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HYPOTHERMIC EFFECT OF SODIUM ACETYLSALICYLATE ON AFEBRILE MONKEYS

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1 Afebrile monkeys (*Macaca cyclopis*) receiving systemic (100-300 mg/kg, i.p.) or central (5-20 mg into the 3rd cerebral ventricle) administration of sodium acetylsalicylate showed a dose-dependent reduction in rectal temperature in a thermoneutral environment (25° C).

2 Administration of sodium acetylsalicylate (10 mg) into the 3rd cerebral ventricle produced a hypothermia with a temperature decrement of 1.0° C, while an intraperitoneal injection of 300 mg/kg was required for a temperature decrement of 0.9° C. The ratio between the total doses given by the two routes was 1 to 120.

3 Following the administration of sodium acetylsalicylate, a decline in rectal temperature was accompanied by a tail cutaneous vasodilatation.

4 The data suggest that sodium acetylsalicylate can lower the normal body temperature by activating heat loss or decreasing the normal (tonic) inhibition of the heat loss mechanism via the central nervous system.

Introduction

It is generally agreed that salicylates produce an antipyretic effect in animals (Chai, Lin, Chen & Wang, 1971; Cranston & Rawlins, 1971; Lin & Chai, 1972; Rawlins, 1973) and humans (Rosendorff & Cranston, 1968) made febrile with pyrogen. Previous studies have also indicated that both pyrogen and salicylate act in the same central thermoregulating structures (most probably the hypothalamus); pyrogen by inhibition of the heat loss mechanism and salicylates by release of such inhibition (Wit & Wang, 1968; Luff, Rawlins & Wright, 1971; Lin & Chai, 1972). Our recent results have also demonstrated that salicylates lower hyperthermia during external heat exposure by activating the heat loss mechanism (Lin & Chai, 1975).

In afebrile animals, however, the effect of salicylates on body temperature is less clear. No temperature change has been observed in afebrile human subjects (Rosendorff & Cranston, 1968) or rabbits (Cranston, Luff, Rawlins & Rosendorff, 1970). In contrast, rats receiving systemic injections of salicylates showed a severe reduction in normal body temperature when placed in a cold environment (Satinoff, 1972; Franscesconi & Mager, 1974). No work has been reported so far in monkeys. Furthermore, little is known as to whether the direct injection of salicylates into the cerebral ventricle will affect normal body temperature.

Thus, the present investigation was carried out with conscious afebrile monkeys in which the thermosteady state was first reached by adapting the animals to a thermoneutral environment (25° C). Sodium salicylate was subsequently administered by different routes and in various doses.

Methods

Fourteen monkeys (*Macaca cyclopis*) of either sex weighing 3.5 to 4.5 kg were used in the present study. Each animal was anaesthetized with sodium pentobarbitone, 30 mg/kg, intravenously. A 22-gauge cannula was surgically implanted into the 3rd cerebral ventricle of each animal with the aid of a stereotaxic instrument for drug administration at a later time. This technique has been described by Chai *et al.* (1971) and Hoo, Lin, Wei, Chai & Wang (1972). At the end of the experiments, the animals were killed and the heads were perfused with 10% formalin-saline. Location of the cannula was verified by gross inspection and histological section with thionin stain. Intraperitoneal

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injections were given by direct puncture of the abdominal wall.

Each animal was allowed at least one week to recover completely from the surgery before being subjected to experiments. Before drug administration, the animal was seated quietly in a primate chair at an ambient temperature of 25 ± 0.5° C until it maintained a steady body temperature for at least 1 hour. During the control period of observation, any animals showing a body temperature below 37.5° C or above 39.0° C were not used. Successive drug administrations were at least one week apart to eliminate any possible tolerance effect. The evening before the experiment, each animal was given water *ad libitum*, but food was withheld. The rectal temperature was monitored by means of a Tri-R-6 flexible thermistor probe and the subcutaneous temperature of the tail was monitored by a needle thermistor. All the recordings were made continuously on a Grass polygraph.

Sodium acetylsalicylate (0.5 to 2.0 g) was freshly dissolved in 10 ml of distilled water, adjusted to pH 7.0-7.2, to make 0.25 to 1.0 M solutions. A volume of 0.1 ml of this solution, containing 5 to 20 mg of sodium acetylsalicylate, was injected into the 3rd cerebral ventricle (intracerebroventricular; i.c.v.). Sodium chloride solutions of equal concentration were prepared as controls to be injected in similar volumes via the same route. For intraperitoneal injection, sodium acetylsalicylate was dissolved in a 0.15 M NaCl solution to make several concentrations ranging from 100 to 300 mg/ml.

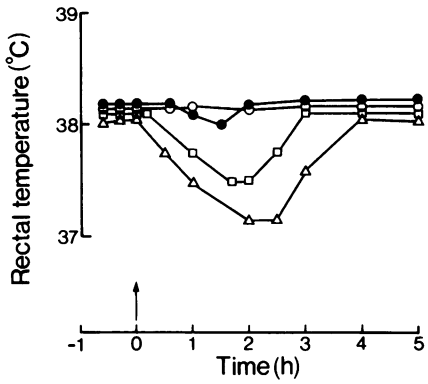

Figure 1 Change in rectal temperature of conscious, afebrile monkeys kept at an ambient temperature of 25° C for 5 h after intraperitoneal administration of sodium acetylsalicylate at arrow. Each point is the average temperature of 8 animals. Control intraperitoneal injection of saline (○); after intraperitoneal injection of sodium acetylsalicylate, 100 mg/kg (●), 200 mg/kg (□) and 300 mg/kg (△).

Results

At a thermoneutral environment (25° C), monkeys treated with sodium acetylsalicylate intraperitoneally or intracerebroventricularly, showed a significant fall in their rectal temperature when compared with animals treated with saline (Figures 1 & 2).

Intraperitoneal administration of sodium acetylsalicylate

Figure 1 shows that sodium acetylsalicylate (100-300 mg/kg, i.p.) induced a dose-dependent hypothermic effect in eight afebrile animals. Each animal served as its own control. With a dose of 100 mg/kg, the rectal temperature began to decrease 35 ± 4.8 min after injection and returned to its control level at 120 ± 12.6 minutes. The maximum decrement in rectal temperature was 0.2 ± 0.12° C (38.3-38.1° C) which was not significant ($P > 0.2$). When the dose was increased to 200 mg/kg, the time to the onset of hypothermia was reduced (12 ± 3.1 min), the maximum decrement in rectal temperature was augmented (0.6 ± 0.09° C; 38.2-37.5° C; $P < 0.05$), and the recovery time was prolonged (180 ± 20.5 minutes). After the injection of 300 mg/kg, the maximum decrement in rectal temperature was further augmented (0.9 ± 0.11° C; 38.1-37.2° C; $P < 0.01$) with a shorter latency of onset (3.5 min) and a longer period of recovery (240 ± 14.5 minutes).

Intracerebroventricular administration of sodium acetylsalicylate

Similarly, Figure 2 shows that injection of sodium acetylsalicylate into the 3rd cerebral ventricle induced a dose-dependent hypothermia in six different afebrile animals. Again, each animal served as its own control. An intracerebroventricular dose of 5 mg of sodium acetylsalicylate produced a slight fall of 0.5 ± 0.12° C (38.3-37.8° C; $P < 0.05$) in rectal temperature. The time to onset of hypothermia was 30 ± 4.2 min and the recovery time was 160 ± 11.6 minutes. When the dose was increased to 10 mg, the time to onset of hypothermia became shorter (4 ± 2.1 min), the maximum decrement in rectal temperature greater (1.0 ± 0.22° C; 38.3-37.3° C; $P < 0.05$), and the recovery time longer (240 ± 16.2 minutes). This is comparable to the hypothermic effect obtained by

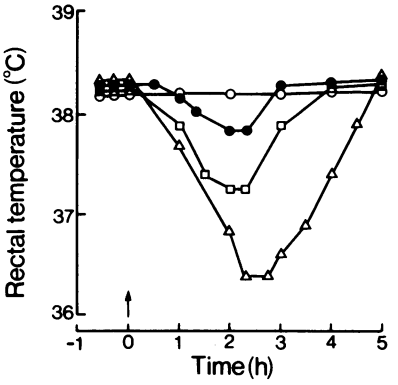

Figure 2 Change in rectal temperature of conscious, afebrile monkeys kept at an ambient temperature of 25° C for 5 h after injection of sodium acetylsalicylate into the 3rd cerebral ventricle at arrow. Each point is the average temperature of 6 animals. Control injection of saline (○); after sodium acetylsalicylate, 5 mg (●), 10 mg (□) and 20 mg (△).

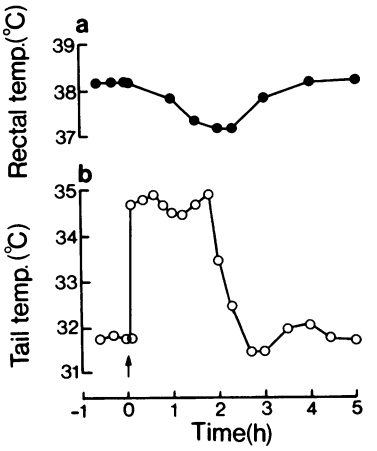

Figure 3 The general pattern of temperature changes of monkeys receiving 10 mg of sodium acetylsalicylate via 3rd cerebral ventricle at arrow. Each point is the average temperature of 6 animals. (a) rectal temperature; (b) tail cutaneous temperature.

the intraperitoneal administration of 300 mg/kg. With a dose of 20 mg of sodium acetylsalicylate, the maximum decrement in rectal temperature was further augmented to 2.0 ± 0.36° C (38.37-36.37° C; $P < 0.01$). The rectal temperature fell almost immediately after injection and returned to the control level much later (300 ± 21.4 minutes).

Figure 3 shows the general pattern of rectal and tail cutaneous temperature changes in response to intracerebroventricular acetylsalicylate. It was found that, concomitant with the onset of the falling rectal temperature, the tail cutaneous temperature increased greatly and rapidly. The tail cutaneous temperature remained at a high level (2-3° C above control) during the decreasing phase of rectal temperature, but then fell rapidly 10-15 min before the hypothermia reached its maximum.

Discussion

The present investigation was carried out in normothermic afebrile monkeys. The afebrile state was carefully checked before each experiment and it was found that, during the control period of observation, the mean rectal temperature of these monkeys was 38.2° C (ranging between 38.1 and 38.4° C). This coincides with the earlier reports of other investigators (Eyre & Kennedy, 1907; Funkhouser, Higgins, Adams & Snow, 1967; Nakayama, Hori, Nagasaka, Tokura & Tadaki, 1971).

Findings from the present experiments are that afebrile monkeys receiving sodium acetylsalicylate (100-300 mg/kg, i.p.) produce a dose-dependent decline in rectal temperature (−0.2 to −0.9° C) when placed in a thermoneutral environment (25° C). These results are consistent with recent reports of other investigators who demonstrated, in addition, that rats treated with salicylates (30-600 mg/kg, i.p.) displayed a more severe reduction in rectal temperature when placed in a 4-5° C environment (Satinoff, 1972; Franscesconi & Mager, 1974). The present results also have shown that the injection of sodium acetylsalicylate (10 mg) into the 3rd cerebral ventricle produced the same hypothermic effect as an intraperitoneal dose of 300 mg/kg (or 1200 mg for a monkey weighing 4 kg). The ratio between these two doses is 1 to 120. This suggests a central site of action. Furthermore, the present study has demonstrated that a decline in rectal temperature, following the administration of sodium acetylsalicylate, was accompanied by a persistent cutaneous vasodilatation. Therefore, these findings suggest that sodium acetylsalicylate lowers the normal body temperature by activating heat loss or decreasing the normal (tonic) inhibition of the heat loss mechanism in the central nervous system.

Similarly, it has been shown that the antipyretic effect of sodium acetylsalicylate is of central origin (Chai *et al.*, 1971). An

intracerebroventricular (4 mg) or an intravenous (100 mg/kg) dose of the drug produced a marked antipyresis in monkeys with fever induced by leukocytic pyrogen. However, in afebrile monkeys of the present study, an intraventricular dose of 10 mg or an intraperitoneal dose of 200 mg/kg was required to achieve a marked decline in rectal temperature. Thus, it seems that the febrile state is more sensitive to the administration of sodium acetylsalicylate. Indeed, it has been observed that, following an intravenous administration of [^{14}C]-acetylsalicylate, the concentration of the drug in the brain of febrile rabbits is significantly higher than that in afebrile animals (Chai, Lin, Hoo & Wang, unpublished data). It is not impossible that the elevated body temperature with high metabolic rate increases the entrance of sodium acetylsalicylate into the brain and thus enhances the heat dissipating mechanism.

On the other hand, the present results seem at variance with those reports that have demonstrated that salicylates have little or no effect on body temperature of afebrile rabbits (Cranston *et al.*, 1970) and humans (Rosendorff & Cranston, 1968). Cranston *et al.* (1970) have shown that salicylates (60 mg/kg i.v. or 1.2 mg i.c.v.) had no significant effect on rectal temperature. Similarly, in human volunteers, salicylates (2 g or 30 mg/kg, i.v.) also had no effect on normal body temperature (Rosendorff & Cranston, 1968). In fact, the present study has clearly demonstrated that sodium acetylsalicylate in low doses (i.e. 100 mg/kg, i.p. or less than 5 mg, i.c.v.) failed to induce hypothermia. Therefore, it seems justifiable to explain the difference between these two sets of results as a difference in the dosage of salicylates being used.

In unanaesthetized monkeys, Myers & Yaksh (1971) found that perfusion of the cerebral ventricles with artificial c.s.f. caused hypothermia when the solution contained 24-48 mM excess calcium ion and hypothermia when 11-34 mM excess sodium ion was perfused. In the present study, a single dose of 0.1 ml containing 0.25-1.0 M sodium acetylsalicylate was administered into the 3rd ventricle. It is unlikely that the sodium or calcium concentrations in the c.s.f. were sufficiently disturbed by our injections to produce hypothermia.

More recently, Feldberg, Gupta, Milton & Wendlandt (1973) have demonstrated that the prostaglandin-like activity of c.s.f. increased greatly during bacterial pyrogen fever, and decreased when the temperature was brought down by salicylates. In addition, Franscesconi & Mager (1974) have shown that the brain levels of tryptophan and hypothalamic 5-hydroxytryptamine were elevated in rats with salicylate-induced hypothermia. Whether these substances play any role in the salicylate-induced hypothermia in afebrile animals remains to be determined.

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