TABLE I. Immunity to Lymphosarcoma 6-C3H-Ed Produced in C3H (He) Mice by Prior Implantation of the Tumor.

Experimental procedure	No. mice	Progressive tumors	Palpable tumors regressed	No. tumor developed	Total negative to first tumor implantation	Result of reimplantation of tumor	
						Not immune	Immune
A-methopterin, 2 mg/kg/day for 7 days	62 (2)*	39	11	10	21	11	10
Tumor inj. in tail (no treatment)	30	11		19	19		19
Tumor inj. in tail (tails tied off)	8 (1)†		7		7		7

* This tabulation includes mice in experiments previously described(9).
† Number in parenthesis died during treatment.

Summary. Experiments are described in which cells of lymphosarcoma 6-C3H-Ed implanted in Jax-C3H (He) mice, in doses which failed to induce tumors when implanted into the tail, in large doses which induced tumors that were destroyed by intermittent occlusion of the blood supply, and by regression of palpable tumors through intensive treatment with A-methopterin, led to development of immunity of the mice against subsequent progressive growth of the same tumor.

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The Mast Cell. Cortisone Action on Connective Tissue.* (19729)

G. ASBOE-HANSEN. (Introduced by D. M. Angevine.)
From the Laboratory for Connective Tissue Research, Department of Anatomy, Faculty of Medicine, University of Copenhagen, Denmark.

During recent years the effect of hormones on connective tissue has been the object of intensive research, particularly after it became known in 1949 that the adrenal cortex influenced a number of connective tissue diseases.

It is known(10,11) that cortisone inhibits new formation of connective tissue and that fibroblasts in healing wounds of treated individuals are smaller and more pyknic than in wounds of untreated subjects.

The effect on the connective tissue ground substance has attracted particular attention, because the formation of ground substance appears to be the essential condition for new formation of connective tissue. Since various observations(1,2,5) indicate that hyaluronic acid, which is an important component of the connective tissue ground substance, is formed by the mast cells, interest has also centered on the effect of hormones on these cells.

The present author(3,13) found that in

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TABLE I. Material and Dosage of Cortisone and ACTH.

	Cortisone,* total dose		ACTH,† total dose (internat.	
	No.	(mg)	No.	units)
Humans	32	750–2600	151	240–860
Rabbits	15	150–275	23	45–105
Mice	20	4,4–11	11	5–14
Guinea pigs	11	8–10	16	5–100

* Cortone (Merck).
† Acton (Frederiksberg Chemical Factories).

human patients the mast cells of dermal connective tissue decreased in number and became degranulated when exposed to the action of ACTH and cortisone. At the same time, the hyaluronic acid of the ground substance was reduced. Cavallero and Braccini(7) confirmed this finding in experiments on the rat.

Since an increased amount of hyaluronic acid is of great pathogenetic significance in various connective tissue diseases(4,9,12), such as rheumatoid arthritis, pemphigus, acute disseminated lupus erythematosus, keloids, fresh cases of scleroderma, and perhaps also in iridocyclitis and glaucoma, the author feels that the suppressive action on the formation of hyaluronic acid is an important link in the clinical effect of cortisone on these diseases. The transmitters of this action might be the mast cells of the mesenchymal tissues.

The mast cells in dermal connective tissue were thoroughly studied during administration of cortisone and ACTH to humans,† rabbits, mice, and guinea pigs.

The following was derived from these experiments: The number of mast cells decreases, and certain morphological changes take place. A varying number of cells are perfectly normal even during and after the treatment, but most of them exhibit characteristic changes, depending somewhat on the dosage. While the granules are normally of approximately uniform size and rather evenly distributed in the cytoplasm, administration of cortisone and ACTH is followed by more or less marked degranulation; the granules are distributed in masses of varying size. A number of the mast cells show a vacuolized cytoplasm; the partitions separating the vacuoles consist of a homogeneous or partly granular substance which stains partly metachromatically, partly orthochromatically with toluidine blue. In some instances it is difficult to make out the cell boundaries. The cells are irregular, ragged in shape, of varying size, but on the whole, small.

† Unaffected skin taken from human patients suffering from various connective-tissue diseases.

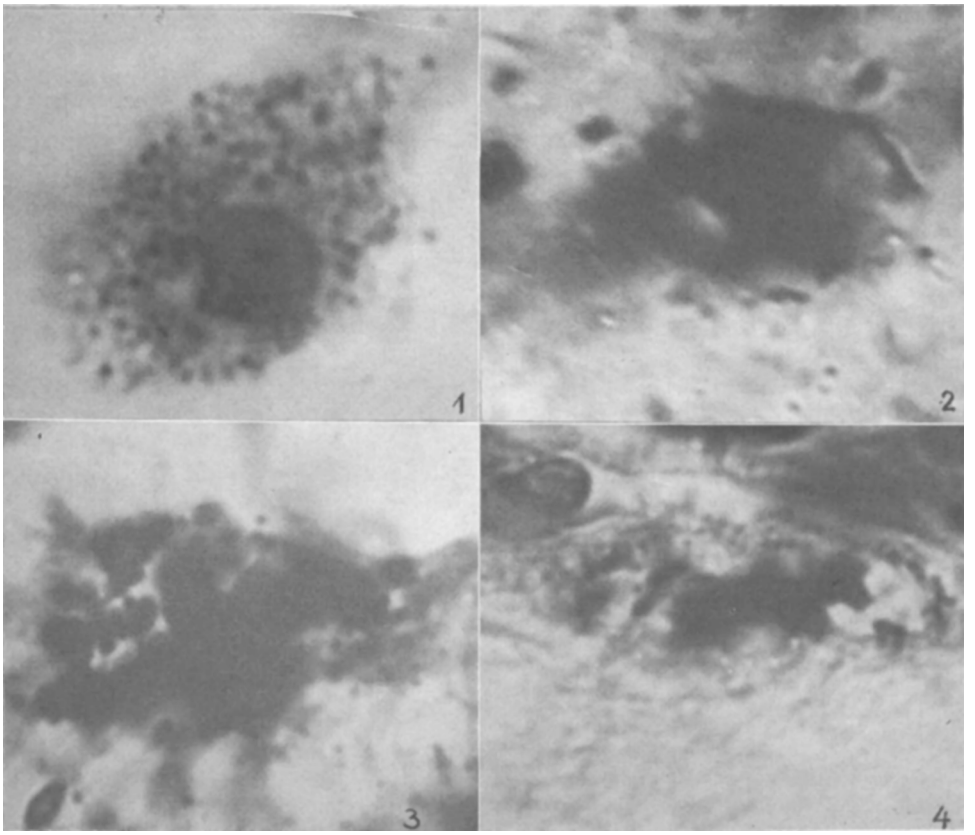
It seems evident that the reduction in the number of mast cells upon simple counting is due to the fact that many have lost their granules and thereby their histological characteristics.

Discussion. While ACTH and cortisone alter the mast cells to shapes which appear degenerated, the thyrotrophic hormone of the pituitary has proved to increase the number of mast cells which become highly granulated at the same time that the content of acid mucopolysaccharides increases. The condition is reversible, yielding to thyroxin(6,8). Similarly, the pathological state observed during administration of cortisone is perhaps reversible. It has not yet been ascertained whether the altered mast cells perish or whether their activity may be revived by discontinuing the hormonal action.

Summary. During administration of cortisone to humans, rabbits, mice, and guinea pigs the number of mast cells in the connective tissue is decreased. At the same time alterations are observed in the number and distribution of their granules, in the shape and outlines of the cells, and vacuolization occurs.

Since mast cells are presumably the origin of hyaluronic acid in the connective-tissue ground substance, this suppressive action is important when viewed against the background of the clinical effect of cortisone on connective-tissue diseases in the increased amounts of hyaluronic acid play an essential pathogenetic role.

After these studies were completed, Frank Bloom (PROC. SOC. EXP. BIOL. AND MED., 1952, v79, 651), has reported his experiments with malignant multiple mast cell tumors (mastocytomata). The tumors rapidly regressed and disappeared, and the neoplastic mast cells showed morphological changes like those described above.



Staining: 1% toluidine blue solution, aqueous. Magnification: approx. 3000 ×.

FIG. 1. Normal mast cell from human skin before administration of cortisone. The fine granules are uniformly distributed throughout the cytoplasm.

FIG. 2. After administration of cortisone, 1800 mg. Mast cell of dermal connective tissue showing vacuolization.

FIG. 3. After administration of cortisone, 1800 mg. Mast cell granules distributed in major and minor clusters and lumps, staining more or less intensely.

FIG. 4. After administration of cortisone, 1800 mg. Mast cell granules of irregular distribution, partly in the periphery, partly in lumps around the central nucleus. Vacuolization of the cytoplasm.

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