lymphocytes, it would seem that the delayed lymphocytocidal action may be a function of the C-11-oxygenated hormones.

Feldman(3) demonstrated that large amounts of adrenal cortex extract had an immediate cytotoxic effect on lymphocytes. The present study confirms this finding. Feldman observed in addition that the extract, in large doses, killed not only lymphocytes but many other types of cells and that the immediate cytotoxic effect took place at room temperature as well as at physiologic temperatures. In this laboratory, small and moderate doses of adrenal cortex extract had no effect on cells other than lymphocytes and had no effect on lymphocytes at room temperature. In these respects the immediate and delayed cytocidal effect appeared distinct.

To explain the relationships between the immediate and the delayed cytotoxic effect of adrenal cortex extract, it may be assumed that a compound in the extract affects two distinct structures or processes of the cell. According to this hypothesis, small and moderate doses of the adrenal cortex extract alter a specific physiologic process in lymphocytes incubated at 37°C and hasten their death. A corollary of this hypothesis is that this physiologic process is absent or not essential to other cells at 37°C and to lymphocytes incubated at room temperature. In contrast, large doses apparently act as a universal protoplasmic poison and affect many types of cells at a wide temperature range.

A similar hypothesis may be used for the effect of x-rays on cells. Small doses of x-rays seem to have a specific action on mature lymphocytes and mitotic cells. Large doses affect all living cells. This "dual mechanism" hypothesis for the action of adrenal cortex and x-rays, seems useful as it explains a number of diverse findings.

Summary and conclusions. The proposed method of assay consists of incubating cellular suspensions derived from the rabbit thymus with adrenal cortex preparations at 37°C for 24 and 44 hours. The number of viable cells in the incubated and treated suspensions were determined by the method of unstained cell counts. The cytocidal unit was defined as that amount of reagent which killed 50% of the cells as compared to a control, incubated suspension. The cytocidal unit was found to be 0.025 γ for 17-hydroxycorticosterone and 0.1 γ for corticosterone and cortisone.

Small doses of the adrenal cortex preparations had a delayed cytocidal effect whereas large doses of adrenal cortex extract and corticosterone had an immediate effect.

Received June 26, 1951. P.S.E.B.M., 1951, v77.

Progressive Growth of C3H Mouse Lymphosarcoma in CF_1 Mice Treated with Cortisone Acetate. (18902)

E. J. FOLEY AND RUTH SILVERSTEIN. (Introduced by R. Tislow.)

From the Biological Research Laboratories, Schering Corporation, Bloomfield, N. J.

The relationships of growth and regression of strain specific transplantable mouse tumors when implanted in alien strains of mice has been established by Lewis(1). It was shown that spindle cell sarcomas arising in inbred strains of mice would grow progressively and kill when implanted in mice of the strain in which they originated. When transplanted in mice of alien genetic constitution however, the tumors would grow and then regress. The sojourn of such tumors for four days in alien strain mice conferred immunity against subsequent implantation of the same tumor.

Similar studies have recently been made by Kidd and Toolan(2) who implanted C3H mammary carcinoma and lymphosarcoma

1. Lewis, M. R., *Bull. Johns Hopkins Hosp.*, 1940, v67, 325.

2. Kidd, J. G., and Toolan, H. W., *Fed. Proc.*, 1949, v8, 360.

TABLE I. Effect of Cortisone Acetate Treatment* on Progressive Growth of Lymphosarcoma 6C3H-ED in CF_1 Mice.

	No. of mice	Died of tumor	Tumor regressed	Died during treatment
Cortisone treated	30	17	6	7
Untreated controls	37	2	35	0

* Cortisone acetate aqueous suspension 40 mg/kg/day subcutaneously for 10 days.

6C3H-ED into A strain mice. The temporary growth of these tumors in A strain mice conferred immunity against subsequent implantations of the same tumors. By appropriate experiments(3,4) it was demonstrated that this immunity was associated with a necrobiotic function of lymphocytes against the implanted tumor cells.

Stoerk(5) has shown in rats and mice that regression of lymphosarcoma during riboflavin deficiency is followed by an acquired resistance against reimplantation with the same tumor. This immunity was gradually lost over a period of one year, and was rapidly diminished when marked loss of normal lymphoid tissue was produced by the administration of cortisone or desoxypyridoxine.

The implication of lymphocytes in tumor immunity processes and in the destruction of tumor cells transplanted into alien strains of mice, suggested that the resistance which alien strain mice possess against heterologous tumors might be obliterated if marked destruction of lymphocytes could be effected at the time of implantation of the tumor. To test this possibility, experiments were made to determine whether intensive treatment with cortisone would render CF_1 mice susceptible to the progressive growth of lymphosarcoma 6C3H-ED. The results of preliminary experiments are presented.

Materials and methods. Young adult CF_1 (Carworth Farms) male mice weighing between 17.5 and 18.5 g were implanted subcutaneously in the right axillary region under light ether anesthesia with small fragments (10 to 12 mg) of the Gardner lymphosarcoma 6C3H-ED, using an 18 gauge trocar. All transplants were from tumor carried by serial passage in JAX C3H mice.

Cortisone acetate was given by daily subcutaneous injection at a dose of 40 mg/kg in 0.25 ml of distilled water. The first dose was given about 4 hours before implantation and was repeated once every day for a total of 10 doses. Implanted mice were palpated on alternate days to determine the growth behavior of the tumor.

All mice were fed Wayne Fox food *ad lib.* and had water available to them throughout the experiments.

Results. The implanted lymphosarcoma grew in all untreated CF_1 mice and became palpable on the fifth or sixth day after implantation, reaching a maximum size of 6000 to 7000 mm^3 in about 2 weeks; after which rapid regression took place. In contrast, the majority of cortisone treated mice grew progressive tumors which cause death of the animals between the twenty-first and twenty-fifth day following implantation. These mice at the time of death carried enormous tumors which had extended to the opposite flank, into the thoracic and neck region, and around the fore and hind legs on the side of the original implantation. The results of several experiments are combined and summarized in Table I.

It is seen in Table I that only 2 of 37 of the untreated CF_1 mice implanted with lymphosarcoma 6C3H-ED died of the tumor, whereas 17 of the 23 mice surviving cortisone treatment died of progressive tumor growth. Thus, treatment with massive daily doses of cortisone acetate reduces the natural strain resistance of CF_1 mice to this tumor. A highly significant difference was found to exist between cortisone treated mice and untreated controls when tested for significance

3. Kidd, J. G., and Toolan, H. W., *Fed. Proc.*, 1949, v8, 373.

4. Kidd, J. G., and Toolan, H. W., *Fed. Proc.*, 1950, v9, 385.

5. Stoerk, H. C., *Fed. Proc.*, 1951, v10, 372.

by the method outlined by Gray(6).

Seven of the 30 implanted mice treated with cortisone died before the tumors reached appreciable size; 4 died on the eighth, 2 on the ninth, and 1 on the eleventh day following the beginning of treatment. It is presumed that these mice died as a result of the large repeated doses of cortisone since 4 out of 18 normal unimplanted male CF_1 mice, similarly treated in experiments made to ascertain lymphoid tissue reducing effects, died between the seventh and tenth day of treatment. Normal mice receiving 40 mg/kg of cortisone acetate subcutaneously for 10 consecutive days show at autopsy profound atrophy of the thymus, reduction in the size of the spleen and considerable loss of weight.

Discussion and summary. The data show that the resistance of alien strain (CF_1) mice to the progressive growth of lymphosarcoma 6C3H-ED (originating in C3H mice) is greatly impaired by intensive treatment with cortisone acetate. Seventeen of the 23 CF_1 mice which survived treatment with cortisone died of progressive growth of lymphosarcoma implanted from C3H mice, whereas only 2 of 37 untreated mice developed progressive tumors.

It should be pointed out that the course of cortisone dosage used to bring about the effects described were excessive, actually constituting a lethal concentration for some of the CF_1 mice. The nature of the change in resistance shown by these experiments has not been established, but it might be inferred that it is associated with the well known depression of normal lymphoid tissue and immune responses which large doses of cortisone induce.

Addendum. After the above manuscript was submitted for publication, a paper by E. L. Howes (*Yale Jour. Biol. and Med.*, 1951, v23, 454) came to our attention. This paper described the progressive growth of adenocarcinoma EO771 or 775 when transplanted in Rockland Swiss mice treated with cortisone.

6. Gray, P., *Arch. Biochem.*, 1947, v13, 461.

Received June 29, 1951. P.S.E.B.M., 1951, v77.

Effect of Roentgen Irradiation upon Protein Absorption in the Mouse.* (18903)

LESLIE R. BENNETT, SARAH M. CHASTAIN, ARTHUR B. DECKER, AND JAMES F. MEAD.
From the School of Medicine, University of California at Los Angeles.

In a study of the effect of X-irradiation upon fat absorption in the mouse Mead, Decker and Bennett(1) showed that contrary to most earlier reports, no impairment of absorption could be demonstrated and that the results of previous workers could often be explained by radiation-induced changes in motility of the gastrointestinal tract.

In a continuation of these experiments we have now investigated the effect of X-irradiation upon protein absorption. The purpose of this study was to determine first, whether the previous findings represented an isolated case or could be generalized to cover other types of nutritive elements; and second, whether the damaged small intestine might permit the passage of appreciable quantities of material with molecular sizes larger than those of the amino acids. Under ordinary circumstances in normal animals, proteins are completely broken down before absorption. However, evidence has accumulated which indicates that in the irradiated animal intestinal permeability may be altered so as to allow the passage of bacteria(2). It was thought that the possibility of the absorption of intact protein could readily be demon-

* This paper is based on work performed under Contract No. AT-04-1-GEN-12 between the Atomic Energy Commission and the University of California at Los Angeles.

1. Mead, J. F., Decker, A. B., and Bennett, L. R., *J. Nutrition*, 1951, v42, 485.

2. Hammond, C. W., and Miller, C. P., *Ann. New York Acad. Science*, 1950, v53, 303.