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Dexamethasone Reverses Adrenalectomy Enhancement of Footshock Induced Analgesia in Mice

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MAREK, P., I. PANOCKA AND B. SADOWSKI. *Dexamethasone reverses adrenalectomy enhancement of footshock induced analgesia in mice.* PHARMACOL BIOCHEM BEHAV **18**(2) 167–169, 1983.—Footshock induced analgesia in mice was significantly enhanced by adrenalectomy. This effect of pituitary-adrenal axis disruption was reversed by dexamethasone. Involvement of pituitary factor(s) in producing this form of stress induced analgesia in mice is suggested.

Footshock Analgesia Adrenalectomy Dexamethasone

A WIDE variety of stressful manipulations produce pronounced pain threshold elevation in the rat and mouse. This analgesia is not a unique phenomenon, but depends on several physiological mechanisms. Watkins and Mayer [23] distinguished, for the rat, four classes of environmental analgesia each of which may be qualified as opiate or nonopiate and as neural or hormonal in nature. Such procedures as prolonged four paw shock, brief front paw shock and conditioning to footshock were found to produce analgesia which appears to be mediated by endogenous opioids [23].

Since various stressful stimuli increase pituitary-adrenal axis activity [17,18], the question arises whether analgesia induced by these stimuli is hormonally mediated. Two types of opiate analgesia in the rat, one produced by immobilization and the other by prolonged footshock, were attenuated by hypophysectomy [23]. ACTH and β-endorphin were found to be released concomitantly by the pituitary following footshock stress [12] and this release was blocked by hypophysectomy [12] or by dexamethasone pretreatment [7]. However, the role of β-endorphin as a putative mediator of hormonal-opiate analgesia may be questioned due to observation that post-stress β-endorphin blood level is severalfold lower than capable of inducing analgesia when injected intravenously [21,22].

Several findings argue that the mechanism of environmental analgesia may differ between rats and mice. For such species distinction indicates different stress susceptibility with regard to analgesic response. Room temperature water swimming (20°C/3 min) which produces pronounced analgesia in mice [24] is incapable of inducing analgesia in rats in which cold water (2, 8, 15°C/3.5 min) is necessary [3]. It must be noted that room temperature water swimming analgesia in mice is opiate in nature [24] which further distinguishes it from the nonopiate cold water swimming analgesia observed in rats [2]. Also, contrary to the rat, both brief (30 sec) and prolonged (10 min) footshock analgesia in mice is blocked by naloxone [4].

The purpose of the present study was to determine whether the pituitary-adrenal axis is involved in producing footshock analgesia in mice. Adrenalectomy enhancement of this sort of analgesic response and its dexamethasone reversal is reported.

METHOD

Male laboratory mice weighing 45 g in average reared on a natural day-light cycle were used. Bilateral adrenalectomy in 36 mice and sham operation (incising and suturing the skin without removing the glands) in 30 mice were performed under ether anesthesia. Mice kept five to a cage at 25°C and fed ad lib were given 0.9% NaCl solution (adrenalectomized and sham operated) or tap-water (intact) to drink. Completeness of adrenalectomy was assessed through autopsy at the end of experiment.

Foot shock was used as an analgesia-producing stimulus. Each mouse was placed on a grid and shocked with 50 Hz sine waves at 0.5 mA rms on a 10 sec on/10 sec off schedule for 10 min. The animals' behaviors were carefully observed during the whole period of footshock. Feces and drops of urine were immediately removed to prevent shortcircuiting. Pain sensitivity was assayed just before and immediately after stress on a hot plate at 56°C. The criterion was a hind paw flick the latency of which was measured with a stop watch. Pre- and post-shock latencies in adrenalectomized (Adrex) and sham operated (Sham) mice were measured at the 8th post-operative day.

Seventeen sham operated, 18 adrenalectomized and 14 intact mice were injected intraperitoneally with 0.9% NaCl solution. Intraperitoneal injections of dexamethasone (Decadron Phosphate, Merck Sharp and Dohme) at a dose of 500 μg/kg were given to 13 sham operate, 18 adrenalectomized

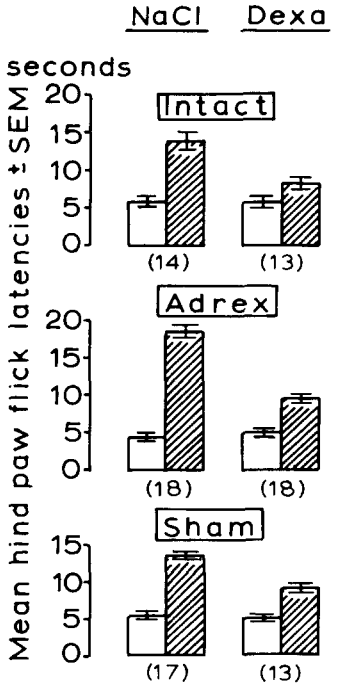

FIG 1. Pain threshold before (White columns) and after footshock (dashed columns) in intact (Intact), adrenalectomized (Adrex) and sham operated animals (Sham). Each group is labelled according to pretreatment with 0.9% NaCl (+NaCl) or dexamethasone (+Dexa). n=number of animals in group.

and 13 intact mice. All injections given at a volume of 0.1 ml/10 g body wt. were made 100 min before footshock. A comparable dexamethasone protocol is known to reverse opiate hypersensitivity in adrenalectomized mice [14].

Three-way split plot analysis of variance was used to evaluate statistical significance of the results. The animals were divided into groups according to the surgery (Intact, Sham and Adrex) and subgroups according to treatment (NaCl or dexamethasone). Pain threshold tests (pre-shock and post-shock) were taken as repeated measures. Individual comparisons were based on F ratios for simple effects.

RESULTS

The results are presented in Fig 1. Overall analysis of variance showed a significant effect of stress, $F(1,87)=174.44$, $p<0.001$, and treatment, $F(1,90)=32.11$, $p<0.001$ and a significant surgery × treatment × test interaction, $F(2,87)=3.70$, $p<0.05$. Pre-shock pain sensitivity differed neither between groups, $F(2,174)=0.75$, non significant (NS), nor between treatments, $F(2,174)=0.04$, NS. Pain threshold was significantly elevated after stress in all NaCl-treated groups, $F(2,174)=11.76$, $p<0.001$. This analgesia did not differ between intact and sham operated groups, $F(1,87)=0.00$, NS, and was significantly greater after adrenalectomy, $F(1,87)=10.07$, $p<0.001$.

Dexamethasone pretreatment significantly decreased the analgesic action of footshock, as compared to NaCl in all animal groups. The $F(1,87)$ values for the intact, sham operated and adrenalectomized groups were 7.03, 22.11 and 5.83, $p<0.05$, 0.001 and 0.05, respectively.

In intact mice dexamethasone reduced post-stress hot plate latencies to the pre-stress level, $F(1,87)=2.90$, NS, whereas in the two other groups the post-shock pain threshold were still higher compared to the pre-shock ones: $F(1,87)=12.11$, $p<0.001$ for adrenalectomized, and 5.50, $p<0.05$ for sham operated animals.

DISCUSSION

Several studies indicate pituitary involvement in environmentally-induced changes in pain sensitivity in the rat. Hypophysectomy attenuates immobilization [1], cold water swimming [3] and some forms of footshock [9,19] analgesia. Moreover, prolonged footshock analgesia was blocked by dexamethasone [15].

Only scanty data exsist on pituitary involvement in modulation of pain sensitivity in the mouse. Acupuncture produced analgesia was reported to be attenuated by hypophysectomy [20] and dexamethasone pretreatment [5]. Integrity of the pituitary was also found to be crucial in naloxone induced hyperalgesia [11].

In the present experiment adrenalectomy enhancement of footshock induced analgesia was reversed by dexamethasone pretreatment which also significantly attenuated analgesia in the intact and sham operated mice. This finding is in accordance with our previous results on swimming analgesia in mice [16] and is interesting since pain threshold elevation induced by both of these stress paradigms was shown to be completely blocked by naloxone [6,24]. The observation of a parallel dexamethasone and naloxone attenuation of footshock analgesia forms an interesting comparison with the rat in which only prolonged severe footshock (30 min/3 mA) analgesia was blocked by dexamethasone and naloxone [15]. Additionally, even high doses of naloxone (20 mg/kg) do not reverse effect of brief (up to 3 min) footshock [13, 15, 21] and cold water [3] in the rat. Contrary, in mice both brief footshock (30 sec) and room temperature swimming analgesia are completely blocked by naloxone (1 mg/kg and 0.1 mg/kg, respectively [4,24]. Such potent naloxone effects and the results of the present study indicate pituitary opioid involvement in producing footshock induced analgesia in the mouse.

In spite of these arguments the involvement of pituitary β-endorphin remains questionable. First, intravenous β-endorphin doses which produce analgesia in mice through much lower than in rats, are still unphysiologically high [21,22]. A possible resolution of this paradox was recently proposed [15]. According to this suggestion analgesic concentration of pituitary opioids in brain areas responsible for pain inhibition may be reached due to hypophyseal portal system or via ventricular system. The latter one is specially suggestive in mice since such humoral pathway was found to be crucial in morphine analgesia in this species [8]. Another argument against involvement of pituitary β-endorphin is lack of changes in baseline latencies which should be elevated in effect of adrenalectomy induced pituitary hypersecretion. It must be noted, however, that experiment was carried out on the 8th post-operative day so that physiological tolerance may have occured.

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