AN INVESTIGATION OF GLEDITSCHIA TRIACANTHOS (LINNÉ).*

BY Y. THOMAS OESTER.

These studies on *Gleditschia triacanthos* (Linné), the common honey locust, had their origin at the University of Notre Dame. As early as 1926, King (1) at Notre Dame discovered that *Gleditschia triacanthos* possessed pharmacodynamic activity. Rudduck (2) extended these pharmacodynamic studies; Simons (3) succeeded in isolating an active principle which possessed marked oxytocic properties. He called this principle *hypoxysin*. Two Japanese workers, Matshshima and Kubota (4) isolated a potent glucoside or saponin from a related species, *Gleditschia horrida*. Their only published pharmacological study was upon the toxicity of the glucoside. Simons showed that his constituent did not respond to the common glucoside tests. The latest work (5) was that of the writer.

EXTRACTION OF ACTIVE PRINCIPLES.

During the past three years investigations have been undertaken with these objectives in mind:

1. Detailed study of the blood pressure reaction of the drug, which Simons (*loc. cit.*) indicated was present in the leaves; also allied pharmacological studies.
2. Determination of the nature of this principle.
3. Determination of its mode of action.
4. Comparison study of this principle, contrasted with the common pressor and depressor substances.

In the preliminary work, the statements of the previous investigators were substantiated, that aqueous percolates of the leaves are the most potent. Various menstrua and methods of extraction were tried but aqueous percolation appears to be the most suitable. There is, however, a large amount of tannin and coloring matter in all aqueous extracts. Their presence offers difficulties in isolating the active constituent. The *hypoxysin* was removed by the method of Simons, and all work was done with the hypoxysin-free extracts.

No successful procedure has been developed for the isolation of the active blood pressure constituent. This, despite the use of the following procedures:

1. Original percolations were made using alcohol, acetone, acidified acetone, chloroform, ether and petroleum ether. Injections of aqueous extracts from dried residues of these percolates gave no marked blood pressure reactions.
2. Normal aqueous percolations, acid and basic, were subjected to a shaking out process with the following solvents; ether, chloroform, petroleum ether, carbon tetrachloride, ethyl acetate, amyl alcohol and benzene.
3. Dried aqueous extracts were treated with the following reagents; alcohol, ether, chloroform, benzene, carbon tetrachloride, acetone, xylene, acidified alcohol and acidified acetone.
4. The following agents, animal charcoal, kaolin, picric acid, hexamethylenetetramine, lead oxide, magnesium oxide and purified talc, failed to separate the potent constituent from the extraneous matter.
5. The procedure as detailed by Fuller (6) was also followed.
6. Steam distillation failed to bring about a separation of the active constituent.

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PHARMACODYNAMIC ACTIVITY.

The following studies were made dealing with the pharmacodynamics of the aqueous extracts after the removal of the *hypoxysin:*

1. The oxytocic nature of *hypoxysin* itself was substantiated by experiments upon the isolated uterus of the guinea pig. Pittenger's procedure (7) was used.

2. Intravenous injections of aqueous extracts produced no marked influence upon the respiration of the etherized dog. A tambour encircling the chest was used to record the respiration.

3. The aqueous extracts exert some influence upon voluntary muscle. In the frog's gastrocnemii, there was a noticeable increase in the amount of work, and fatigue was delayed.

4. There is very little effect upon ventricular contraction when the extract is injected intravenously. Only during the period of lowest blood pressure was there a noticeable decrease in ventricular systole. The Guthrie myocardiograph was used to record the ventricular response.

5. In direct perfusion and irrigation of frog's and turtle's hearts, recorded by the suspension cardiograph, outlined by McGuigan (8), there was at first a slight decrease in frequency although an improvement in force. Upon extended perfusion or irrigation, however, both frequency and force are decreased.

6. The blood pressure activity of the Gleditschia extracts consists chiefly in a marked lowering of blood pressure. Following this there is a gradual return to normal. The reaction seems to be more effective than that following the use of the common nitrites.

7. The depressor effects appear to be due to direct action of the drug on the musculature of the blood vessels because:

(*a*) The heart frequency and force are not markedly affected by the injections of the drug.

(*b*) Adrenalin, acting as a stimulant to the nerve endings in the arterial wall, normally brings about a constriction. This action is unimpaired by the previous use of the Gleditschia extract. From this result it is apparent that the extract does not affect the nerve endings. Therefore it is assumed that the arterial musculature and Rouget cells produce the change in blood pressure.

8. Direct experimentation on arterial rings from the sheep's carotid artery showed that there is a decrease in tone upon the application of Gleditschia extract. The experimental procedure used was the same as that outlined for the isolated uterus.

DEPRESSOR CONSTITUENT.

A large number of experiments upon the blood pressure of the etherized dog lead to the following conclusions:

1. The blood pressure constituent may be extracted from the crude drug by the use of an aqueous menstruum.

2. The blood pressure constituent is not soluble to any marked extent in the following solvents: ether, ethyl acetate, carbon tetrachloride, carbon disulphide, petroleum ether, amyl alcohol, acetone or benzene.

3. Changes in p_H within a small range, either above or below 7.35, do not affect the activity of the principle to any marked degree.

4. No toxicity was ever noted on any of the injections, although some represented as much as four Gm. of the crude drug.

5. Aqueous extracts of the drug gave negative tests for alkaloids and glucosides. It appears from the general nature of the extracts that the active constituent is a complex neutral principle, of a gummy consistency.

SUMMARY.

The common methods of separating active constituents are of little value when applied to aqueous extraction of *Gleditschia triacanthos.*

There appear to be two active constituents, *hypoxysin*, isolated by Simons, and a second which has marked depressor action.

From a preliminary investigation of the extracts it would seem that the active blood pressure constituent is a neutral principle of gum-like consistency.

The depressor action is best obtained from aqueous extracts.

The depressor activity seems to be caused by a direct action of the drug upon the musculature of the blood vessel.

The drug has very little effect upon the heart rate or force. The respiration is uninfluenced by the injections of the drug.

Administration of the drug causes an increase in the amount of work performed by voluntary muscle, also a delay of fatigue.

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ASSAY OF BARBITURIC ACID DERIVATIVES.

Barbituric acid derivatives, when mixed with silver nitrate, form soluble silver compounds which are only very slightly dissociated and therefore do not form silver oxide with alkali. This reaction may be used for their assay, as follows: 0.2 to 0.3 Gm. is dissolved in 30 mils of water with the addition of 1 Gm. of anhydrous sodium carbonate, and $N/10$ silver nitrate is run in until there is obtained a distinct turbidity which remains for some time. One equivalent of silver then corresponds to one molecule of the barbituric acid derivative. The method may be used for diethylbarbituric, phenylethylbarbituric, cyclohexenylethylbarbituric, diallylbarbituric, isopropylbrompropenylbarbituric and butylbrompropenyl barbituric acids, but not for *N*-methylated derivatives such as evipan and prominal.—H. BUDDE in *Apothekerzeitung*, 49 (1934), 295, through *Quarterly Journal of Pharmacy and Pharmacology*.

CINCHOPHEN POISONING.

A. J. Quick (New York) regards the acute yellow atrophy of cinchophen poisoning as a form of allergic reaction—Arthus's phenomenon. This is a severe localized inflammatory process which may be produced in animals by injection of a protein. The reaction is due to a metabolic derivative, not to the cinchophen itself. Quick advises certain precautions in its administration: the drug should be given under medical supervision and large doses should be avoided; it should not be given to persons sensitive to foreign proteins, and such proteins should not be given at the same time; cinchophen should never be given intravenously, and it should be given with caution to patients with damaged livers. In acute yellow atrophy glucose and insulin (5 to 10 units three times a day) should be given; calcium gluconate and liver extract are also of value.—*Am. J. Med. Sci.*, Jan. 1934, 115–121. Through *The Prescriber*.