Effect of Heparin Na on the Action of Various Bronchoconstrictors in Guinea Pig Tracheal Strips

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ISHIHARA, Y. and KITAMURA, S. Effect of Heparin Na on the Action of Various Bronchoconstrictors in Guinea Pig Tracheal Strips. Tohoku J. exp. Med., 1981, 133 (1), 61-65 — The effect of heparin Na on the actions of various bronchoconstrictors was examined. Guinea pig tracheal strips were suspended in bioassay glass chambers and superfused with Krebs-Henseleit solution, pH 7.4, at 37°C saturated with oxygen and carbon dioxide gas (95:5, v/v). Contraction of the tissue was detected by an isotonic transducer and displayed on a polyrecorder. Heparin Na attenuated histamine-, prostaglandin F2α-, acetylcholine-, serotonin-induced contractile responses dose-dependently, and shifted the dose-response curves of histamine, prostaglandin F2α, acetylcholine, serotonin and bradykinin downward. — heparin Na; histamine; serotonin; PGF2α; acetylcholine

The mast cell granule releases histamine, slow-reacting substance of anaphylaxis (SRS-A) (Kitamura et al. 1978) and prostaglandin F2α upon immunologic challenge to the mast cell. However, the exact fate of heparin, which is also contained in the mast cell granule, during this release phenomenon is unknown. Heparin inhibits blood coagulation in vivo and in vitro.

Heparin was introduced into clinical medicine 40 years ago and has been used for the treatment of myocardial infarction, cerebral thrombosis, venous thrombosis, and pulmonary thrombosis, and also for animal experiments in vivo. Present investigation was conducted to examine the effect of heparin Na on the action of various bronchoconstrictors in guinea pig tracheal strips.

MATERIALS AND METHODS

Male Hartley strain guinea pigs, weighing 250-300 g, were sacrificed. Their trachea was removed, cut spirally 1.0-1.5 mm in width and 3.0-4.0 cm in length, and one end of the tracheal strips was attached to the notch at the bottom of the bioassay glass chamber and the other end to the lever of this system. The tracheal strips were superfused at a rate of 20 ml/min with the Krebs-Henseleit solution (pH 7.4) saturated with oxygen and carbon dioxide gas (95:5, v/v) at 37°C. A load of 0.5 g was applied to the strips and

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allowed to equilibrate for 1 hr. Drugs were infused by a low speed infusion pump (Harvard Apparatus, USA) at a rate of 0.97 ml/min. Contraction of guinea pig tracheal strips was detected by an isotonic transducer (ME Commercial, Tokyo) and displayed on a polyrecorder. The grade of contraction was expressed as the contraction index (C.I.) that is the area (cm²) enclosed by the response curve and the base line measured with a planimeter. Upward deflection from the base line, i.e. contraction, is shown with a plus sign. For the comparison of contraction indices Student’s t-test was used to establish significance.

Drugs used were acetylcholine hydrochloride (Ach, Daiichi Pharmaceut. Co., Tokyo), histamine dihydrochloride (Hist, Takara-Kohsan, Tokyo), serotonin creatinine sulfate (5HT, Daiichi Chemical Co., Tokyo), bradykinin (Brady, Protein Research Foundation, Osaka), prostaglandin F₂α (PGF₂α, Ono Pharmaceut. Co., Osaka), and heparin Na (Shimizu Seiyaku Co., Shimizu).

RESULTS

Fig. 1 shows the effect of different doses of heparin Na on contractile responses induced by Hist (250 ng/ml) and PGF₂α (250 ng/ml) in guinea pig tracheal strips. C.I. is on the ordinate and the concentration of heparin Na (U/ml) on the abscissa. It is obvious from this figure that heparin Na attenuated Hist- and PGF₂α-induced contractile responses dose-dependently.

In the same way, Fig. 2 shows the effect of different doses of heparin Na on contractile responses induced by Ach (100 ng/ml) and 5HT (250 ng/ml) in guinea pig tracheal strips. It is obvious from this figure also that heparin Na attenuated Ach- and 5HT-induced contractile responses dose-dependently.

The effect of continuous superfusion of heparin Na (1 U/ml) on the dose-response curves of Hist (left side) and PGF₂α (right side) is shown in Fig. 3. The

![Graph](image-url)

Fig. 1. Effect of heparin Na on histamine- and PGF₂α-induced contractile responses in guinea pig tracheal strips.
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solid circles represent the dose-response curves of Hist and PGF2α with continuous superfusion of solutions without heparin Na, while the open circles represent those with heparin Na. It is also obvious from this figure that dose-response curves of

Fig. 2. Effect of heparin Na on acetylcholine- and serotonin-induced contractile responses in guinea pig tracheal strips.

Fig. 3. Effect of heparin Na on dose-response curves of histamine and PGF2α in guinea pig tracheal strips. •—•, without heparin Na; ○—○, with heparin Na.
Hist and PGF$_2$α moved downward with continuous superfusion of heparin Na.

Fig. 4 shows the effect of heparin Na (1 U/ml) on the dose-response curves of Ach (left side), 5HT (middle), and Brady (right side). This figure also indicates that the dose-response curves of Ach, 5HT, and Brady moved downward with continuous superfusion of heparin Na.

**DISCUSSION**

The physiological function of heparin, if any, is the subject of speculation. There is suggestive evidence that it acts as a natural anticoagulant to aid in maintaining the blood in a fluid state, as was postulated by Howell and Holt (1918) many years ago. However, the failure of most investigators to find heparin in normal blood argues against this possibility.

In the present experiments, it was elucidated that heparin Na attenuated the contractile responses of guinea pig tracheal strips induced by Hist, PGF$_2$α, Ach, 5HT, and Brady, although its precise mechanism is still unknown. It is well known that Hist, SRS-A (Kaliner and Austen 1973), and prostaglandins (Piper and Vane 1971) are released from lung mast cells upon immunologic challenge, and it has also been suggested that heparin is released from lung mast cells upon immunologic challenge (Yurt et al. 1977). However, in the latter case, the role of heparin remains to be elucidated. Although the present investigation may suggest that heparin attenuates the harmful effect of bronchoconstrictive chemical mediators released...
into the circulating blood, whether or not the amount of heparin released upon immunologic challenge is sufficient to do this is still to be determined. It might, anyhow, be possible to use heparin in patients with status asthmaticus in place of glucocorticoid or methylxanthines.

Heparin is not a single substance and the potency of commercial preparations ranges from 140 to 190 units/mg. So, we plan to do similar experiments using various kinds of heparin from different companies in near future, and were also intending to extend this experiment to an in vivo study.

References