and eyes and periodic bursts of activity. But while resting it lies with stiffly extended legs, and the periods of activity are characterized by generalized muscle spasms which sometimes propel the animal across the floor. If set on its feet, the cat tends to take several quick, spastic, trot-like steps before it falls. This is the phase of the activity which most closely resembles the morphine mania seen in intact cats. It differs mainly (1) by the inability to assume or maintain an upright position, (2) by pronounced extensor spasm of the legs and (3) by quicker onset of fatigue.

Both intact and decorticated cats a few hours after morphine administration show a heightened "startle" reflex to noise (clapping of hands) and to tapping the animal. This effect is exaggerated in the decorticated cats and closely resembles the tetanus of early strychnine poisoning. Two of the decorticated animals in this condition were given a half-anesthetic dose of sodium amytal (25 mg. per kg., intraperitoneal) which uncovered vigorous running movements not seen in intact cats similarly treated.

Summary. The long-surviving cat after bilateral cerebral decortication with degenerative changes in the striate nuclei responds to morphine with an excitement which is quite similar to the effect on intact cats, but with certain modifications described in the text. The excitant action of morphine must, therefore, be mediated by subcortical centers.

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A Method for Determining Blood Volume in Rats.

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Methods for determining the blood volume of small animals are available, but all have one of two objections: (1) they require so much blood as to affect the subsequent blood volume of the animal, or may even cause its death; (2) they require the laborious preparation of standards consisting of dye-containing serum in uniform capillary tubes. Such standards are then presumed to be permanent.

Our method requires but 50 cmm. of blood, and the standard can

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be set up for each period of work in a few minutes. Moreover, the blood volume is determined directly, and not calculated from the serum volume by means of the hematocrit reading.

Adult albino rats have been used. Three mg. of nembutal per 100 gm. of body weight are injected intraperitoneally. As soon as the animal is sufficiently quiet, the jugular vein is exposed at the base of the neck by an incision parallel to its course. Then 0.3 cc. of 5% vital red is injected into the dilated bulb. The manner of injection is important. A one cc. tuberculin syringe fitted with a fine hypodermic needle is used. Exactly 0.3 cc. of the dye is drawn into the syringe, reading the mark against a strong light. The needle, pointing toward the rat's head, is introduced into the upper border of the pectoral muscle, grazes the upper edge of the clavicle and enters the vein through the muscle. The point is carried onward until it is visible in the jugular bulb. It is moved up and down to be sure it has not picked up the vein wall, after which the dye is injected under inspection. We wait 20 to 25 seconds while the vein clears of dye. Then the needle is quickly withdrawn and pressure exerted momentarily over the muscle. Leakage does not occur. This is the only method we have been able to find or to devise by which small intravenous injections can be given quantitatively in the rat.

After 3½ minutes the tail is immersed in water at approximately 45°C. for one minute. Then, 4½ minutes from the time of the injection, a vein in the tail is punctured, and 50 cmm. of blood is withdrawn into a graduated pipette. The puncture wound is sealed with collodion. The 50 cmm. of blood are placed in 2.95 cc. of physiologic saline (3 cc. from which 50 cmm. have been removed) and centrifuged. The supernatant fluid is pink.

The standard is prepared by adding 0.3 cc. of 5% vital red to 4 cc. of physiologic saline, the resulting volume being 4.3 cc. This is the primary standard. Standard A is made by adding 50 cmm. of this to 5.95 cc. of physiologic saline (6 cc. from which 50 cmm. have been removed). Standard B is made by adding 100 cmm. of the primary standard to 5.9 cc. of physiologic saline.

We use a microcolorimeter† requiring 2 cc. of fluid in each cup. The standard cup is connected by a sidearm to a 1 cc. tuberculin syringe graduated to 0.01 cc. The volume of the standard in the standard cup (VS) is therefore 2.00 minus the amount in the syringe, read to the second decimal. In an actual determination

† The colorimeter used is the Vim-Sheftel Microcolorimeter, made by the MacGregor Instrument Co., Needham, Mass.

2 cc. of the unknown are introduced into the cup for the unknown. This is the pink supernatant fluid obtained after centrifugalization. Two cc. of standard A or B are introduced into the standard cup, the one being selected which appears slightly darker than the unknown. Standard is then withdrawn into the syringe until the colors match. The formulas are: for standard A, blood volume = 17.2/VS; for standard B, blood volume = 8.6/VS.

Results. The average blood volume in 20 rats was 4.3 cc. per 100 gm. of body weight, varying from 4.1 to 5.3 cc. The smaller animals have the larger blood volumes in terms of body weight. Only 3 animals in this series had body weights below 160 gm., and only these had blood volumes of 5 cc. or more per 100 gm. of body weight.

If the course of disappearance of dye is followed at 15 minute intervals after injection, it is found that a gradual loss occurs reaching 11 to 12% in one hour.

As a control, 4 rats were injected by the same technique with 0.3 cc. of 0.4% vital red, and exsanguinated at the end of 4½ minutes. One cc. or more of serum was obtained and this, undiluted, was read in the colorimeter against a standard set up in normal serum. These rats had an average blood volume of 4.8 cc. per 100 gm. body weight, varying from 4.4 to 5.2.

Method for Repeated Determinations. This method is applicable when dye from a previous injection is still present in the blood stream. The only change is in the making up of standards A and B. It is estimated from the size of the animal which standard, A or B, will be required. If there is doubt, both must be made up. Take 2.95 cc. of the selected standard and add to it 50 cmm. of blood taken just before the second dye injection. Centrifuge, and place 2 cc. of the supernatant fluid in the standard cup. The remainder of the procedure and calculation is unchanged.

Our results agree with those of Cutting and Cutter,[1] who determined serum volume but who give hematocrit values in their protocols from which blood volume can be calculated. They disagree with the results of Went and Drinker[2] who, in 7 animals, found an average blood volume of 7.4 cc. per 100 gm. of body weight with a variation of 6.9 to 7.9.

1 Cutting, W. C., and Cutter, R. D., *Am. J. Physiol.*, 1935, **113**, 150.

2 Went, S., and Drinker, C. K., *Am. J. Physiol.*, 1929, **88**, 468.