A Study of the Constituents of Corn Smut.*

By William Stuart.

In connection with some studies upon corn smut, which were published in the twelfth annual report of the Indiana Experiment Station,[1] the question as to whether corn smut actually contained some principle injurious to farm animals was given some attention. This portion of the work, which was performed by the writer under the supervision of Dr. Arthur, was not completed in time for publication with the other studies mentioned. This work consisted in making extracts of the corn smut, and determining, by means of standard alkaloidal reagents, whether it contained an alkaloid or not. It also included a study of the physiological action of the extract upon horses, when administered to them either hypodermically or per orum. For the latter portion of the work the writer is greatly indebted to Dr. R. A. Craig, of the Veterinary Department, who administered the doses and observed its effects.

In the preparation of the extract valuable assistance was received from Mr. J. W. Sturmer, of the Purdue School of Pharmacy.

TESTS FOR ALKALOIDAL SALTS.

The methods employed in testing for alkaloidal salts were to make an alcoholic extract of the smut spores and such detritus as would pass through a fine sieve. A hundred grams of the smut spores were weighed out and, after thoroughly moistening them in an open dish with a 33⅓ per cent. solution of alcohol, they were again passed through a sieve to break up all lumps, then transferred to a percolator previously fitted up for the purpose. Sufficient alcohol, of the same strength as that previously mentioned, was added to cover the spores. Maceration of the spores was continued for twenty-four hours before any of the liquid was allowed to pass over into the receiving flask, the latter being so adjusted as to prevent it. At the end of this period the receiving flask was lowered so as to permit of about two drops passing over into the flask per minute. The percolation was continued until the percolate was colorless, sufficient

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* Abstract of an article published in the Thirteenth Ann. Rep. of the Ind. Exp. Sta., pp. 26–32, Jan., 1901.

(1) Arthur and Stuart, Twelfth Ann. Report Ind. Exp. Sta., p. 84–135, Jan ,1900.

alcohol being added from time to time to keep the surface of the spores covered with the liquid. The first 50 cc. of the percolate was set aside and the balance collected and evaporated down to 50 cc. on a steam bath. This was added to the first amount saved making 100 cc. of the extract. Each cc. of the extract representing one gram of the spores.

In testing the extract for alkaloids a certain amount of it was taken and evaporated to dryness on a steam bath. The residue was treated with a five per cent. solution of sulphuric acid, and filtered. The filtrate was then subjected to tests with the following reagents:

1. Potassium mercuric iodide (Mayer's solution).
2. Phosphotungstic acid.
3. Iodine in potassium iodide solution.
4. Picric acid.

A small portion of the filtrate was poured out into each of four watch-glass crystals and then a drop or two of the reagents added. The reactions obtained by this method were as follows:

Reagent 1. A slight milky turbidity was produced.

Reagent 2. A decided milky turbidity was obtained.

Reagent 3. No visible reaction.

Reagent 4. No visible reaction.

A number of tests with the same and with fresh lots of extract prepared in the same manner gave similar results.

TESTS FOR TOTAL ALKALOIDS.

In testing for total alkaloids a modified "Prollius Fluid"[2] was used. Two methods were employed. The first was to treat two grams of the smut for four hours with 50 cc. of "Prollius Fluid" in a well stoppered conical flask. The contents of the flask were vigorously shaken at intervals during that period. After macerating four hours the supernatant solution was drawn off and filtered. The filtrate was evaporated to dryness on a steam bath and the residue treated with a five per cent. solution of sulphuric acid. The acid solution was filtered and the filtrate tested as mentioned for the alcoholic extract. The reactions obtained were in each instance similar to those given for alkaloidal salts.

The second method employed consisting in macerating ten grams of the smut spores in 100 cc. of "Prollius Fluid" for twenty-four hours. The

[2] Modified Prollius Fluid: Ether, 250 c. c.; Chloroform, 100 c. c.; Alcohol, 25 c. c.; 28% Ammonia, 10 c. c.

flask containing the spores being agitated at frequent intervals during that period. The supernatant liquid was drawn off and filtered, and 50 cc. of it transferred to a separatory funnel and subjected to the "shaking out" process as outlined in Sturmer and Vanderkleed's "Course in Quantitative Analysis: 61-64, 1898, under 'Process 1.—General for Total Alkaloid.'" The results obtained from this method by the reagents were quite similar, although more marked, to those of the preceding ones.

Reagent 1. A slight turbidity was obtained which, on standing for some time, deposited a dark brownish substance on the bottom of the glass.

Reagent 2. A marked cloudiness was obtained which, on standing for some time deposited a whitish crystalline precipitate on the bottom of the glass.

Reagent 3. No visible reaction or any deposit after standing.

Reagent 4. No visible reaction, but on standing a slight deposit was noticed on the glass.

TESTS FOR ALKALOIDS IN COMMERCIAL EXTRACTS OF ERGOT AND CORN SMUT.

The uniformity of the results obtained from the reagents employed, the first two giving positive and the last two negative reactions in each instance, led to an examination of the commercial extracts of both ergot and corn smut.

Ergot of rye test.—The commercial fluid extract of ergot was obtained from a leading wholesale druggist in the city, whose supply was obtained from the well-known firm of Park Davis & Co., of Detroit, Michigan. The fluid extract was evaporated to dryness over a steam bath, the residue treated with dilute sulphuric acid and filtered. Tests of the filtrate were made, and the reactions obtained were as follows:

Reagent 1. A yellowish brown, curdy-like precipitate was obtained.

Reagent 2. A cloudy white precipitate was obtained which on standing changed to a purplish brown, curdy-like substance.

Reagent 3. A reddish brown precipitate was obtained.

Reagent 4. No visible reaction obtained.

Corn smut ergot test.—The material used was obtained from the same local druggist, who in turn received his supply from the well-known firm of Merrill & Co., Cincinnati, Ohio. The fluid extract was treated in the

same way as in the preceding test and the reactions obtained were somewhat similar.

Reagent 1. A precipitate was formed, but it was not so marked as in the ergot of rye.

Reagent 2. Reaction much the same as that in rye ergot.

Reagent 3. Reaction not quite so marked as in the rye ergot.

Reagent 4. No reaction was obtained.

A brief summary of the work shows that a substance was obtained in all the extracts made which gave positive reactions with the first two reagents used and negative ones with the last two.

Commercial extracts of rye ergot and of corn smut gave similar reactions to those obtained from the corn smut extract prepared in the laboratory, in the case of reagents one and two, and in addition gave marked results with reagent three.

PHYSIOLOGICAL EFFECT OF AN ALCOHOLIC EXTRACT OF CORN SMUT UPON HORSES.

The study of the physiological effect of an alcoholic extract upon horses was carried on in conjunction with that of the alkaloidal tests in the laboratory, the alcoholic extract used being prepared by the writer in the same manner as that described in the preceding pages. The experimental work upon the horses was performed by Dr. R. A. Craig, of the Veterinary Department of Purdue University.

The appended notes upon the amounts and effects of the doses administered were taken by him and have been kindly placed at my disposal.

Horse No. 1.—A gelding, poor in flesh, but healthy, was given 15 cc. of the extract subcutaneously. The dose seemed to have no effect. The next day 30 cc. were given in the same way. In twenty-five minutes he stopped eating. The pulse and breathing were quickened and the peristaltic movements of the intestines were increased. Forty-five minutes after the drug was given faeces were passed. No further effects were noted.

Horse No. 2.—A gelding in good condition was given 25 cc. subcutaneously. In twenty minutes he became restless, stopped eating, and the pulse and breathing were quickened. A moist evacuation of faeces occurred in twenty-five minutes. An hour after giving the injection its effects had passed off. Two days afterwards 45 cc. were given. The horse

soon became restless, the intestinal murmurings were loud and an evacuation of faeces soon followed. When made to turn in the stall his movements were slow and unsteady. One hour after giving the injection his pulse was sixty and his respirations forty-three per minute. He refused to eat and remained dull till noon the following day. After an interval of a few days the horse was given 130 cc. per orum. In forty minutes he stopped eating, his pulse and breathing were quickened, but outside of this no other effects of the drug were noted.

A brief summary of the results show that an injection of 25 to 30 cc. of the drug caused restlessness and increased peristaltic movements of the intestines. This was followed shortly by evacuation of the contents of the rectum. At the same time the pulse and respiration were quickened. The effects of the dose passed off in an hour.

The injection of 45 cc. produced, in addition to the above symptoms, a dullness and an unsteady gait when made to move. The effects of the dose were much more lasting. The horse remained dull and refused to eat for twenty-four hours.

A 15 cc. subcutaneous injection and a 130 cc. per orum dose produced but little effect.

While the results of both the chemical and physiological tests of the corn smut are at variance with those obtained by some other investigators,[3] they are in accordance with results of a number of chemists,[4] and to some extent in their physiological action to that obtained by Dr. Mitchell,[5] whose experiments were performed upon the frog. The concordance of the results obtained from both the chemical and physiological tests would indicate the presence in minute quantity of some narcotic in corn smut. What this narcotic is, and why, when corn smut is consumed in large quantities by farm animals, it does not produce more harmful results, are questions which are yet to be determined.

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[3] Kedzie, Bull. Mich. Exp. Sta., No. 137 : 45, 1896.
Mayo, Bull. Kans. Exp. Sta., No. 58 : 69, 1896.

[4] Dulong, Journ. de Pharm. 14 : 556, 1828.
Cressler, Amer. Journ. Pharm. for 1861 : 306.
Parsons, Rep. Dept. Agric. for 1880 : 136–138, 1881.
Hahn, Amer. Journ. Pharm. 53 : 496, 1881.
Rademaker and Fischer, Med. Herald for 1887 : 775.

[5] Mitchell, Jas.—The Physiological Action of Ustilago maidis on the Nervous System, Inaug. Thesis, Univ. Pa., 1883. Therap. Gaz., Detroit, 10 : 223–227, 1886.