EFFECT OF PROSTAGLANDIN D₂ ON PAIN AND INFLAMMATION

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Various studies about the role of the prostaglandin (PG) E and F series in the pain and inflammatory reactions have been reported. However, there are few reports concerning these actions of PGD₂ (1), one of the arachidonic acid metabolites that is similar to PGE₂ and F_{2α}. In chicken urate crystal arthritis, PGD₂ was by far the prevailing PG in the exudates, reaching levels up to 10 times those of PGE₂ (2). In addition, Nugteren and Hazelhof reported that in many tissues, D prostaglandins were the main products of the endoperoxide breakdown (3). Therefore, in this paper, the involvement of PGD₂ in the development of hyperalgesia and the inflammatory reaction was also studied in comparison with PGE₁ and E₂.

Male ddY strain mice (20–22 g) and male Wistar strain rats (100–110 g) were used. PGs used were supplied by Ono Pharmaceutical Co., Ltd. To test the hyperalgesic action of PGD₂, the ACh- and bradykinin (BK)-writhing tests were used. PGs were administered i.p. 10 min before injection of ACh and BK. All drugs injected i.p. were dissolved in 0.2 ml of 0.9% NaCl solution. Various doses of ACh were injected i.p., and the ED₅₀ value of ACh that elicited writhing was determined. In the ACh- and BK-writhing tests, the number of mice showed writhing response was measured during a 10 min period after ACh and BK injection. In case of testing the central action of PGs on pain sensitivity, PGs dissolved in 0.9%

NaCl solution were injected intracerebro-ventricularly (i.c.v.) in a volume of 5 μ l. Five min after i.c.v. injection of PGs, pain sensitivity was measured in three ways: hot-plate, Haffner's and acetic acid-writhing tests. In order to investigate the role of PGD₂ in the inflammatory reaction, the paw edema test was used. Various doses of PGs dissolved in 0.1 ml of 0.9% saline were injected into the right hind paw of rats, and the edema was measured by a mercury displacement manometry for 3 hr after the injection. Furthermore, in order to test whether PGD₂ could potentiate the inflammatory action, paws were pretreated with PGs 10 min before subplantar injection of BK.

Table 1 shows the effects of PGD₂ (1 μ g/kg) pretreatment on ACh- and BK-writhing. The maximal doses of PGs, which did not cause writhing when given alone, were used. PGD₂ shifted the ED₅₀ value of ACh to a low dose similar to PGE₁ and PGE₂. Giving BK alone (1, 0.1 or 0.01 mg/kg i.p.), manifestation of writhing was 20, 10 or 0%, respectively. When BK was injected in combination with PGD₂, marked potentiation was observed at a BK dose of 0.01 mg/kg. In the cases of higher doses of BK, however, the number of writhings decreased rather than being the same as that obtained by BK alone. A similar effect was obtained by mixtures containing PGE₁ or PGE₂.

The change of pain threshold by i.c.v. injection of PGD₂ was investigated (Table 2).

Table 1. Effects of PGD₂, E₁ and E₂ on ACh- and BK-writhing in mice

Pretreatment drugs (i.p.)	Dose (μg/kg)	ED50 value of ACh (Confidence limit) (mg/kg) (a)	% of mice showing a writhing response BK (mg/kg) (i.p.) (b)		
			1	0.1	0.01
Saline		0.64 (0.46–0.94)	20	10	0
PGD ₂	1	0.08 (0.03–0.17)	0	30	40
PGE ₁	0.1	0.21 (0.06–0.44)	0	10	60
PGE ₂	1	0.11 (0.02–0.38)	0	10	40

Manifestation of the writhing response was observed during a 10 min period after ACh or BK injection. (a) None of these dose-response relationships differed significantly from linearity, and those from the experiments with PGs did not deviate significantly from parallelism with the saline-treated control. (b) Each group included 10 mice.

Table 2. Effects of i.c.v. injection of PGD₂, E₁ and E₂ on pain sensitivity in mice

	Dose (μg/kg)	Hot-plate test (sec) (a)	Haffner's test N/12 (b)	Acetic acid writhing test (c)
Normal control		5.0±0.36	0	18.7±2.66
Saline		5.5±0.82	0	15.6±2.18
PGD ₂	0.5	5.3±0.42	0.5	15.8±2.12
	5	6.4±0.61	1.0	15.4±2.01
	50	7.6±0.56*	2.0	12.6±1.26*
Normal control		4.5±0.58	0	26.6±2.47
Saline		5.8±0.89	0	20.9±3.78
PGE ₁	0.5	8.5±1.00	2.5	16.9±4.28
	5	8.3±0.63*	4.0	17.5±3.24
	50	9.8±1.51*	4.0	9.0±2.44*
PGE ₂	0.5	4.8±0.60	0	25.2±2.15
	5	5.0±0.58	0	36.0±4.90*
	50	3.6±0.38*	0	23.8±4.61

(a) Hot-plate test: Mice were placed on the hot-plate (55±0.5°C) and latencies of their first response (licking the feet, jumping or rapidly stamping the feet) were recorded. Each value showed the mean ±S.E. of 12 animals.

(b) Haffner's test: The pressure of 500 g on the tail root of the mouse was given, and the biting response was used as an index. Pain sensitivity was expressed as the ratio of the number of animals showing negative and false positive reaction (only false positive reaction was changed into 0.5)/number tested (n=12).

(c) Acetic acid-writhing test: Number of writhing responses occurring during a 10 min period after a lapse of the first 5 min after i.p. injection of 0.6% acetic acid (0.1 ml/10 g body weight) was measured. Each value showed the mean ±S.E. of 12 animals. *Significantly different from the saline control (P<0.05)

There was no significant difference between the saline control and the normal control (non-treated). Increase in doses of PGD₂ from 0.5 to 50 μg/kg decreased the pain sensitivity dose-dependently with each method, but PGD₂ had a less potent analgesic effect than PGE₁. Conversely,

PGE₂ caused an increase of pain sensitivity.

PGD₂ was injected into the hind paw of rats, and the potency for edema formation was compared with PGE₁ and PGE₂. PGD₂ (10, 100, 1000 ng) produced the paw swelling dose-dependently (data not shown). This inflammatory effect of PGD₂ was less

potent than PGE₁ and E₂ and lasted for a short time. Effects of the addition of various PGs on the paw swelling induced by BK (10 ng) were examined (data not shown). Each dose of PGs used had no greater effect than saline when given alone. However, pretreatment with PGD₂ (1 ng) caused potentiation of the BK paw swelling. One twentieth ng of PGE₁ had a similar potentiating effect, but PGE₂ (0.05 ng) gave no potentiation.

It was observed that PGD₂ injected into a peripheral site elicited hyperalgesia in the ACh- and BK-writhing tests, similarly to PGE₁ and E₂. When PGD₂ was injected into the cerebral ventricle, it showed an analgesic effect. Thus there was the different result that PGD₂ was hyperalgesic peripherally and analgesic centrally. The same effect was reported on BK (4, 5) and substance P (6, 7). Pretreatment with the low dose of PGD₂ (1 ng) resulted in potentiation of BK paw swelling. However, PGD₂ was about one-twentieth as potent as PGE₁. These results indicate that PGD₂ appears to possess hyperalgesic action in the peripheral pain reaction and performs the roles of chemical

mediator and potentiator in the inflammatory process.

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