Influence of Cortisone on the Toxicity of Nitrogen Mustard in Rats. (20364)

LOUISE A. HUTCHINSON,* GOULD A. ANDREWS, AND RALPH M. KNISELEY.

From the Medical Division, Oak Ridge Institute of Nuclear Studies, Oak Ridge, Tenn., under contract with the U. S. Atomic Energy Commission.

Cortisone and nitrogen mustard are sometimes combined in the therapy of lymphomas, leukemias, and other neoplastic conditions. There is relatively little information available on the combined effects of these 2 drugs. Nitrogen mustard and radiation are recognized to produce similar effects(1). A number of reports have been published on the action of cortisone on modifying the effects of total body irradiation. It has been reported that only 31% of mice receiving irradiation during cortisone administration survived, whereas there was 100% survival with either irradiation or cortisone alone(2), and a less pronounced but similar increase in mortality was noted in another laboratory in which cortisone was started immediately after irradiation(3). Thiersch(4) reports a delay in bone marrow regeneration in irradiated rats treated with cortisone. However, Mirand *et al.*(5) using mice found that cortisone afforded protection against radiation effects. Alpert, Zimmerman, and Scheer(6) making observations on combined therapy of malignant disease in humans observed that patients who failed to respond to either cortisone or nitrogen mustard alone, would not respond to the combination. However, their experience suggested that cortisone had some protective effect against the leucopenia resulting from mustard. In a discussion of the above work, Schoenback(7) observed that in experiments on mice he had not noted any reduction in the toxicity of mustard when used with ACTH or cortisone.

The following experiment was performed to observe the effects of cortisone on the LD_{50} (5-7 days) of HN_2 in rats.

Materials and methods. A pilot experiment showed that the LD_{50} dose of HN_2 administered subcutaneously was 1.2 mg/kg for male Wistar rats 5 months old and weighing 300-400 g. In the subsequent experiment a total of 60 rats were used. Two groups of 20 animals were started on cortisone 5 mg subcutaneously daily and a third group of 20 was started on control injections of .2 cc distilled H_2O subcutaneously. On the third day an LD_{50} dose of HN_2 was given to one group of 20 rats receiving cortisone and the group receiving control injections. The third group received only cortisone. All animals were observed for a period of 7 days following administration of the mustard, and during this time the daily cortisone and control injections were continued. A second experiment duplicating the conditions of the first was set up for the purpose of obtaining histological and bone marrow specimens. Animals were sacrificed on the second, fourth, fifth, seventh, and ninth days after injection of HN_2. Histologic sections of adrenal, spleen, and bone marrow were prepared. Smears of bone marrow were also made. The adrenals and spleen were weighed. In addition to these studies on sacrificed animals, similar information was obtained on some of those that died in both series.

Results. Table I gives the results of the mortality study. It shows no striking effect of the cortisone in either enhancing or protecting against HN_2 toxicity.

In the second experiment which was set up for the purpose of obtaining histologic data, the spontaneous death rate in the group of animals receiving nitrogen mustard alone was higher (13 animals) than in the group on HN_2 plus cortisone (8 animals). Because animals were being sacrificed at random in these groups, no effort was made to interpret this observation.

Bone marrow studies showed pronounced destruction on the second day and extensive depletion by the fourth day in Groups I and

* Present address: Department of Internal Medicine, Tulane University Medical School, New Orleans, La. Work done at Oak Ridge under Resident Physician contract.

TABLE I. Effect of Cortisone in Rats Given an LD_{50} Dose of HN_2.

Days after administration of HN_2	No. of rats dead		
	Group I Mustard alone	Group II HN_2 and cortisone	Group III Cortisone only
0-3	0	0	0
3-4	1	2	0
4-5	4	6	0
5-6	5*	2	0
6-7	0	0	0
Total deaths	10	10	0
Survivors	10	10	20

* One traumatic death.

II which received HN_2 and HN_2 plus cortisone respectively. Persisting cells were largely plasma cells and reticulo endothelial elements. Megakaryocytes never disappeared completely. Mast cells remained plentiful. Starting on the fifth day regeneration was noted, and this progressed so that hyperplasia was present on the seventh and ninth days. In Group III (cortisone alone) the marrow picture showed no distinct changes during the experiment.

The spleens in all groups showed a definite shrinkage in size from the second through the ninth days. In the initial period this decrease in size appeared greater in Groups I and II than in Group III.

In Group III (cortisone alone) histologic studies showed preservation of the white pulp while in both other groups there was extensive atrophy of lymphatic tissue and a lack of evidence of activity of lymphopoietic centers of the follicles. There appeared to be some regeneration of the white pulp by the ninth day in Groups I and II, but this was minimal and did not compare with the striking regenerative change seen in the bone marrow.

The weights of adrenal glands indicated a definite shrinkage in Group III (cortisone alone) at intervals studied up through the ninth day. In the other 2 groups, changes in size were not striking or consistent except on the fifth day after the HN_2, when there appeared to be definite hypertrophy in Group I, with Group II being intermediate in size between Groups I and III.

Discussion. Under the conditions of the present experiment, the LD_{50} of nitrogen mustard was the same in rats receiving cortisone as in rats not receiving cortisone. The cortisone which was started 2 days before the HN_2 was given, failed to modify the severe bone marrow depression. The studies on the weights and histologic changes of adrenals and spleens were inconclusive because of the small number of animals used in the sacrifice experiments and because of difficulties in interpreting changes after spontaneous death. Both the cortisone and mustard appeared to cause splenic atrophy. In the adrenal the stress produced by the mustard resulted in enlargement which was maximum at the fifth day, and in the animals receiving both drugs this effect was partially neutralized by the cortisone which alone tended to produce atrophy.

This experiment represents an arbitrary set of conditions in the use of the 2 drugs together. Both have complex and widespread metabolic effects. It is likely that changes in the dosages and time sequence would produce results different from those which we obtained. It is entirely possible that some of the effects of cortisone are beneficial while others are harmful to the animal suffering from nitrogen mustard toxicity.

Summary. Cortisone failed to produce alteration in the LD_{50} of nitrogen mustard in rats under one set of experimental conditions.

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