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THE POSSIBLE INFLUENCE OF ETHER ANESTHESIA ON THE ACCURACY OF THE CAT METHOD OF DIGITALIS ASSAY.*

BY H. B. HAAG.

Assays of tincture of digitalis were carried out on decerebrated cats to determine whether ether anesthesia affected the accuracy of the cat method in the assay of digitalis.

Of the many objections raised to the cat method of digitalis assay one frequently brought forward is the possible influence of the anesthetic used. Berardi has recently called attention to the fact that etherized dogs manifest toxic symptoms following doses of digitalis which control animals tolerate with impunity. Here in our laboratory results have been obtained which have led us to believe that digitalis somewhat lowers the resistance of animals to poisoning by ethyl alcohol. Since ethyl alcohol and ether are related both chemically and pharmacologically the question arose in our minds as to the possible influence which ether might exert in the cat method of Hatcher. In an attempt to determine this digitalis was assayed on several series of decerebrate cats, and results here obtained compared with those in which etherized animals were employed.

The method used in preparing the decerebrate beasts was, with the exception of a few modifications, that described by Pollock and Davis of Chicago, the efficacy of which depends on the production of cerebral anemia following ligation of both carotids and the basilar artery. To avoid the use of ether, the animals were anesthetized with a mixture of nitrous oxide and oxygen, and were then tied to the operating table, the head being placed in a head holder. Both carotids were next exposed and ligated and a cannula inserted into the trachea. The gas mask was then removed, the anesthetic being continued by way of the cannula. The mouth was held open by a suitable mouth-gag, and the tongue brought outside the oral cavity and held in this position by a cord. A median line incision was made in the soft palate from the posterior edge of the hard palate, thence caudad to the free border of the soft palate. The resulting flaps were retracted so as to better expose the underlying field. Next the mucous membrane and muscles of the base of the skull were dissected laterally in such a way as to bring into view the anterior border of the foramen magnum and the tympanic bullae. By means of a curette the bony field was cleared of adhering shreds of mucous membrane and muscle. A trephine opening was then made at a point midway between the tympanic bullae, about one centimeter anterior to the anterior border of the foramen magnum. For this procedure a foot-driven dental burr, of the Vulcanite variety, was used. This operation exposed the underlying *dura mater* which was then punctured and removed; this was to allow escape of the

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cerebro-spinal fluid and bring more fully into view the basilar artery. The artery was gently dissected and a Crile clamp applied, effort being made to place it so as to occlude the artery just caudad to the origin of the posterior superior cerebellar artery; following closure of this vessel the marked extensor position so characteristic of decerebrate rigidity ensued. Frequently at this point respiration became somewhat embarrassed, necessitating artificial respiration for a few minutes. The entire operation generally required about thirty minutes, occasioned no shock and very little hemorrhage.

Pollock and Davis state that in their laboratory cats decerebrated by their method frequently survived for twenty-four hours without any special attention. None of our control decerebrate animals lived as long as twenty-four hours, but we believe that the condition of the respiration and circulation remained fairly intact for three or four hours after the operation. And as the assays were made during this interval, the results might be considered indicative of the potency of the drug as determined on an animal of normal vital functions.

To allow recovery from the anesthesia and the operation, the assay was not begun until thirty or forty minutes after ligation of the basilar artery. The animals were then arranged in the usual way; the tincture, diluted one to ten with saline, being given by way of the femoral vein at the rate of one cubic centimeter every two minutes until death. The results of this are furnished by Table I.

TABLE I.—RESULTS OF DIGITALIS ASSAY ON DECEREBRATE CATS.

Cat No.	Sex.	Wt. in Kgm.	Total cc. dil. tr.	Mgm. leaf × Kgm. body wt.
1	Female	1.915	15.6	81.4
2	Male	2.670	17.0	64.0
3	Female	1.715	13.8	80.4
4	Female	2.650	20.0	75.4
5	Male	3.165	13.0	41.0
6	Male	4.060	25.0	61.3
7	Female	1.990	11.6	58.2
8	Female	2.595	17.0	65.5
9	Female	2.270	14.5	63.8
10	Female	2.965	17.0	57.3
12	Female	2.020	11.2	55.4
13	Female	1.700	15.5	91.1

Average mgm. leaf × Kgm. body weight = 66.2.

TABLE II.—RESULTS OF DIGITALIS ASSAY ON ETHERIZED CATS.

Cat No.	Sex.	Wt. in Kgm.	Total cc. dil. tr.	Mgm. leaf × Kgm. body wt.
1	Female	2.86	19.10	66.70
2	Male	2.77	23.10	83.30
3	Male	3.20	21.20	66.20
4	Female	2.75	20.40	74.10
5	Male	2.70	21.10	78.10
6	Female	2.10	14.80	70.50
7	Female	2.31	14.20	61.50
8	Male	3.35	26.80	80.00
9	Male	3.54	25.00	70.60
10	Female	2.215	22.10	99.80

Average mgm. leaf × Kgm. body weight = 75.0.

For purposes of comparison a second assay was made using etherized cats, precaution being taken that the anesthetic be very light throughout the entire procedure. Table II is a summary of these results.

DISCUSSION AND CONCLUSIONS.

Judging by our experiments we believe that the method of decerebration as elaborated by Pollock and Davis is free from many of the objections which embarrass methods heretofore described.

A comparison of the tables shows a difference of 8.2 mgm. leaf per kilogram cat in the two assays: this might be taken to indicate an unfavorable influence exerted by the procedure of decerebration. In our experience, however, it is rarely possible to have different series of etherized cats give results on the same preparation of digitalis which check more closely than this. So far as the number of experiments justify, it may be assumed that the light ether anesthesia necessitated in the Hatcher-Brody cat method of digitalis assay does not materially influence the resulting minimal lethal dose.

We owe our thanks to Parke, Davis and Co. for their generosity in furnishing the digitalis leaf used in these experiments.

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STUDIES OF THE VITAMIN POTENCY OF COD-LIVER OILS—XXI—THE STIMULATION OF REPRODUCTION BY FAT-SOLUBLE VITAMINS.*

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In a previous paper³ it was reported that the addition of a vitamin-rich cod-liver oil to the ration of laying birds at the rate of one pint per 100 birds per week caused a material increase in the number of chicks hatched. These data regarding the influence of cod-liver oil on reproduction were obtained in a study conducted with three different breeds of domestic fowl maintained under a variety of conditions as regards ration and management.

The present investigation was undertaken to secure additional data concerning the effect of fat-soluble vitamins on reproduction, particularly with respect to the effect of various amounts of supplementary vitamin feeding.

EXPERIMENTAL PROCEDURE.

The experimental birds in this instance were eight months old Rhode Island Red pullets. The experimental period was of thirty-two weeks' duration, which

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