicifuga, administered to rats by stomach tube, do not exert an appreciably inhibitory effect on epileptiform convulsions produced by injections of camphor.

4. Cimicifugin exerted no action on isolated surviving preparations of intestinal and uterine muscle. A suspension of the fluidextract of cimicifuga, however, tends to relax and paralyze such muscle preparations, thus indicating a depressant and poisonous property of the oily constituent of the drug.

5. Intravenous injections of suspension of fluidextract of cimicifuga produced marked depression of the circulation and respiration.

6. The results of the present investigation afford no scientific basis for the extravagant claims in regard to the therapeutic value of cimicifuga which are found in some of the old textbooks on medical practice and treatment.

BIBLIOGRAPHY.

(1) J. Young, *Am. J. Med. Sci.*, 9 (1831), 310.
(2) N. S. Davies, *Trans. Jour. Am. Med. Assoc.*, 1 (1848), 351.
(3) T. Lauder Brunton, "Pharmacology, Therapeutics and Materia Medica," Third Edition, Philadelphia (1889).
(4) H. A. Hare, "Practical Therapeutics," Eighteenth Edition (1922).
(5) William Osler, "Principles and Practice of Medicine," Sixth Edition (1905).
(6) Inaba and Janusche, *Z. ges. exptl. Med.*, 1 (1913), 129.
(7) D. I. Macht, *J. Pharmacol. and Exper. Therap.*, 4 (1913), 547.
(8) D. I. Macht and C. F. Mora, *Ibid.*, 16 (1920), 219.

PHARMACOLOGICAL RESEARCH LABORATORY,
HYNSON, WESTCOTT & DUNNING, INC.,
BALTIMORE, MARYLAND.

VEIN ISLETS AS MEANS OF IDENTIFYING DRUGS AND DETECTING ADULTERANTS.*

BY C. J. ZUFALL AND ALLEDA BURLAGE.

If one traces the vein of a leaf as it dwindles in size, he finds that it becomes too small to be seen with the naked eye and finally as he views it under the microscope he finds that it is composed of only one or two vessels which connect with similar branches of other veins. The small area of the leaf enclosed by these smallest branching veins was given the name "vein islet" by Benedict (3).

In some species the vein islet is composed of only a few cells while in others it is composed of many. The size of this area, however, in full-grown leaves, is fairly constant for any species of plant, as shown by Zalenski (1), Shuster (2), Benedict (3), Levin (4) and Ensign (5).

In 1929 Levin (4) used the size of vein islets to distinguish between drugs of closely related species. His work included the several species of Barosma, Erythroxylon, Cassia and Digitalis, and is the most reliable investigation of the subject. This led us to extend the investigation into the detection of adulteration and substitution of other drugs.

The investigators referred to above used various methods of determining the size of the vein islets, most of which were long and quite tedious. Zalenski measured the "combined length of the veins in one square millimetre." Benedict (3) made photographs of the leaves by means of an enlarging camera and counted the vein islets. Ensign showed that Benedict's method was inaccurate because, in uncleared leaves, the chlorophyll hides the minute veins in from 17 to 62 per cent of the cases. Ensign concluded "that any study of leaf venation made from uncleared leaves is wholly unreliable" (5).

---

* Scientific Section, Miami meeting, 1931.