Cortisol-induced migration of eosinophil leukocytes to lymphoid organs[1]

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Summary. Cortisol induces a migration of eosinophil leukocytes from the blood to several lymphoid organs, such as the spleen, lymph nodes and thymus, but not to other tissues. This migration explains the cortisol-induced blood eosinopenia, which is much more pronounced in intact than in splenectomized animals.

ACTH or cortisol administration is known to induce a dramatic decrease in the number of eosinophil leukocytes in the blood[2-5]. Several hypotheses have been proposed to explain the mechanism of cortisol-induced blood eosinopenia. Cortisol may induce a lysis of eosinophils in the blood stream[6], an inhibition of eosinopoiesis in the bone marrow[7-10] and/or a migration of these cells to other tissues[11]. Cortisol, however, does not destroy eosinophils in vitro[12]; therefore, the 1st possibility seems unlikely. The changes in eosinopoiesis cannot explain the kinetics of the cortisol-induced eosinopenia nor the kinetics of the increase in the number of blood eosinophils after the suppression of cortisol treatment[13]. Therefore, the most plausible hypothesis appears to be a cortisol-induced migration of eosinophils to tissues.

To verify the above hypothesis, we studied the effect of cortisol treatment or of adrenalectomy on the tissue eosinophils in various organs. We found that there is a cortisol-induced migration of eosinophils to the spleen, in addition to some other organs. Therefore, we also decided to test the effect of splenectomy on the cortisol-induced decrease in the number of blood eosinophils.

Material and methods. 95 female adult Sprague-Dawley rats were used in the present experiments. The following groups of animals were studied: cortisol-treated, splenectomized cortisol-treated, splenectomized untreated, adrenalectomized untreated, and control rats. The cortisol-treated animals were i.v. injected with 5 mg hydrocortisone sodium succinate (Solu-Cortef®, Upjohn) per 100 g b. wt, and were sacrificed 24 h after cortisol administration. Splenectomy was performed 4 days before cortisol administration. The adrenalectomized rats were given saline solution to drink ad libitum and were sacrificed 7 days after adrenalectomy.

Various organs (table 1) were obtained from control and cortisol-treated animals; some organs (table 1) were also obtained from adrenalectomized untreated animals. All the above organs were fixed in 10% neutral formalin for subsequent histological studies[14]. The eosinophils were counted in 9 sections from each organ, and the tissue eosinophilia was expressed as the average number of eosinophils per mm^2 of tissue section. Blood samples were obtained from control animals, splenectomized untreated animals and 6 h after cortisol administration from both intact and splenectomized animals. EDTA was added to the blood samples; they were stained with 1% eosin Y in acetone. The eosinophils were counted in a Neubauer chamber.

Results. An increase in the number of tissue eosinophils is observed in several lymphoid organs, but not in other tissues, 24 h after cortisol administration (table 1). This increase is observed in spleen red pulp, in lymph node medullary substance, hilus, capsule and subcapsular space and in the thymus capsule. A decrease in the number of tissue eosinophils is observed in lymph node cortical substance, medullary substance, hilus, capsule and subcapsular space and in the thymus cortical substance 7 days after adrenalectomy (table 1). When comparing the results obtained in adrenalectomized animals with those obtained in intact cortisol-treated animals, a significant increase in tissue eosinophilia is observed in spleen red pulp, in lymph node cortical substance, medullary substance, hilus, capsule and subcapsular space and in thymus cortical substance, medullary substance and capsule in cortisol-treated animals (table 1).

Table 2 shows a dramatic decrease in the number of blood eosinophils 6 h after cortisol treatment in intact animals ($p < 0.001$). Only a slight decrease in the number of blood eosinophils is observed in splenectomized animals under a similar cortisol treatment as compared to untreated splenectomized animals ($p < 0.01$). Comparing splenectomized cortisol-treated animals with intact cortisol-treated animals, it is clear that the cortisol-induced blood eosinopenia is much more pronounced in intact animals than in splenectomized animals ($p < 0.005$). A small increase in the number of blood eosinophils is observed in untreated splenectomized animals as compared to controls ($p < 0.02$).

Discussion. Our results demonstrate a cortisol-induced migration of eosinophil leukocytes from the blood to

Table 1. Effect of adrenalectomy or cortisol treatment on tissue eosinophilia in various organs

Organs	Tissue eosinophilia per $mm^2 \pm$ SEM		
	Control rats	Adrenalectomized untreated rats	Intact 24 h cortisol-treated rats
Spleen red pulp	52.16± 9.20	49.00± 14.03[a]	107.00± 11.40[de]
Lymph node hilus	69.00± 10.15	39.25± 13.29[b]	382.09± 71.52[dg]
Lymph node capsule and subcapsular space	43.89± 7.10	13.33± 3.76[b]	141.83± 23.44[df]
Lymph node cortical substance	8.17± 1.78	1.63± 0.55[b]	8.69± 1.06[af]
Lymph node medullary substance	54.46± 6.05	18.00± 4.26[c]	134.62± 12.08[dg]
Thymus capsule	19.92± 3.94	9.17± 0.93[a]	35.10± 5.85[be]
Thymus cortical substance	6.33± 1.76	2.00± 0.71[b]	9.65± 1.43[af]
Thymus medullary substance	13.25± 2.46		21.70± 3.52[ae]
Oral mucous membrane	19.00± 5.81		7.29± 2.18[b]
Various other organs[h]			not significant

[a] not significant, as compared to controls; [b] $p < 0.05$, as compared to controls; [c] $p < 0.01$, as compared to controls; [d] $p < 0.001$, as compared to controls; [e] $p < 0.05$, as compared to adrenalectomized; [f] $p < 0.01$, as compared to adrenalectomized; [g] $p < 0.001$, as compared to adrenalectomized; [h] there is no significant statistical difference in stomach, duodenum, small intestine, large intestine, liver, kidney, ovary, lung and myocardium of intact 24 h cortisol-treated rats, as compared to controls.

several lymphoid organs, but not to other tissues. The finding that cortisol-induced blood eosinopenia is much less pronounced in cortisol-treated splenectomized animals than in intact cortisol-treated animals suggests that cortisol-induced blood eosinopenia is due, at least in part, to a migration of these cells to the spleen. The decrease in the number of tissue eosinophils in lymphoid tissues after adrenalectomy suggests that eosinophil migration to lymphoid organs may also occur in physiological conditions under the effect of endogenous levels of corticosteroid hormones.

The role of eosinophil leukocytes in lymphoid organs is unknown. Lymphoid organs seem to be target organs for cortisol. Specific receptors for cortisol have been described in rat thymus lymphocytes[15-17], as well as in other populations of lymphocytes[18]. Cortisol is known to inhibit glucose uptake and metabolism in thymus lymphocytes[19] and subsequently to decrease macromolecular synthesis[20]. Cortisol induces an involution of lymphoid organs[21] and a depression of the immune response[22]. Eosinophil leukocytes are thought to be involved in the immune mechanisms[23,24]. It is possible that the migration of eosinophil leukocytes to lymphoid organs under the effect of cortisol plays a role in the regulation of the immune reaction by cortisol. It is not known whether this migration is secondary to some of these conditions, to a release of an eosinophilotactic substance produced by T lymphocytes[25] or another eosinophilotactic factor, or is mediated by a direct influence on the cells, as was proposed to explain a specific eosinophil migration to other organs by other hormonal stimuli[14,26]. The possibility of trapping eosinophils in lymphoid tissue under the influence of glucocorticoids to remove these cells from the blood stream cannot be excluded. Further studies are necessary to elucidate the role of eosinophil leukocytes in the immune reaction.

Table 2. Effect of cortisol treatment and/or splenectomy on blood eosinophilia

Experimental condition	Number of eosinophils per mm^3 of blood ± SEM
Control rats	356.15 ± 50.22
Intact 6 h cortisol-treated rats	8.80 ± 5.39
Splenectomized untreated rats	574.17 ± 93.99
Splenectomized 6 h cortisol-treated rats	218.89 ± 40.28

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Effects of oestrogen and progesterone on rat pineal N-acetyl transferase activity and melatonin production

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Summary. We have extended previous studies on pineal β-receptors to include effects of oestradiol or PMSG treatment in the immature female rat. Neither manipulation has any effect on norepinephrine-induced N-acetyl transferase (NAT) activity in vitro. In the adult ovariectomised rat oestrogen/progesterone priming exerts a small sensitising effect to β-stimulation with isoproterenol. Progesterone alone, in vitro, inhibits the release of melatonin from pineals of adult ovariectomised rats.

We have reported[3] that the sensitivity of pineal β-receptors is responsive to changes in hypothalamic function. During precocious sexual maturation after hypothalamic lesions the pineals of female rats are more sensitive to norepinephrine. The cause of this increase is obscure, though high plasma oestrogen titres could be responsible. We have extended previous work to look at the effects on β-receptor sensitivity of different methods of priming the immature female rat with oestrogen. Oestrogen titres were increased either by injection of oestradiol benzoate (OB) in oil or by pregnant mare serum gonadotrophin (PMSG), a treatment known[4] to elevate blood oestrogen. The influence of oestrogen treatment on pineal adenylyl cyclase has been described[5] in the adult rat. Thus oestrogen/progesterone inhibits norepinephrine-stimulated adenylyl cyclase in vitro, whilst Illnerová[6] found no effect of this treatment on isoproterenol-stimulated NAT activity in vivo. We have repeated the experiments of Weiss and Crayton[5] and have determined the effects of oestrogen and progesterone on pineal NAT after stimulation with a very low concentration[7] of isoproterenol (5×10^{-9} moles/l). In addition preliminary experiments show that progesterone inhibits the release of melatonin in vitro from pineals of ovariectomised rats.