Diminished Corticosterone Levels in Nude Mice Implanted with MCF-7 or ZR-75-1 Human Breast Tumor Cells

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Summary. Corticosteroid levels were studied in the plasma of athymic mice implanted with human breast tumor cells, either from MCF-7 or ZR-75-1 cell lines. There was a highly significant decrease in plasma corticosterone levels in the mice implanted with these tumor cells. There was no significant effect on corticosterone of GW 39 colon cancer cells, LS 174 T colon cancer cells, or Calu-3 lung cancer cells.

There have been multiple studies of cortisol levels in women with breast cancer. Both significant [1, 2] and non-significant [3, 4] increases have been reported. However, the cause of the fluctuations is unknown. They might be due to the

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tumor itself, or they might reflect the anxiety of preoperative patients. Indeed, in one report, postoperative patients had normal plasma cortisol levels [5].

We have studied corticosteroids in athymic mice bearing human breast tumors derived from MCF-7 or ZR-75-1, two well established human breast cancer cell lines [6, 7].

Materials and Methods

Female nu/nu athymic mice, 3–4 months of age (Frederick Research Lab, Md.) were used. Cell suspensions (5×10^6 cells/100 µl/mouse) of ZR-75-1 cells (ATCC, Md.) or MCF-7 breast cancer cells (obtained from Dr. Charles McGrath, Michigan Cancer Foundation) were injected subcutaneously into the mammary pad region. Age matched control animals were injected with 100 µl of Tyrode's solution (DIFCO). One to two days prior to cell injections, a 21 day estradiol (E2) pellet with 0.5 mg E2 (Innovative Research, Inc.) was inserted into the scapular region of each animal with a 13 gauge trocar. Serum E2, which is necessary for the growth of breast tumors in athymic mice, was thus maintained at a level of 300–350 pg/nl.

Five to six weeks after injection, when the tumors measured 5–10 mm in diameter, each mouse was bled from the retro-orbital plexus at 5 pm, when corticosterone levels peak in mice [9]. Blood samples were centrifuged and stored frozen (–20 °C) until assayed for corticosterone.

We also evaluated the effect on corticosterone of three other types of implanted tumor cells: 1) GW 39 colon cancer cells, 2) LS 174 T colon cancer cells (ATCC, Maryland), and 3) Calu-3 lung cancer cells. The tumor-bearing animals and the controls in this group did not have the implanted 21 day estrogen pellets.

Results

The presence of human derived breast tumors in athymic mice significantly reduced plasma concentrations of corticosterone when compared to vehicle-injected control mice (Table 1). The presence of the MCF-7 or ZR-75-1 derived tumors led to decreases in corticosterone concentrations of six and four fold respectively. However, the implanted colon cancer or lung cancer cells had no significant effect on corticosterone levels (Table 2).

Tabelle 1. Serum corticosterone concentrations in nude mice implanted with either ZR-75-1 or MCF-7 breast cancer cells, and in unimplanted, vehicle-injected controls

Tumor type	ZR-75-1	Controls	
No. Mice	7	10	$t = 3.1$
Corticosterone (ng/ml)	89 ± 35	363 ± 146	$p < 0.001$ (2 tailed)
Tumor type	MCF-7	Controls	
No. mice	10	10	$t = 4.26$
Corticosterone (ng/ml)	39.5 ± 31.3	241.5 ± 139.4	$p < 0.01$

Tabelle 2. Serum corticosterone concentrations of nude mice implanted with colon or lung tumor cells, and of unimplanted controls. There was no significant difference in the corticosterone concentrations in this group of animals ($F = 2.60$, p n.s. by one way analysis of variance)

Tumor type	GW 39 (colon)	LS 174T (colon)	Calu-3 (lung)	Controls
No. mice	8	7	8	8
Serum corticosterone	179 ± 55.9	237 ± 83.5	203 ± 58.2	154 ± 36.4

Discussion

Almost any type of stress, whether physical or neurogenic, will cause immediate, marked increase of ACTH secretion by the anterior pituitary gland, followed within minutes by greatly increased adrenocortical secretion of cortisol. The cortisol secretion, in turn, causes rapid mobilization of amino acids and fats from their cellular stores, making these substances available for energy and for synthesis of other compounds, including glucose, needed by different tissues of the body [10].

Other investigators have reported elevated cortisol levels in advanced breast cancer patients. The levels were diminished by hypophysectomy and were, therefore, probably caused by a stress reaction, rather than by ectopic ACTH synthesis by the tumor itself [11]. Furthermore, although the subject is controversial [12], emotional stress and attitude may influence the survival of breast cancer patients [13].

In light of the human studies, our finding of dramatically diminished corticosterone levels in athymic mice with implanted tumors derived from human breast cancer cells is difficult to explain. Certainly the diminished levels are not due to any type of stress reaction. Perhaps the MCF-7 and ZR-75-1 cells are producing an agent which specifically lowers corticosterone, either by a direct action on the adrenal cortex, or by inhibiting the release of ACTH from the anterior pituitary.

It is reasonable to assume the secretion of such an agent, since breast cancer cells make many biologically active substances. Lippman et al. have found that MCF-7 and ZR-75-1 breast cancer cells elaborate Transforming Growth Factor (TGF) alpha and beta, a Platelet Derived Growth Factor (PDGF), and an autostimulatory mitogen, IGF-1 [14]. One of these substances, or some other, as yet unidentified, agent might be causing the diminished corticosterone levels we report here. Indeed, Hotta and Baird [15] have shown that TGF-beta inhibits steroidogenesis by suppressing Low Density Lipoprotein metabolism.

Alternatively, the implanted breast tumors may cause conversion of corticosterone to a non cross-reacting metabolite, thereby lowering the apparent serum corticosterone concentration. We hope that further studies may help to clarify this matter.

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