TABLE I. Effect of ACTH on Distribution of Radioberyllium 11 Days after Injection of Be^7. % injected dose of Be^7.*

Tissue	Carrier-free Be^7		Be^7 + carrier	
	Be only	Be + ACTH	Be only	Be + ACTH
Liver	13.7 ± 1.7	10.4 ± 1.3	19.5 ± .3	17.8 ± .9
Femurs	2 ± .1	1.6 ± .1	1.9 ± .2	2 ± .04
Kidneys	.15 ± .02	.17 ± .03	.51 ± .06	.49 ± .02
Spleen	.58 ± .03	.47 ± .04	3 ± .2	2.8 ± .5
Lungs	.07 ± .004	.08 ± .004	.22 ± .02	.22 ± .03
Carcass	27.9 ± 1	24.3 ± .6	32.4 ± 1.4	31.7 ± 1.3
Total	44.4 ± 2.7	37.1 ± .8	57.5 ± 2	55 ± .7

* Each value is a mean based on 3 mice, except in the Be^7 + carrier control group, which consisted of 2 mice. Deviation is expressed as the standard error of the mean.

lomatous infiltration and possibly of the fibrosis(1,8) with resultant reduction of the alveolar-arterial difference in oxygen partial pressure(9).

The lack of effect of ACTH both on the survival of mice acutely poisoned with Be and on Be distribution suggests that there is no direct reaction between beryllium and either ACTH or any substance produced by ACTH activity. On the other hand, successful antidotal action against acute experimental beryllium poisoning in mice in the case of aurintricarboxylic acid is associated with the known ability of this substance to react chemically with Be under physiological conditions even though it produces no redistribution or increased excretion of Be(10,11).

Summary. Adrenocorticotrophic hormone (ACTH) had no appreciable effect on the distribution or retention of radioberyllium, injected into mice with and without carrier beryllium sulfate. The survival of mice acutely poisoned with beryllium sulfate was not influenced by ACTH.

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Protective Effect of Serotonin and of Para-Aminopropiophenone Against Lethal Doses of X-Radiation. (19706)

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In a previous publication(1) it was demonstrated in rats that pretreatment with pitressin or epinephrine afforded pronounced protection against total body x-irradiation. The protective effect was assumed to result from the production of a temporary tissue anoxia

TABLE I. Effect of Serotonin, PAPP and $NaNO_2$ on Survival of Rats after 880 r Total Body X-Irradiation.

Treatment (IP)	Time of treatment relative to x-radiation, min	Survival, 28-day		% methemoglobin	
		No.	%	Start of x-ray	Avg during x-ray period
Serotonin					
Control (H_2O)	5 before	3/55*	6		
20 mg/kg	"	30/31	97		
4 "	"	10/38	26		
PAPP					
Control (propylene glycol)	30 before	26/77	34	1.5	—
32 mg/kg	"	29/31	94	76.9	77.2
16 "	"	29/30	97	73.1	71.3
6 "	"	14/30	47	55.8	54.4
$NaNO_2$					
Control (saline)	10 or 30 before	5/44	11	2	—
60 mg/kg	30 "	9/30	30	52	54.7
60 "	10 "	2/23	9	25.4	42

* First number indicates survivals; following number indicates total animals, *i.e.* 3 survived out of 55.

by these agents. Further investigation of this pre-protection by vasoconstrictor agents was made employing serotonin (5-hydroxytryptamine), the vasoconstrictor agent present in blood platelets(2). In addition, to substantiate further the concept of the possible significance of the amount of oxygen in the tissue with regard to radiation effects, studies have been made on methemoglobinemia as a means of producing a reduced tissue oxygen tension.

Methods. Male Sprague-Dawley rats, weighing 270 ± 10 g, were irradiated in pairs, one serving as control, the other as a treated animal. Each pair was exposed to total body x-irradiation for 22 minutes in a single exposure. Radiation factors were: 200 kv, 6 ma, ½ mm Cu 1 mm Al filter, target distance approximately 29 cm, and 40 r/min., dosage rate measured in air.*

Either 1 or 5 mg of serotonin creatinine sulfate† in ½ ml of water were injected intraperitoneally 5 minutes before exposure. Methemoglobinemia was produced by the administration of either sodium nitrite or para-aminopropiophenone. Sodium nitrite was dissolved in isotonic saline to a concentration of 20 mg $NaNO_2$ per ml. Doses of 0.75 ml were administered intraperitoneally 10 or 30 minutes before exposure. Dosage levels of para-aminopropiophenone‡ were 8, 4, and 1.5 mg per animal, each administered intraperitoneally in 1 ml of propylene glycol. Blood methemoglobin levels were determined by a modification of the method of Evelyn and Malloy, as described by Storer and Coon(3). All controls received equivalent amounts of the appropriate solvent. Animals were housed in individual cages and weighed daily until death or termination of the experiment after 28 days.

Results. As would be expected with an 880 r dose in total body exposure, the majority of deaths occurred between 6 and 14 days. The effect of serotonin creatinine sulfate on survival rate after total body x-irradiation is shown in Table I. A dose level of 4 mg/kg body weight injected 5 minutes before exposure produced little, if any, protection; however, 20 mg/kg administered at the same interval prior to exposure produced a striking protective effect, as indicated by a survival rate of 97%, as compared with 6% survival for the control group.

Sodium nitrite and para-aminopropiophenone (PAPP) in varying doses were used to

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‡ We are grateful to the Dow Chemical Co., Midland, Mich., for supplying us with this preparation.

produce different blood levels of methemoglobin. Table I shows the methemoglobin level at the beginning of the experiment and the average level during the irradiation period. With PAPP, dosage levels of 32 and 16 mg/kg body weight produced average methemoglobin levels of 77 and 71% during the period that irradiated animals were exposed. Sixty mg/kg body weight of $NaNO_2$ 30 minutes prior to irradiation produced an average methemoglobin level of 54.7% during the period of exposure, compared to 54.4% resulting from a dosage of 6 mg/kg of PAPP. The survival rates obtained with these doses of PAPP and $NaNO_2$ are also shown in Table I. The two higher doses of PAPP, 32 and 16 mg/kg, gave excellent protection as evidenced by the 94 and 97% survival at 28 days. $NaNO_2$ (60 mg/kg) and PAPP (6 mg/kg) administered 30 minutes before exposure gave very nearly the same slight protection when the 28-day survival rates were compared with their respective controls. One group injected with $NaNO_2$ (60 mg/kg) 10 minutes before x-irradiation showed no protective effect with an average methemoglobin level of 42% during the period of exposure. The control animals in the PAPP experiments received 1 ml of propylene glycol 30 minutes prior to exposure and the survival rate in this group was higher than in the untreated or saline treated controls.

Discussion. The protective effect elicited by serotonin creatinine sulfate is assumed to lie in its vasoconstrictor property(2), producing a transient tissue anoxia in a manner similar to that of epinephrine(1). The possible influence of this agent on metabolism has not as yet been fully investigated.

Apparently there is a correlation between the degree of methemoglobinemia and protection against radiation. The protective effect of methemoglobinemia at certain concentrations may be due to the decreased supply of oxygen to the tissues, thus rendering them anoxic. The results with $NaNO_2$ and PAPP reported here agree with similar observations obtained with mice by Storer and Coon(3). The possibility that PAPP and $NaNO_2$ may affect the oxygen uptake of the tissue directly has to be taken into consideration. Cole, Bond, and Fishler(4) have reported that the mortality of mice receiving 600 r or 750 r single-dose whole body x-irradiation was reduced markedly following pre-irradiation intraperitoneal injection of $NaNO_2$ (100-125 mg/kg). They discuss the possibility that nitrite protection is mediated via its effect on catalase activity. The dose level of $NaNO_2$ per kg employed by these authors is higher than that used in the present investigation. However, it was found that 60 mg/kg was very close to the lethal dose for the rats employed. Unfortunately, the above authors did not carry out any methemoglobin determinations, permitting a comparison of methemoglobin levels with the degree of protection. The observation that administration of propylene glycol apparently exerts a slight protection is in agreement with similar observations of Salerno, Mattis, and Friedell(5).

The findings on the protective action of serotonin and PAPP, when administered prior to a lethal dose of x-irradiation, seem to substantiate the concept of a relationship between tissue oxygen tension and radiosensitivity. Alterations of susceptibility to the effects of radiation by changing oxygen tension in the tissues are probably due to the reduction in the formation of reactive decomposition products of water. In such a concept, cognizance should be taken, of course, of the effects of those decomposition products of water (such as the OH radical), which are formed on irradiation in the absence of dissolved oxygen.

In this connection, it may be opportune to refer to the observations of Bennett, Chastain, Flint, Hansen, and Lewis(6) that life span of rats receiving 600-1400 r whole body roentgen irradiation under anoxia was considerably shortened. This occurred in spite of the protection the anoxia gave against the lethal action of x-rays in the immediate post-irradiation period of 28 days. However, the main object of our investigation was to gain further insight into the mechanism of radiation injury and not so much to find protective means against radiation.

Summary. 1. The survival rate of rats, exposed to lethal x-ray dosage, was found to be significantly increased after pretreatment with

serotonin creatine sulfate (20 mg/kg) or para-aminopropiophenone (32 mg or 16 mg/kg). 2. The protective effect of these agents is assumed to be due to their property of producing a temporary tissue anoxia.

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Inhibition of Bacterial (*Escherichia Coli*) Modification of Erythrocytes. (19707)

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It is an established fact that certain viral and bacterial antigens may be adsorbed on red blood cells; this may result in hemagglutination (direct microbial hemagglutination), in modification of the erythrocytes or both. These modified red blood cells are specifically agglutinated by the homologous antimicrobial antibody (indirect microbial hemagglutination). It was shown recently from this laboratory(1,2) that boiled suspensions of *E. coli* serogroups 0111 and 055 as well as sterile broth culture filtrates are capable of modifying red blood cells from a variety of animal species. These modified red cells are agglutinated and, in the presence of complement, lysed by the group-specific *E. coli* antiserum. It was demonstrated furthermore that this agglutination and hemolysis is specifically inhibited by the homologous bacterial antigen. The experiments to be recorded in this communication revealed that certain materials inhibit the modification of red blood cells by *E. coli* antigen.

Material and methods. In addition to *E. coli* serogroups 055 and 0111 used in the previous experiments several strains of *E. coli* 026 were employed, since the latter serogroup, too, has been found to be associated with epidemic diarrhea of infants(3). For the modification of red blood cells boiled suspensions from Kolle flasks were used, as described previously(2). The microorganisms were suspended in the materials to be tested for inhibitory activity, namely, serum from man and various animals, human plasma fractions, bovine albumin (Armour), egg white, egg yolk, various fractions of rat liver, and, for control purposes, physiological saline solution. The human plasma fractions were obtained from Cutter Laboratories through the courtesy of Dr. F. F. Johnson and the rat liver fractions through the kindness of Dr. Charles U. Lowe. The mixtures were kept at room temperature for 15 minutes and then used for modification of red blood cells. The treated erythrocytes were washed 3 times in physiological saline solution. The hemagglutination and hemolysis tests were carried out as described in detail previously(2).

Results. It was found that a variety of human and animal sera, including sera from healthy adults and children, human cord serum, calf, beef, horse, sheep, chicken, duck and turkey sera in dilutions of 1:10 and 1:100 either completely or partially inhibited the modification of sheep red blood cells by *E. coli* 026 antigen. Quantitative experiments revealed that the more concentrated the *E. coli* antigen the more serum is required to produce inhibition of modification of red cells. The