consumption and Ca distribution of the muscle in high K medium. These results seem to support the assumption (2) that Ca ions enter into the intracellular space through the membrane of the smooth muscle depolarized by high K solution, and participate in increase of oxygen consumption, while Ca ions entering into the "tightly bound fraction" play some roles in developing muscle tension.

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THE EFFECTS OF PARACHLOROPHENYLALANINE ON NON-TOLERANT RATS AND OF CHOLINERGIC BLOCKING DRUGS ON TOLERANT RATS TO MORPHINE

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Received for publication June 6, 1970

The small amounts of morphine raise while the large amounts of morphine lower the body temperature of non-tolerant rat. On the other hand, morphine raise and its antagonist, nalmophine, lower the body temperature of tolerant rat (1-3).

Feldberg and Meyers presented the new concepts that norepinephrine and 5-hydroxytryptamine (5-HT) in hypothalamus regulate the body temperature (4-6).

Collier (7) and Sharpless and Jaffe (8) are making efforts to explain the phenomena of tolerance, dependence and abstinence syndrome by pharmacological denervated supersensitivity (disuse supersensitivity). Their theories are based on the existence of interaction between morphine and humoral transmitters in the brain.

These evidences mentioned above stimulate us to study the relationship between morphine and humoral transmitters. In the present investigation, the changes of body temperature and spontaneous activity of rat were chosen as the indicators resulting from the interaction between morphine and humoral transmitters.

Thermistor probe was inserted 6 cm into rectum of the male rat of Donryu strain and taped on the tail. Rat can move relatively freely during measuring body temperature without touching and knowing what is being done. Under these conditions, the rectal temperature of the rat was measured outside of the chamber, the temperature of which is maintained at 20±0.5°C and the relative humidity at 70±10%.

The body temperature of non-tolerant rat was lowered significantly by the subcutaneous (s.c.) administration of 50 mg/kg of morphine-HCl, while this lowering action was completely prevented by the pretreatment with parachlorophenylalanine (300 mg/kg, i.p.) 72 hours before morphine injection. Parachlorophenylalanine is known to lower 5-HT level in brain to about 10% of normal without decreasing norepinephrine level in brain significantly (9) and has no effect on the body temperature of rats by its administration.

**FIG. 1-A.** The effects of morphine-HCl (20 mg/kg, s.c.) and scopolamine hydrobromide (5 mg/kg, s.c.) on the body temperature of tolerant rats. From one hour before injection to three hours after injection, the rectal temperature was measured every 15 minutes, while from three to five hours after injection, every 30 minutes. Measurements of body temperature started at about 9.30 a.m. Each point represents the average from six rats and S.E. for each point is below one per cent of the observed values. (---) ; Morphine was given at arrow mark on the left side. (----) ; Scopolamine was given at arrow mark on the left side. (-----) ; Scopolamine (at arrow mark on the left side) was given 15 minutes before morphine injection (at arrow mark on the right side).

**FIG. 1-B.** Effects of cholinergic blocking drugs on the spontaneous activity of tolerant rats induced by morphine-HCl (20 mg/kg, s.c.). Measurement of spontaneous activity started at about 9.30 a.m. Spontaneous activity of tolerant rats given only morphine-HCl (20 mg/kg, s.c.) was measured as control (=100). Then, next day, cholinergic blocking drugs were given subcutaneously to these tolerant rats 10 minutes before morphine injection and immediately after morphine injection, spontaneous activity was measured. The results are given as average±S.E. from at least five rats.

Tolerant rats were made by the repeated s.c. injection of 20 mg/kg of morphine-HCl twice a day (at 9.00 and 17.00) for more than a week and were maintained by the same way. The s.c. administration of 20 mg/kg of morphine-HCl to such tolerant rat raised the body temperature. The rise of body temperature by morphine was completely blocked by the s.c. administration of scopolamine hydrobromide (5 mg/kg) 15 minutes...
before morphine injection (Fig. 1-A) and was partially inhibited by 0.5 mg/kg of scopolamine hydrobromide. The lowering effect of naloxone (5 mg/kg, s.c.) on the rectal temperature of tolerant rat was not prevented by neither parachlorophenylalanine, α-methyl tyrosine nor scopolamine. Only the change of environmental temperature from 20 to 30°C could prevent its lowering action by naloxone.

Spontaneous activities of two tolerant rats were measured in a transparent plastics cage (20 cm in width, 30 cm in length and 13 cm in height) by M/P 40Fc Motility Meter, Motron-Produkter, Stockholm, Sweden, at 20 ± 0.5°C.

The s.c. administration of 20 mg/kg of morphine-HCl to tolerant rats increased their spontaneous activities. Such increase of spontaneous activity was partially inhibited by cholinergic blocking drugs (Fig. 1-B). The inhibitory action of methscopolamine, which seems not to penetrate blood-brain barrier and is thought to have greater blocking action to peripheral cholinergic receptor than scopolamine (10), indicate that increase of spontaneous activity is not only mediated by peripheral cholinergic mechanism but also by central cholinergic mechanism. Moreover, scopolamine has greater inhibitory action on spontaneous activity than atropine, which is thought to have less blocking action than scopolamine on central muscarinic neuron (11, 12). This result also suggests that increase of spontaneous activity is partially mediated by central cholinergic mechanism.

The s.c. administration of 20 mg/kg of morphine-HCl to tolerant rat increase the body weight of rat to around 6 to 9% in four hours after morphine injection. This increase of body weight was also partly blocked by these cholinergic blocking drugs.

Above data suggest us the possible participations of 5-HT and cholinergic mechanism to some signs of the rat induced by the administration of morphine.

Details on the relationships between morphine and 5-HT and cholinergic mechanism are now under investigation and to be published in the near feature.

Acknowledgement: The authors wish to thank Prof. E. Hosoya for his encouragement and interest.

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