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.

1. Crumpler, H. R., Dent, C. E., Harris, H., Westall, R. G., *Nature,* 1951, v167, 307.

2. Fink, K., Henderson, R. B., Fink, R. M., PROC. SOC. EXP. BIOL. AND MED., 1951, v78, 135.

3. ———, *J. Biol. Chem.,* 1952, v197, 441.

4. Fink, K., Cline, R. E., Henderson, R. B., Fink, R. M., *ibid.,* 1956, v221, 425.

5. Altman, K. I., Haberhood, G. L., Hempelmann, L. H., *Radiation Res.,* 1956, v5, 466.

6. Kay, R. E., Barly, J. C., Entenman, C., USN NRDL-TR-98, Naval Med. Res. Inst., 1956.

7. Awapara, J., *The Leukemias, Etiology, Pathol-physiolog. and Treatment;* Henry Ford Hospital Intern. Symposium, Chap. 20, Academic Press, N. Y., 1957.

8. Brucer, M., Andrews, G., *Oak Ridge Nat. Lab. Report.*

9. Awapara, J. and Sato, Y., *Clin. Chim. Acta,* 1956, v1, 75.

10. Datta, S. P., Dent, C. E., Harris, H., *Biochem. J.,* 1950, v46, XLII.

11. Fink, K., Fink, R. M., *Fed. Proc.,* 1958, v17, 219.

12. Caravaca, J., Grisolia, S., *J. Biol. Chem.,* 1958, v231, 357.

13. Heppel, L. A., Rabinowitz, Jesse C., *Enzymology of Nucleic Acids, Purines, and Pyrimidines,* 1958, v27, 633.

14. Cronkite, E. P., Fliedner, T. M., Bond, V. P., Rubini, J. R., Brecher, G., Quastler, H., *Ann. N. Y. Acad. Sci.,* 1958.

Received October 31, 1958. P.S.E.B.M., 1959, v100.

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

CHARLES R. ENSOR, BARBARA BOHNER AND GRAHAM CHEN
Research Division, Parke, Davis & Co., Detroit, Mich.

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.

TABLE I. Anticonvulsant Effect of Dilantin Sodium by Intravenous Administration in Mice.

Current, ma	Time after dosing: 1 min.	5 min.	15 min.	60 min.
	*PD_{50} ± S.E., mg/kg intrav.			
6	13.6 ± .8	9.9 ±.6	7.8 ±.3	8.0 ±.3
9	19.6 ±1.1	15.5 ±.5	11.0 ±.2	10.4 ±.3
12	25.5 ±1.0	14.2 ±.4	14.6 ±.2	10.9 ±.3
24	25.5 ± .4	20.5 ±.7	15.5 ±.1	12.0 ±.3
48	28.0 ± .7	21.5 ±.9	17.3 ±.2	14.8 ±.4

* 50% protective dose and stand. errors.

sions, or (b) by ascertaining the current strength necessary to produce tonic-extensor seizures in 50% of the animals (CS_{50}) when a protective dose of Dilantin was given. Three groups of 10 mice each were used for the 3 doses of Dilantin (for a fixed current strength) or for the 3 current strengths (for a protective dose of Dilantin) that would give tonic-extensor convulsions in 10 to 90% of animals. PD_{50} or CS_{50} and standard errors were estimated graphically by the method of Miller and Tainter(5).

Results. Table I gives the results obtained from experiments in which convulsions in mice were produced by a fixed current strength at 6, 9, 12, 24 or 48 milliamperes. The anticonvulsant effect of Dilantin was determined at 1, 5, 15 and 60 minutes after intravenous injection. The data indicate first, as is to be expected, that the amount of Dilantin (PD_{50}) required for abolishing the tonic-extensor seizures was proportionally greater for convulsions induced by higher current strength. A significant feature is that this was also found to be the case for Dilantin one minute following intravenous administration. Second, as revealed by the lower protective doses of Dilantin from 1 to 60 minutes after injection for convulsions which were induced by the same current strengths, there was an increasing anticonvulsant effect of Dilantin during this period.

Rate of onset and duration of anticonvulsant effect of Dilantin are more clearly shown by the graphic data in Fig. 1, which was constructed with the results of another experiment by plotting current strength required to produce tonic-extensor seizures in 50% of mice (CS_{50}) against time interval after the animals had received 15 mg/kg of Dilantin sodium intravenously. It is seen that the anticonvulsant effect of Dilantin (in terms of the CS_{50} value) became increasingly greater immediately following injection, reached a peak at about 30 minutes and peak value was maintained for the next 1½ hours. A significant anticonvulsant effect was evident 8 hours after administration. Similarly, the effect of 5 mg kg of Dilantin sodium was also investigated. This is the dosage recommended for management of epilepsy in humans. The control CS_{50} was 5.00 ± 0.10 and significant increases were shown at 15 minutes, CS_{50} = 5.30 ± .09 (P = .01) and at 30 minutes (CS_{50} = 5.52 ± .12 (P = .01). No significant differences were obtained at 1 minute, 5 minutes and 1, 2, 4 and 6 hours. Apparently this is the minimal dose of Dilantin sodium required to show an anticonvulsant effect on electrically-induced seizures in mice.

Discussion. In view of the fact that Dilantin is effective in abolishing tonic-seizures in mice one minute after intravenous administration, it appears that Dilantin reaches the site of action in the central nervous system and reacts with the receptors very quickly. Since the tonic-extensor seizures induced in mice at higher current strength can be abolished by Dilantin with a corresponding increase in dosage, those cases of status epilepticus which were not promptly controlled by 250 mg of intravenous Dilantin sodium, could very likely be relieved by higher doses of the drug.

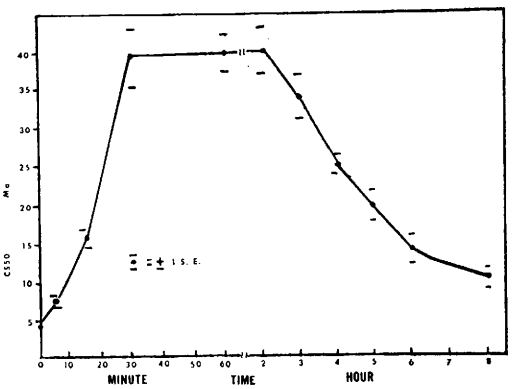

FIG. 1. Rate of onset and duration of anticonvulsant effect of a fixed dose of Dilantin sodium (15 mg/kg) following intrav. administration in mice.

The prolonged anticonvulsant effect of Dilantin shown by intravenous administration in mice, if true also in man, is certainly a very desirable property of the drug in management of status epilepticus or for prophylactic control of seizures in neurosurgery.

Summary. 1. Rate of onset and duration of anticonvulsant effect of intravenous Dilantin sodium against electrically-induced tonic-extensor seizures have been investigated in mice. At 15 mg/kg, the anti-convulsant effect of Dilantin reached a peak approximately 30 minutes after injection, and was maintained for 1½ hours; it then began to decline gradually, persisting more than 8 hours. 2. Dilantin, one minute after intravenous injection, was capable of abolishing the tonic-extensor seizures in mice at doses in proportion to the current strengths employed to produce convulsions.

1. Schwab, R. S., *Rhode Island M. J.*, 1953, v36, 574.

2. Murphy, J. T., Schwab, R. S., *J.A.M.A.*, 1956, v160, 385.

3. Toman, J. E. P., Swinyard, E. A., Goodman, L. S., *J. Neurophysiol.*, 1946, v9, 231.

4. Chen, G., Ensor, C. R., *Arch. Neurol. Psychiat.*, 1950, v63, 56.

5. Miller, L. C., Tainter, M. L., PROC. SOC. EXP. BIOL. AND MED., 1944, v57, 261.

Received November 3, 1958. P.S.E.B.M., 1959, v100.

Canine Hepatitis Virus: Attempts to Find Relationship with Human Hepatitis.*† (24550)

VIGGO BECH‡ (Introduced by R. H. Green)

Section of Epidemiology and Preventive Medicine, Yale University School of Medicine

Two groups of investigators(2,8) described successful isolation and propagation of canine hepatitis virus in cell cultures of dog kidney. Since then the growth of this virus in cell cultures of kidney tissue from other animals has been reported(5,9). Fastier(6,7) described agglutination of fowl erythrocytes by this virus using infected tissue culture fluids as antigens. This agglutination was inhibited by immune sera from dogs naturally or experimentally infected with canine hepatitis virus. Although there is no epidemiologic evidence to suggest that the infectious agent of human hepatitis is the same as that of canine hepatitis, the possibility of some immunologic relationship between the two was felt to warrant exploratory investigation using recently developed methods.

Materials and methods. Virus. A strain of canine hepatitis virus, which had been passed 37 times in dog kidney tissue cultures, was kindly supplied by Dr. V. J. Cabasso, Lederle Medical Research Dept. *Cell cultures.* Kidneys from young puppies were trypsinized according to the method of Rappaport(16). Cultures were prepared by seeding each tube with about 300,000 cells contained in 1 ml growth medium. In the same way cultures were prepared from rhesus monkey and young pig kidneys. The growth medium consisted of 0.5% lactalbumin hydrolysate in Hanks' balanced salt solution (BSS)(13). For dog and pig kidney cell cultures this was supplemented with 10% calf serum; for rhesus kidney cell cultures with 2% calf serum. From continuous cell lines§ of human origin: Detroit-6(1);

* Work supported by Office of Surgeon General, Dept. of Army.

† I am greatly indebted to Dr. Leon Whitney and Dr. George D. Whitney, who generously donated the dogs and kept and examined them during experiments. I am also indebted to Dr. Robert W. McCollum for assistance.

‡ Visiting scientist, Yale Univ., Present address: State Serum Inst., Copenhagen, Denmark.

§ Original cell lines were kindly supplied by: Drs. I. W. McLean and W. A. Rightsel, Parke Davis and Co., Detroit, Mich., (Detroit-6 cells); Dr. Alice E. Moore, Sloan-Kettering Inst., N. Y., (Hep-2 cells); Dr. Jørgen Fogh, Virus Laboratory, Berkeley, Calif., (FL cells).