SITES OF ANALGESIC ACTION OF CIRAMADOL

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Ciramadol, a benzylamine derivative [1-cis-2-(dimethyl-amino-3'-hydroxy-phenyl-methyl)cyclohexanol], has been reported to be an agonist-antagonist analgesic drug in animals (1) and humans (2). However, the profiles and sites of analgesic action of the drug in animals have not yet been shown.

Satoh et al. (3) have demonstrated that the flexor reflex induced by intra-arterial injection of bradykinin in the rat was an objective and sensitive index for evaluating various analgesic drugs. Pentazocine and cyclazocine were shown to produce analgesic effects in the bradykinin-induced flexor reflex test (4), though the drugs did not elicit antinociceptive effects without marked impairment in the tail-pinch, tail-flick and hot-plate methods.

In the present experiments, antinociceptive effects of ciramadol were evaluated using three analgesic tests: the tail-pinch, hot-plate and bradykinin-induced flexor reflex tests. Further, in order to search for the sites of analgesic action of ciramadol in rats, the drug was not only administered systemically, but also applied locally to the lumbar subarachnoid space and nuclei reticularis paragigantocellularis and gigantocellularis (NRPG-NRGC), which are sensitive sites to morphine in the production of analgesia (5, 6).

The tail-pinch tests were performed in male dd-K mice (14–17 g) and Sprague-Dawley rats (180–200 g) according to the method described by Takagi et al. (7) and Akaike et al. (8), respectively.

The hot-plate tests were done using male

dd-K mice (22–26 g). The latencies of paw-licking reactions of mice placed on a hot plate at $55 \pm 0.1^\circ\text{C}$ were determined. Each mouse was tested only once at 30 min after subcutaneous administration of ciramadol or physiological saline.

The bradykinin-induced flexor reflex test was carried out on conscious male Sprague-Dawley rats (160–260 g) according to the methods described by Satoh et al. (3). Bradykinin (2–4 μg in 0.2 ml physiological saline) was injected into the right common iliac artery at intervals of 10 or 20 min through a cannula implanted retrogradely into the left common iliac artery from the left femoral artery. Bradykinin-induced flexor reflexes of the right hind limb were recorded on a kymograph. When, after administration of ciramadol, the magnitude of the reflexes was less than 25% of the smallest value among control trials, the effect was regarded as analgesic. The ED₅₀ values and 95% confidence limits were determined according to the method of Litchfield and Wilcoxon (9).

For injection of ciramadol into the lumbar subarachnoid space (intrathecal injection), a polyethylene tube (13 cm length and 0.5 mm o.d.) was inserted through a puncture in the atlanto-occipital membrane so that the tip rested on the dorsal aspect of the rostral margin of the lumbar enlargement and fixed to the skull with dental cement. Such an operation was done under ether anesthesia immediately before cannulation for bradykinin injection. Ciramadol was dissolved in physiological saline and injected intrathecally in a volume of 10 μl .

Microinjection of ciramadol into the NRPG-NRGC was done according to the method described by Akaike et al. (8). Ciramadol dissolved in physiological saline at various concentrations was injected into the NRPG-NRGC in a volume of 0.5 μ l from an injection cannula introduced through a guide cannula which had been unilaterally implanted under pentobarbital anesthesia one week before experiments so that the tip of the guide cannula was located 4 mm above the NRPG-NRGC. At the end of each experiment, the microinjection site was histologically verified to be in the NRPG-NRGC. Each rat was given only one dose of ciramadol, subcutaneously, intrathecally or into the NRPG-NRGC.

Subcutaneous administration of ciramadol did not produce any detectable effect with the tail-pinch tests in mice at doses of 5, 10, 20, 50 and 100 mg/kg (10 mice per each dose) and in rats at doses of 5 and 50 mg/kg (3 and 4 rats, respectively). Furthermore, ciramadol in subcutaneous doses of 5, 10 and 50 mg/kg did not significantly influence latencies of paw-licking and jumping reactions (10 mice per each dose) in the hot-plate test.

On the other hand, the bradykinin-induced flexor reflexes of rats were suppressed by subcutaneous ciramadol in a dose-dependent manner at doses of 0.5, 1, 2 and 4 mg/kg. Such an analgesic effect appeared within 15 min after administration of the drug and lasted for 70 min or more. The ED₅₀ value (95% confidence limits) was 1.18 (0.63–2.23) mg/kg. The suppressive effect of ciramadol was antagonized by naloxone (0.1 mg/kg, i.v.) given 30 min after ciramadol administration (Fig. 1).

Intrathecal injection of ciramadol in doses of 0.1, 0.5 and 1 μ g/rat produced a dose-dependent analgesic effect in the bradykinin-induced flexor reflex test. The ED₅₀ value was 0.32 (0.15–0.63) μ g/rat. Such an effect appeared within 5 min after the injection and

Fig. 1. A suppressive effect of ciramadol subcutaneously administered on bradykinin-induced flexor reflexes in a rat and reversal of the inhibition by naloxone. Minus signs indicate time before ciramadol administration.

lasted for 60 min or more in 5 out of 10 rats in which the analgesic effects were produced by intrathecal ciramadol. Ciramadol microinjected into the NRPG-NRGC produced a dose-dependent suppression of bradykinin-induced flexor reflexes in doses of 1, 2 and 5 μ g/rat. The ED₅₀ value was 1.5 (0.84–2.7) μ g/rat. The suppressive effects appeared within 5 min after the microinjections and recovered within 60 min in 7 out of 10 rats in which the effects were produced by microinjections of the drug into the NRPG-NRGC. Longer lasting analgesic effects observed in four rats injected intrathecally and in three rats microinjected into the NRPG-NRGC were reversed by naloxone (0.1 mg/kg, i.v.) given 80 or 100 min after ciramadol applications (Fig. 2).

Ciramadol did not elicit any motor impairment at the doses used in the bradykinin-induced flexor reflex tests, suggesting that the suppressive effects of ciramadol on the bradykinin-induced flexor reflexes were hardly due to inhibitory effects of the drug on

Fig. 2. Examples of inhibitory effects of ciramadol microinjected into the NRPG-NRGC (A) and applied into the lumbar subarachnoid space (C) on the bradykinin-induced flexor reflexes in rats and antagonism to the inhibition by naloxone. (B) shows microinjection sites in the NRPG-NRGC of rats. Filled marks, but not open marks, indicate the microinjection sites where ciramadol (2 µg/rat) produced analgesia. A filled circle marked with a small arrow shows a microinjection site in an experiment from which the data represented in (A) were obtained. Minus signs indicate time before ciramadol application.

the motor systems.

The present experiments demonstrated that an analgesic effect of ciramadol administered systemically was detected with the bradykinin-induced flexor reflex test, but not with the tail-pinch and hot-plate tests. The analgesic effect was antagonized by naloxone (0.1 mg/kg, i.v.), indicating that ciramadol acted through specific opioid receptors. Such a pharmacological profile of ciramadol is similar to that of pentazocine. Judging from the ED₅₀ values of ciramadol and pentazocine (4) in the bradykinin-induced flexor reflex test (1.18 and 1.45 mg/kg, s.c., respectively), the analgesic potencies of both drugs are approx. equivalent. Such results fairly correspond to clinical data that the analgesic potency of ciramadol to post-operative pain was nearly equal to that of

pentazocine (2).

Micro-applications of ciramadol to the lumbar subarachnoid space and NRPG-NRGC produced dose-dependent analgesic effects in the bradykinin-induced flexor reflex test. The potency in the former case was 4.7 times more than that in the latter case, suggesting that the action of ciramadol on the spinal cord more greatly contributes to the production of analgesia by the drug than that on the supraspinal structures like NRPG-NRGC. On the other hand, in the tail-pinch test, the analgesic potency of morphine applied to the lumbar subarachnoid space has been shown to be 13 times less than that of the drug microinjected into the NRPG-NRGC (6). Moreover, the analgesic effect of pentazocine in the bradykinin-induced flexor reflex test was much less in spinal rats than in intact rats (4). These observations suggest the main site of analgesic action of ciramadol is different from that of morphine and pentazocine; the former is at the spinal level, but the latter is in the supraspinal structures such as the NRPG-NRGC.

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