(4) Official squill preparations retain over half of their activity for a period of one year.

(5) The simple determination of the M. L. D. of a squill preparation by the well-known cat method does not yield a reliable indication of potency. The M. L. D. of a standard substance, such as ouabain or scillaren A, must be determined by the identical technique used for the sample, and the results expressed in terms of the standard instead of "cat units."

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FOOT-NOTE: The Sandoz Chemical Works very kindly supplied the scillaren A and B used in this work.

SALIVA TESTS. II. HEROIN.

BY JAMES C. MUNCH.*

Success in using the mouse test for the detection of morphine (1) suggested the possibility of employing it for the detection of heroin in the saliva of horses and in pharmaceutical products (2). A standardized technique was developed for this saliva test (1).

A series of normal mice, weighing approximately 20 Gm., were injected subcutaneously with heroin, and various symptoms observed over a period of half an hour. In general, the *S*-tail curve resembles that produced by morphine. Literature reports (2) indicate that morphine and heroin are equally potent, threshold doses being stated to be 10 gamma per 20-Gm. mouse, or 0.5 mg. per Kg. We found the threshold dose of heroin to be much smaller (Table I). In addition to the tail curve, mice injected with heroin showed a series of symptoms differing from those following the administration of morphine. The mice tended to become much more restless, and hyper-irritable. A common heroin symptom was the

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development of a definite running reflex. Very shortly after the injection of an effective dose, mice tended to go to the periphery of their cages, and start an endless parade round and round from right to left, or vice versa. If two injected mice were placed in the same cage, but facing in opposite directions, they developed a traffic code; on meeting, one mouse consistently swerved to the right, the other to the left.

Heroin was injected subcutaneously into horses. Definite evidence of stimulation was observed by a veterinarian after 32.5 mg. per horse. The injection of 16 mg. per horse, or one-half the previous dose, failed to affect the horse so far as the verterinarian could determine. The mouse test on samples of saliva was positive with each dose (Table II). The horse receiving 16 mg. was also bled thirty minutes after injection. The blood was allowed to clot and tests conducted upon the serum. No effect was observed on mice, following the injection of 1 cc. of serum; 2 cc. uniformly produced a positive tail-curve.

TABLE I.—MOUSE-TAIL RESPONSES AFTER INJECTION OF HEROIN.

Dose, Mg./Kg.	Per Cent Mice Showing Positive Reaction.
0.025	0
0.04	25
0.05	100
0.0625	100
0.07	100

TABLE II.—DETECTION OF INJECTED HEROIN IN HORSE'S SALIVA.

Horse No.	Weight.	Heroin Injected. Mg./Horse.	Gamma/Kg.	Veterinary Deductions.	Mouse Test. Tail-Curve.	Excitement.
135	570	325	570	Stimulation	Positive	+ + +
9366	500	162.5	325	Stimulation	Positive	+ + +
135	570	65	115	Stimulation	Positive	+ +
135	570	32.5	57	Stimulation	Positive	+
135	570	16	28	Normal	Positive	0

The running reflex and the increased irritability appeared to differentiate heroin from morphine. Heroin solutions in the saliva of untreated horses, in normal horse serum, and in distilled water, were identical in action. Saliva collected from horses receiving 16 to 65 mg. per horse gave a morphine, rather than a heroin type of response, suggesting that heroin is broken down to morphine in the body of the horse. Following larger doses of heroin, positive mouse-tail tests were not observed until an hour after administration, and the effect persisted in salivas collected twenty-four hours afterward. However, salivas collected after 15 and 30 minutes produced marked excitement, followed by depression. This excitement and depression decreased in intensity with decreasing doses, and was not observed after the administration of 16 mg. of heroin. After the subcutaneous administration of heroin to horses, some substance is excreted in the saliva which produces effects upon mice similar to those produced by morphine.

Mouse tests were made by direct intraperitoneal injection of a series of very old solutions of pharmaceuticals containing heroin, such as Linctus Compound, Syrup of Heroin Compound and Elixir of Glycerin and Heroin Compound. The undiluted products produced typical tail-curves, restlessness and the running reflex of heroin, but convulsions developed, and the animals died within ten to thirty minutes. Marked opacity of the pupils developed shortly before death.

Dilute solutions produced typical tail-curves, marked depression rather than stimulation, and blindness.

CONCLUSIONS.

1. Physiological actions upon mice serve to detect heroin in saliva, as well as in pharmaceutical preparations.
2. Heroin is more effective than morphine on mice.
3. After administering heroin, the saliva of horses contains a substance giving a morphine reaction on mice.

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DRUG EXTRACTION. I. A STUDY OF VARIOUS MENSTRUA FROM THE STANDPOINT OF SWELLING EFFECTS, PENETRATION AND EXTRACTION.

BY WILLIAM J. HUSA AND LOUIS MAGID.

(*Concluded from page 1103, November Journal.*)

EXTRACTION OF BELLADONNA ROOT OF DIFFERENT DEGREES OF FINENESS.

Filtration Method.—A technique was used by Husa and Fehder (14), whereby imbibition and extraction of powdered drugs could be determined in a process of maceration. An outline of the method as applied to powdered belladonna root is as follows:

Ten grams of powdered belladonna root were macerated for 15 minutes with 90 Gm. of menstruum in a 250-cc. Erlenmeyer flask in a thermostat at 30° C., during which time it was agitated every five minutes. The mixture was then filtered and after allowing 15 minutes for completion of draining, the filtrate was weighed and the weight of the filter paper with the wet marc was also determined. Similar macerations were made for periods of 1 hour, 5 hours and 24 hours, with less frequent agitation during the longer intervals.

In the filtrate, the percentage of dissolved solids was determined by weighing exactly ten Gm. of filtrate in a tared 50-cc. beaker, evaporating to dryness on a water-bath and drying to constant weight in an oven at 105° C. The filtrate was assayed for alkaloidal content according to the U. S. P. X assay for Tincture of Belladonna except that 25 Gm. of the filtrate were used. The wet marc was also assayed for alkaloidal content according to Type Process B, designated for belladonna root by the U. S. P. X.

The amount of moisture in the powdered drug was determined by the U. S. P. X method for drugs containing no constituents volatile at 100° C. The amount of dry marc was calculated as follows: (weight of drug taken for extraction) minus (weight of moisture in drug) minus (weight of dissolved solids in filtrate) = (weight of dry marc). The weight of the liquid imbibed by the marc was calculated as