Effect of Adrenalectomy on Healing of Indomethacin-Induced Gastric Erosions in Rats

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We studied the effects of adrenalectomy and replacement therapy on healing of gastric erosions in adult Sprague-Dawley rats developed 4 h after subcutaneous injection of indomethacin in a dose of 25 or 35 mg/kg. Adrenalectomy was performed 1 week before or 4 h after indomethacin administration. Healing was evaluated by changes in the area of erosions over 24 h after indomethacin administration at fixed time intervals. Plasma corticosterone was measured. Adrenalectomy (irrespective of the time of intervention) decelerated healing of gastric injuries. Injection of corticosterone in a physiological dose of 4 mg/kg at the initial stage of healing (4 h after indomethacin administration) improved healing of erosions.

Key Words: *gastric mucosa; erosions; indomethacin; glucocorticoids; healing*

Glucocorticoids in pharmacological doses promote the development and decelerate healing of erosions and ulcers in the gastric mucosa [4,9-11]. It was hypothesized that glucocorticoids synthesized during stress produce an ulcerogenic effect. Our studies argued against this concept. We showed that endogenous glucocorticoids secreted during activation of the hypothalamic-pituitary-adrenocortical system possess gastroprotective properties [2,3,5-8].

Here we studied the effects of endogenous glucocorticoids on healing of gastric injury. The role of glucocorticoid deficiency and replacement therapy with corticosterone in healing of indomethacin-induced gastric erosions was evaluated.

MATERIALS AND METHODS

Experiments were performed on adult Sprague-Dawley rats weighing 220-260 g. The animals were kept at 19-21°C under standard laboratory conditions (daytime 8.00-20.00). Indomethacin in a dose of 25 or 35 mg/kg (5 ml/kg, Chinoin) was injected subcutaneously to model hemorrhagic erosions of the gastric mucosa. Indomethacin was suspended in physiological saline containing 1 drop of Tween 60 (Theodor Schuchardt) and administered after 24-h starvation.

Glucocorticoid deficiency was induced by adrenalectomy under Nembutal or ether anesthesia. Adrenalectomy or sham operation (control) was performed 1 week before and 4 h after administration of indomethacin in doses of 25 (series I) and 35 mg/kg, respectively (series II).

Since the area of erosions reached the maximum 4 h after indomethacin administration, the study of healing was started at this term. In series II adrenalectomy was performed 4 h after injection of indomethacin. Experimental rats received replacement therapy with the natural glucocorticoid corticosterone (4 mg/kg in 1 ml/kg propylene glycol subcutaneously, Serva). Adrenalectomized and sham-operated rats receiving 1 ml/kg propylene glycol served as the control. After treatment the animals fed a standard diet. Healing was studied by measuring the area of erosions under a microscope. The area of erosions was compared 7, 12, and 24 (series I) or 12 and 24 h after indomethacin injection (series II).

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Plasma corticosterone level was measured spectrofluorometrically [1]. The blood was taken after decapitation or collected through a cannula introduced into the jugular vein 1 day before indomethacin administration under Nembutal anesthesia.

The differences between plasma corticosterone concentrations were estimated by *t* test or Welsh's alternate *t* test for dissimilar dispersions. The differences between areas of erosions were evaluated by Mann—Whitney test.

RESULTS

Adrenalectomy performed 1 week before indomethacin administration potentiated the development and decelerated healing of gastric erosions in rats (Fig. 1). Four hours after indomethacin administration erosions in adrenalectomized rats were much larger than in sham-operated animals. Significant differences in the area of erosions were observed throughout the observation period (Fig. 1). In sham-operated and adrenalectomized rats the area of erosions markedly decreased (healed) 7 and 12 h after indomethacin administration, respectively. Replacement therapy with corticosterone was given to evaluate whether the effect of adrenalectomy was determined by glucocorticoid deficiency. Indomethacin stimulated corticosterone production in sham-operated rats, while adrenalectomy abolished this effect (Fig. 2, *a*, *b*). Treatment of adrenalectomized rats with corticosterone in a dose of 4 mg/kg simulated the hormonal response observed in sham-operated animals after administration of indomethacin (Fig. 2, *a*). Blood hormone concentration in adrenalectomized rats remained high 4 h after corticosterone administration (Fig. 2, *a*, *b*), but decreased 8 and 20 h after treatment (12 and 24 h after injection of indomethacin, Fig. 2, *b*).

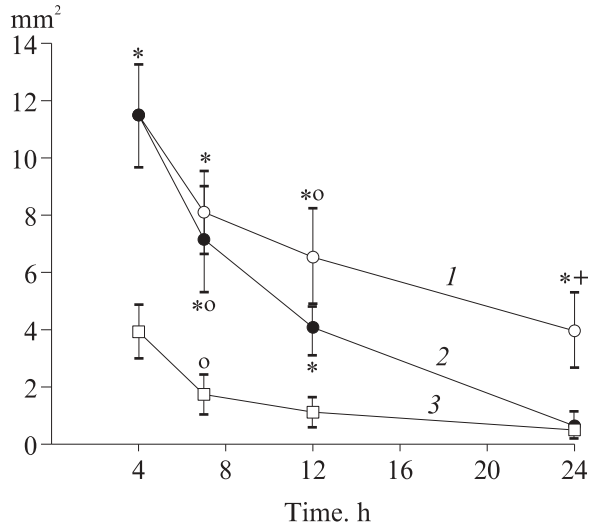

Fig. 1. Effect of adrenalectomy performed 1 week before indomethacin administration (25 mg/kg) and replacement therapy with corticosterone on healing of erosions in rat gastric mucosa (*n*=5-16). Here and in Fig. 3: abscissa, after indomethacin administration; ordinate, area of gastric injury. Adrenalectomized rats (*1*), adrenalectomized rats receiving replacement therapy (*2*), and control (*3*). $p<0.05$: *compared to the control; +differences between adrenalectomized rats receiving and not receiving replacement therapy; °first significant difference from the initial level.

Administration of corticosterone in a dose of 4 mg/kg accelerated healing of the gastric mucosa in adrenalectomized rats (Fig. 1). The area of gastric injury in these animals significantly decreased 7 h after indomethacin administration (similarly to sham-operated rats). Twenty-four hours after indomethacin

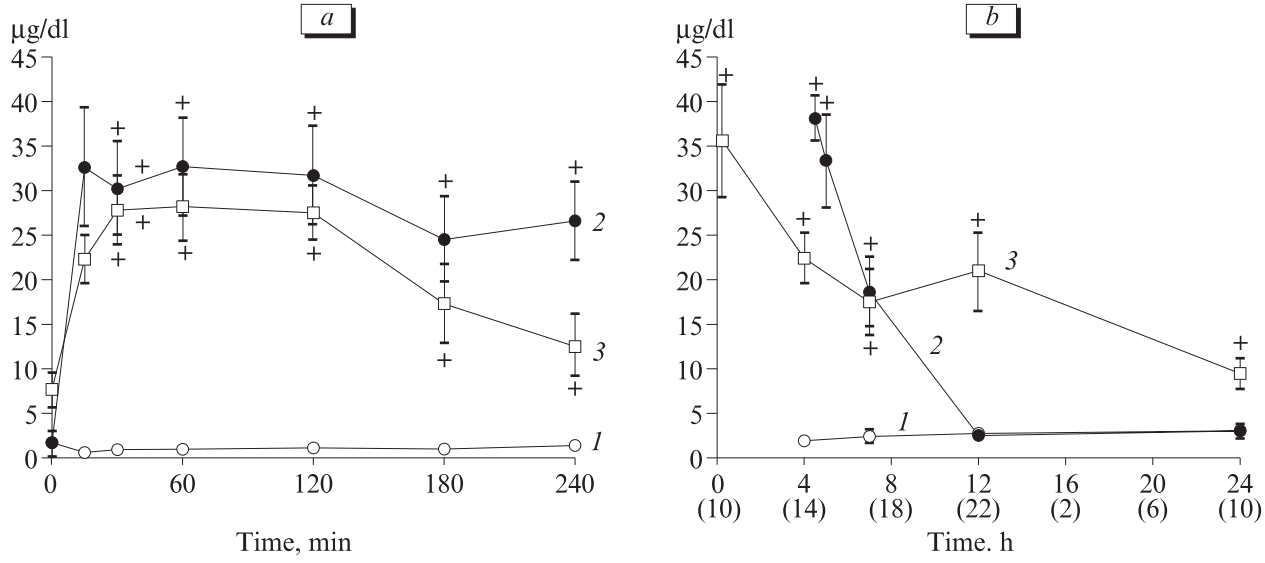

Fig. 2. Plasma corticosterone concentration in rats (*n*=5-16). Blood from cannulated (*a*) and decapitated animals (*b*). Abscissa: time after indomethacin administration; time of day is shown in brackets. Ordinate: plasma corticosterone concentration in rats. +$p<0.05$ compared to adrenalectomized rats not receiving hormone therapy.

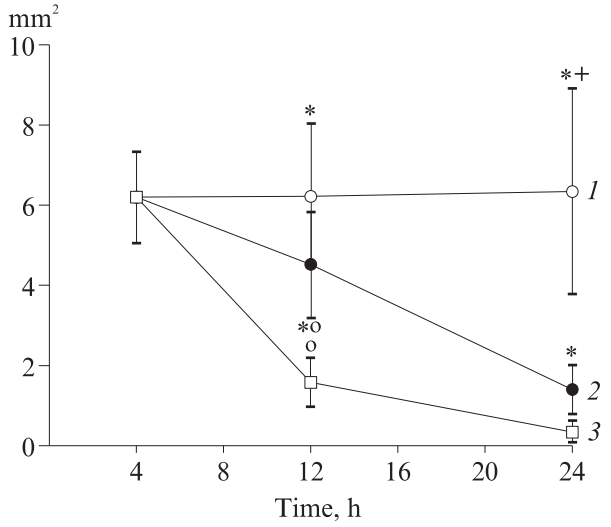

Fig. 3. Effect of adrenalectomy performed 4 h after indomethacin administration (35 mg/kg) and replacement therapy with corticosterone on healing of erosions in rat gastric mucosa.

administration the area of erosions in rats receiving replacement therapy decreased compared to that in adrenalectomized animals not treated with corticosterone and did not differ from that observed in the sham-operated rats. Adrenalectomy decelerated healing of erosions, which was related to glucocorticoid deficiency. Therefore, these hormones promote recovery of the gastric mucosa after injury.

Since in series I adrenalectomy was performed 1 week before indomethacin administration this procedure affected not only healing, but also the development of erosions. Therefore, healing was studied in rats differing in the initial degree of gastric injury. To avoid this disadvantage, in series II adrenalectomy was performed 4 h after indomethacin administration (*i.e.*, after development of erosions). Moreover, in series II the dose of indomethacin was increased (35 mg/kg) to stimulate the development of erosions. Adrenalectomy inhibited, while corticosterone in a dose of 4 mg/kg promoted healing of gastric injuries in adrenalectomized rats (Fig. 3). Sham-operated animals were characterized by good healing of erosions, whose area decreased 12 h after treatment. In adrenalectomized rats gastric injury did not heal even 24 h after treatment. It should be emphasized that the area of erosions in adrenalectomized rats receiving hormone replacement therapy markedly decreased 12 h after indomethacin administration. Twenty-four hours after injection of indomethacin the area of erosion in these rats was lower than in adrenalectomized animals.

It should be noted that the significant effect of corticosterone on healing of erosions in adrenalectomized rats developed only 20 h after treatment. In this period the hormone was not detected in the blood. This delayed effect of corticosterone was probably related to its effect on the synthesis of new compounds involved in healing of gastric injury.

Our results show that glucocorticoid deficiency induced before and after the development of gastric injury decelerates healing of erosions. Replacement therapy with corticosterone abolished the effect of insufficient hormone production. Therefore, glucocorticoids are involved in healing of gastric injury produced by indomethacin. These results and published data [6-8] indicate that patients with insufficient production of glucocorticoids are at high risk of ulcer disease produced by nonsteroid antiinflammatory preparations (*e.g.*, indomethacin).

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