Potentiation of an Effect of Morphine in the Rat by Sera from Morphine-Tolerant and Abstinent Dogs and Monkeys*

By CONAN KORNETSKY and GLENN F. KIPPLINGER**

With 1 Figure in the Text

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KRUGER, EDDY, and SUMWALT (1941) concluded in a review that there was no evidence that serum from a morphine-tolerant animal had any effect on the magnitude of response of test animals to morphine. In all of the experiments reviewed, death of the test animal was used as a criterion of morphine effect. The present study was initiated with the belief that if serum from a morphine-tolerant animal had any effect on the subsequent response of test animals to morphine, it would most likely manifest itself when sublethal doses of morphine were used.

Method

In order to test the above hypothesis, five mongrel dogs (one male and four females) were placed on gradually increasing doses of morphine sulfate (MS) so that at the end of a two-week period they were receiving 60 mg/kg daily in two 30 mg/kg doses. At the beginning of the two-week period, the initial doses of 5 mg/kg of MS to the dogs caused defecation, vomiting, and deep depression as indicated by difficulty in arousing the animal. When animals were aroused, they exhibited a marked ataxia. Later doses of 30 mg/kg produced only mild ataxia, if any, and a somnolent state from which the animal readily could be aroused. This decrease in the depressant effect of MS in the later doses indicated that tolerance to MS was obtained. Although the dogs who received MS lost weight during the first week of daily drug administration, their weights stabilized at about 10 percent below their control weights during the second week. The weights of the control animals did not change during this

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** Now at the Department of Pharmacology and Toxicology, University of Texas — Medical Branch, Galveston, Texas.
period. Both groups of dogs were placed on a diet of canned meat during the first week because the MS animals would eat the canned meat, but they would not eat their usual dry chow. None of the dogs were emaciated or obviously fat at the end of the two-week period. As an additional indication of nutritional state, serum glucose and total protein levels were determined from the serum of two experimental animals and two control animals. The serum glucose levels and the total protein levels for the MS-treated animals were 66 and 80 mg-% (glucose) and 5.6 and 5.7 gm-% (protein). The levels for the control animals were 70 and 80 mg-% (glucose) and 5.9 and 4.9 gm-% (protein). These are fasting levels.

Drug administration was abruptly terminated at the end of this two-week period, and 48 hours later the animals were exsanguinated. Upon termination of MS administrations, the signs of abstinence were mild and lasted about 24 hours. They consisted of mild, but noticeable, hyperirritability and restlessness. Control dogs (one male and three females as in the experimental group) were exsanguinated at the same time as the animals that received daily MS injections. All dogs were exsanguinated under anesthesia. Animals were anesthetized by rapidly injecting intravenously 25 mg/kg of thiamylal sodium (concentration: 50 mg/ml). If anesthesia did not appear to be complete, the animal was given an additional 10 mg/kg. There was no trend for either group to require more thiamylal on a mg/kg basis to produce anesthesia.

Sera was prepared individually from the blood of each animal and frozen. Thimerosol (1:20,000), an organic mercurial antiseptic, was added as a preservative to the serum. Sera was prepared by bleeding the animal into a clean two-liter flask, allowing the clot to form, freeing the clot from the walls of the vessel, allowing the clot to retract, decanting the serum, centrifuging to remove any clot debris, adding preservative, and freezing. The animals were exsanguinated in the morning, the blood decanted in the afternoon, and centrifuged and frozen the following morning. After bleeding the animal all steps were carried out at 4°C. Despite precautions, it was not possible to prepare the sera without some degree of hemolysis. Sera from individual donor animals were not pooled at any time.

Sera prepared from the blood of two rhesus monkeys also were used1. One of the monkeys had been receiving a total daily dose of 12 mg/kg of MS for 33 months. The blood from this monkey and one non-tolerant control monkey were drawn without anesthesia or exsanguination 48 hours after the last dose of MS was given to the tolerant monkey.

Three milliliters of serum from a donor animal was injected intraperitoneally into each of 114 male Wistar rats whose mean weight was

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