TABLE I. Survival Time Response of Hypophysectomized Rats Weighing 80 to 150 g Given 0.5 mg of Endotoxin Intraperitoneally Followed Immediately by Intravenous Injection of Corticotropin (ACTH).

		Total dose of corticotropin in milliunits							
		0.0		0.5		1.5		4.5	
Day	Sex	No. rats	Mean survival time, min.	No. rats	Mean survival time, min.	No. rats	Mean survival time, min.	No. rats	Mean survival time, min.
1	♂	6	92.7 ± 14.9*	5	113.8 ± 19.5*	11	145.7 ± 8.2*	9	160.8 ± 12.1*
2	♀	9	136.7 ± 10.8	7	122.7 ± 10.7	11	148.8 ± 9.1	10	140.8 ± 6.4
3	♂	9	114.4 ± 9.4	6	130.7 ± 17.5	7	105.4 ± 10.5	7	196.7 ± 31.5
4	♀	9	123.2 ± 7.7	9	157.0 ± 11.2	9	163.8 ± 11.2	11	168.7 ± 10.8
5	♂	7	125.6 ± 9.0	5	113.0 ± 10.9	6	161.8 ± 22.4	8	173.2 ± 19.1
Combined		40	120.1 ± 8.4	32	130.9 ± 6.6	44	146.0 ± 5.6	45	166.1 ± 7.2

* Stand. error of mean.

approximately 0.3 and 1.0 for hydrocortisone and corticotropin, respectively. 3. The effect of sex and age of animals and time of administration on response to hydrocortisone were studied.

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Urinary Excretion of Beta Aminoisobutyric Acid (BAIBA) in Irradiated Human Beings.* (24548)

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Recently a previously unknown amino acid, beta-aminoisobutyric acid (BAIBA) has been identified in human urines by Crumpler, Dent *et al.*(1), and Fink *et al.*(2). Further studies by Fink using C^{14} labeled thymine indicate that BAIBA is a product of thymine metabolism(3,4) (Fig. 1). Since thymine is a pyrimidine peculiar to deoxyribonucleic acid (DNA), it appears likely that under certain circumstances BAIBA excretion may reflect alterations in DNA metabolism. Aminoaciduria has been reported following total body irradiation(5,6). Analysis of the amino acids studied indicated that glycine and taurine were excreted in increased amounts from presumed protein catabolism. Pyrimidine metabolites such as BAIBA have not been studied previously to our knowledge following total body irradiation, however. Awapara(7) noted increased BAIBA excretion followed nitrogen mustard or local x-ray treatment in some leukemic patients. In June 1958, in the course of an industrial operation involving uranium, a critical geometry accidentally occurred. Eight individuals nearby were exposed to the resultant mixed neutron and

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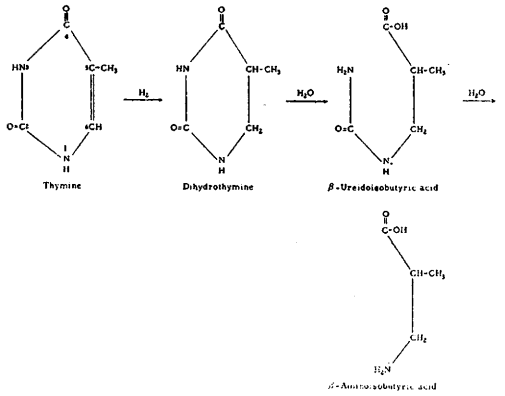

FIG. 1. Thymine metabolism.

gamma radiations. Five of the 8 persons were heavily exposed and developed early symptomatology. The remaining 3 with less exposure had no symptoms. A full description of the incident and the clinical courses of the involved personnel has been reported by Brucer and Andrews(8). The present studies concern measurements of these irradiated subjects, in whom large masses of radiosensitive tissues were undergoing dissolution and alteration.

Materials and methods. Frozen 24 hour urine aliquots were thawed and processed for BAIBA measurement using a modification of the method described by Awapara and Sato (9). Ten ml aliquots of the urines were mixed with 1 g portions Darco G-60 charcoal and heated briefly in a water bath. Following centrifugation 6.0 ml of the charcoal treated urines were placed on Dowex-2 columns (previously converted to the hydroxide form) for desalting. After the urinary supernatants had entered the resin beds, the columns were washed with cold boiled distilled water until the effluent became neutral. The columns were then eluted with enough 4N acetic acid to collect 10 ml. Eluates were lyophilized and resultant dried materials dissolved in 1 ml portions of distilled water. These concentrates were frozen in preparation for chromatography. Two-dimensional chromatography was used for identifying BAIBA. Ten lambda (.01 ml) of the concentrates were spotted on 8 inch squares of Whatman No. 4 paper held in the frame described by Datta *et al.*(10). Solvent 1 was the organic phase of n-butanol, acetic acid and water (5:1:4) and solvent 2 consisted of 2,4, lutidine saturated with water. After drying, the papers were sprayed with ninhydrin reagent and the color developed by heating at 100°C. Known amounts of BAIBA of varying concentrations were dissolved in water and chromatographed simultaneously. BAIBA was readily identified by Rf measurement (Rf 1 = 0.45 ± .02; Rf 2 = 0.32 ± .02). Color intensity and size of the ninhydrin spots were graded 0 to 5+ by 2 independent observers. Semiquantitative estimation of BAIBA in urines in μM/L were performed by comparison with standards and calculated on the basis of 24 hour urine volumes. Duplicate determinations were reproducible. Identical amounts of BAIBA have been added to BAIBA "negative" urine and also to equal volumes of water and then processed as above. Analyses of the urine samples always showed lower BAIBA content than BAIBA in water samples. The reasons for this are under study. The BAIBA detected in the experimental urines thus does not represent all of the BAIBA actually excreted. However the losses we believe are uniform(7).

Results. Table I shows the measured daily urinary BAIBA levels of 8 individuals hospitalized after radiation exposure. BAIBA is expressed in μM/Liter of urine. The 24 hour urine volumes are also recorded. The first urine samples for these studies were obtained on third day after exposure. BAIBA excretion was ranked according to quantity excreted, then the exposure doses supplied by Brucer and Andrews(8) were compared to this order. The fair parallelism of excretion with dose is evident. In Fig. 2 is shown the total μM BAIBA excreted per day in these patients.

Discussion. The data presented demonstrate that there is an apparent increase considerably above normal in BAIBA excretion in irradiated human beings. There are good indications that the excretion pattern is dose dependent. In our laboratory, maximum excretion in urines of normal individuals and in urines of patients with non-neoplastic disease has been between 0 to 150 μM/Liter of urine. Awapara(7) states that BAIBA excretion in normal individuals is an uncommon

TABLE I. Urinary BAIBA Excretion in 8 Patients following Irradiation.

Patient	1	4	3	5	2	6	7	8
Estimated exposure in rads(7)								
Neutrons	66	57	55	45	39	14	8	4
Gamma	210	182	175	143	114	43	34	17
Total	276	239	230	188	163	57	42	21
BAIBA excretion, μM/l, and 24 hr urine vol								
Day 3	3+ 920	3+ 1975	2+ 3390	0 3150	+ 2320	2+ 1410	0 1220	0 2040
4	4+ 1300	3+ 1035	3+ 1610	2+ 1980	2+ 2290	0 1360	0 2940	0 3480
5	3+ 1700	2+ 1840	2+ 2015	+ 2250	+ 2420	*	*	*
6	3+ 1980	" 1970	" 1270	+ 1360	0 2815			
7	2+ 1850	" 1630	" 1780	+ 3080	+ 2575			
8	" 2680	+ 1870	" 2600	+ 3420	+ 2560			

0 = <75 μM/l BAIBA approximately.
+ = 75 *idem*
2+ = 125 "
3+ = 250 "
4+ = 500 "

* No further specimen received.

event. These observations are consistent with more quantitative observation on normals by Fink *et al.*(11). There has been some variation in technics. However, we are quite confident that the amounts excreted by the irradiated human beings are significantly elevated, in spite of the semiquantitative nature of the method. Current studies on irradiated dogs showed a great increase in animals receiving 400 r whole-body irradiation appearing within 18 hours after exposure. BAIBA was not detected pre-irradiation and in normal dog urines. Investigation of the suggested dose dependency is underway in dogs.

The mechanism of production of increased BAIBA excretion is not clear. Studies by Caravaca and Grisolia(12) suggest that BAIBA may arise in part directly from sources other than thymine such as carbamyl amino acids. However, the significance of this pathway in mammals needs further evaluation. The pattern of excretion seen here follows the extensive necrosis of radio-sensitive cells of lymph nodes, bone marrow and GI tract. DNA destruction is manifested by pyknosis, phagocytosis and disappearance of large amounts of Feulgen staining material. The pathway of destruction of DNA after radiation injury is not known. It is however a potential source of urinary BAIBA after irradiation. It has been shown that intravenous H^3 thymidine in human beings and other animals is either incorporated into DNA or catabolized(14). Therefore in respect to this pyrimidine deoxyriboside there are at least 2 pathways by which it is utilized. It is believed that some of the thymidine not incorporated into DNA is promptly degraded through a series of steps to BAIBA.† Accordingly when total DNA synthesis is decreased from any cause, metabolism of thymine compounds may be shunted towards BAIBA production.

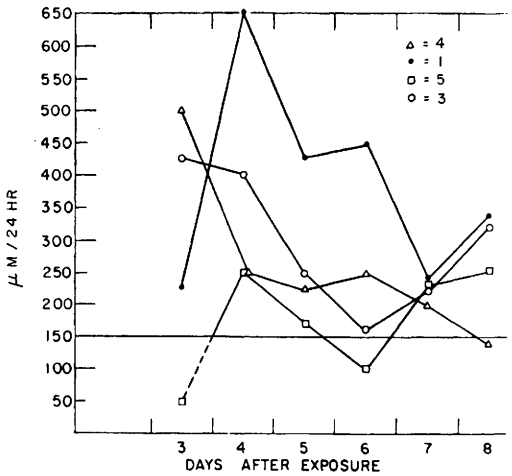

FIG. 2. Daily urinary BAIBA excretion after irradiation.

These alternatives are subject to experimental attack. If the BAIBA comes from destruction of cells by irradiation and if DNA

† Chromatography of urine from one of our patients who received intravenous H^3 thymidine was done by Dr. George Gerber at the University of Rochester, School of Medicine and Dentistry, Div. of Exp. Radiol. He has isolated and characterized tritium containing BAIBA from the urine.

in cells were labeled before irradiation with H^3 thymidine, some of the BAIBA excreted after irradiation should have a tritium label if DNA breakdown contributes BAIBA. Increases of urinary BAIBA detected following irradiation may prove useful in early detection of serious exposure.

Summary. 1. Beta-aminoisobutyric acid (BAIBA) has been found in increased amounts in urine from a group of human beings accidentally exposed to serious radiation. 2. BAIBA levels excreted appear related to the estimated doses received. 3. The implications of BAIBA excretion as related to DNA metabolism and irradiation-induced aplasia are discussed.

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Anticonvulsant Effect of Dilantin Sodium by Intravenous Administration in Mice. (24549)

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Dilantin sodium®* given intravenously has, in recent years, been advocated for treatment of status epilepticus and prevention of seizures in neurosurgery(1,2). The principal advantage of Dilantin, as contrasted to hypnotic and anesthetic agents, is high anticonvulsant potency unaccompanied by depressant effects on the respiratory mechanisms. Because the recommended dose of 250 mg of intravenous Dilantin sodium was found in some cases incapable of promptly suppressing epileptic seizures, the question has been raised as to whether this lack of prompt anticonvulsant effect could be a result of insufficiency in dosage or might be caused by some factor which delays its action on the central nervous system (personal communication). Accordingly, rate of onset and duration of anticonvulsant effect of Dilantin on electrically-induced seizures were investigated in mice at various dosages following intravenous administration.

Methods. The drug (Steri-Vial Dilantin sodium), dissolved in water, was injected *via* the marginal tail vein of male albino mice weighing 18 to 22 g. Electroshock as described by Toman, Swinyard and Goodman (3) with slight modification(4) was employed to produce convulsive seizures. The anticonvulsant effect of Dilantin was determined either (a) by finding a dose of the drug required to abolish the tonic-extensor seizures in 50% of the mice (PD_{50}) when a fixed current strength was chosen to induce convul-

* Dilantin® is trademark applied to dihpenylhydantoin by Parke, Davis & Co.