therefore, antinicotinic properties which are lacking almost entirely in atropine.

1. Kunkel, A. M., Wills, J. H., and Monier, J. S., PROC. SOC. EXP. BIOL. AND MED., 1956, v92, 529.

2. Miller, Lloyd C., and Tainter, M. L., *ibid.*, 1944, v57, 261.

3. Shea, S. M., *Brit. J. Pharmacol. Chemotherap.*, 1956, v11, 171.

Received September 17, 1957. P.S.E.B.M., 1957, v96.

Amino Aciduria in Primates Following Irradiation.* (23611)

C. G. HUNTER (Introduced by C. H. Best)
(With the technical assistance of J. G. Scott†)
Department of Physiological Hygiene, School of Hygiene, University of Toronto

The acute radiation syndrome presents a symptom complex that is dose dependent. In the lower lethal dose range for the primate, the metabolic damage is expressed in 2 phases, an initial phase of predominantly alimentary dysfunction and a later phase associated with the catabolism of body tissues. In both phases, nitrogen balance becomes negative, but little change in the total daily urinary nitrogen excretion takes place until as an antemortem event, the daily urine nitrogen excretion may be greater than during a pre-radiation control day.‡ Some qualitative change in the nitrogen partition occurs and amino acid excretion is increased. This increase has been stated to be due to the radiation injury sustained by the animal(1). This paper presents a study of the urine amino acid nitrogen excretion of 4 whole body irradiated monkeys and of the 2 survivors (after physiological recovery) when sham irradiated and given the same dietary intake as in their radiation experiment.

Material and method. Adult male imported Rhesus monkeys (*M. mulatta*), tuberculin tested and separately housed, were placed in a metabolic cage. Their body weights varied from 6.8-12.4 kg and they were in robust health. They were fed by hand on Parkes "41" rat cubes and hand watered, so an excellent estimation of food and water intakes was possible. After a control period, the animals were whole body irradiated or sham irradiated and the urine amino acid nitrogen (amino N) and total nitrogen (total N) excretion followed until death or obvious recovery. The urines, voided into flasks with toluene as a preservative, were collected at the same time each day and analyzed for amino N by the copper titration method(2), and for total N by a semi micro method(3). Several urines were subjected to paper chromatography(4). *Radiation factors.* One animal was exposed to X irradiation, 240 Kv, HVL. 1.2 mm Cu with stepped Cu/Al filter, F.S.D. 150 cm, dose-rate 9 r/min. and the other 3 to gamma radiation (Co^{60}, 1.3, 1.1. Mev.) dose-rates 13-15 and 103-105 r/min. With both the X and gamma irradiation, the radiation procedure was arranged so that the dose received was relatively homogenous throughout the animal (5). Measurement of dose-rate was made with a Victoreen 100 r dose meter at various selected positions that the animal would subsequently assume. Thiopentone anaesthesia was given for the X irradiation(6).

Results. The daily urine amino N excretion of the 4 irradiated animals is shown in Table I. Since amino N is but a part of the total urinary N excretion, total N excretion is recorded as well. The values for the 4 animals during the control period were in close agreement, with the exception of monkey C, a smaller animal.

Immediately following radiation, only monkey C showed an amino N excretion greater

* Experiments carried out at the M.R.C. Radiobiological Research Unit, A.E.R.E., Harwell, England.

† Dept. of Biochemistry, University of Western Australia, Perth, Australia.

‡ Unpublished observations, Hunter, C. G.

TABLE I. Daily Urine Amino N and Total N (in Parentheses) Excretion, g/day, in 4 Monkeys after a Single Exposure. Animal G—two exposures to whole body X and gamma irradiation in lower lethal dose range for the primate.

	Non-survivors		Survivors	
Monkey	C	G	A	E
Dose	500 r x	460 r γ	480 r γ	480 r γ
Pre-irrad. (No. days)	6	5	5	4
Mean	.02 (1.98)	.092 (4.37)	.094 (4.42)	.078 (2.94)
Range	.017–.022 (1.52–2.48)	.064–.124 (4.142–4.645)	.079–.113 (4.24–4.52)	.072–.084 (2.64–3.23)
Days post-irrad.				
1	.038 (2.54)	.08 (2.90)	.079 (3.60)	.072 (2.17)
2	.031 (1.10)	.076 (2.88)	.097 (3.89)	.08 (2.10)
3	.03 (2.58)	.076 (2.60)	.104 (2.88)	.089 (1.70)
4	.034 (2.16)	.072 (2.58)	.113 (2.87)	.079 (1.23)
5	.022 (2.37)	.088 (2.26)	.113 (2.87)	.075 (1.15)
6	.028 (3.48)	.065 (2.28)	.087 (2.27)	.084 (1.99)
7	.08 (3.90)	.088 (2.04)	.085 (2.13)	.111 (1.109)
8	.08 (3.81)	.119*(3.31)	.064 (1.49)	.104 (1.15)
9	.053 (1.76)	.16 (3.20)	.083 (1.898)	.112 (1.65)
10	.042 (3.28)	.163 (3.88)	.083 (3.20)	.123 (1.58)
11		.098 (2.55)	.099 (2.48)	.154 (2.40)
12		.103 (3.14)	.098 (2.49)	.133 (2.06)
13			.089 (2.80)	.154 (2.62)
14			.088 (2.79)	.10 (3.31)
15			.094 (2.52)	.065 (3.22)
16			.072 (3.40)	.077 (3.70)

* Day 1 after 2nd irradiation.

than in the control period. By the end of the first week all values were within the pre-irradiation range, but the amino acid index $\left(\frac{\text{amino N}}{\text{total N}} \times 100\right)$ had increased slightly. With the onset of severe terminal catabolism in animal C and animal G (after the second exposure), the amino N values were higher than during the control period. One survivor only, animal E, excreted significantly more amino N than before irradiation and this occurred during N retention at the end of the second week.

Table II records the results of the sham irradiation experiment. The mean control values of both animals were slightly higher, perhaps associated with an increase in weight since irradiation (12 months previously). With the identical dietary intake as in their irradiation study, monkey A lost more amino N and total N than after irradiation. Monkey E on the other hand, gave an opposite result losing more amino N and total N after irradiation.

Two dimensional chromatograms were run on the urine of monkey A. Normal monkey urines were found to contain 2 amino acid groupings: glutamic acid/glycine and taurine/glutamine in descending order. After irradiation slight quantitative increase was demonstrated on days 4 and 5, but the only

TABLE II. Daily Urine Amino N and Total N (in parentheses) Excretion, g/day, in 2 Monkeys Sham Irradiated and Given the Same Dietary Intakes of Their Irradiation Experiment.

	Monkey A	Monkey E
Days pre-sham irrad.	3	3
Mean	.112 (3.77)	.12 (3.61)
Range	.104–.12 (3.46–4.10)	.103–.154 (3.36–4.00)
Days post-sham irrad.		
1	.112 (3.28)	.076 (3.80)
2	.09 (2.80)	.078 (2.23)
3	.091 (2.80)	.068 (1.94)
4	.112 (3.62)	.065 (2.82)
5	.114 (3.42)	.073 (2.21)
6	.084 (2.10)	.074 (1.65)
7	.057 (3.68)	.067 (1.55)
8	.096 (3.06)	.076 (2.82)
9	.104 (2.65)	.053 (2.42)
10	.122 (3.20)	.06 (2.503)
11	.122 (3.20)	.072 (2.78)
12	.126 (3.88)	
13	.127 (4.56)	
14	.13 (4.84)	

qualitative difference was the appearance of small amounts of leucine for 9-10 days.

Discussion. Amino aciduria has been described in man following accidental over-exposure to ionising radiation(1,7). Two lethally irradiated men excreted increased quantities of urine amino acids during the first week, the peak excretion being on the sixth day, when values about 3 times the normal were excreted. Three other males and one female, much less severely irradiated, also showed increases during the first week. In these reports urine amino acid excretion was estimated by paper chromatography and the pattern of the amino acids changed from the four acids, glycine, taurine, serine and alanine, excreted in normal human urine to some 14-15 in the irradiated. In spite of the paucity of data given in these reports, radiation injury appeared to increase urinary amino acid excretion, the cause for this increase not being determined. It was obvious that the increase appeared quickly after the exposure being detected on the first day although the amount excreted was not proportional to the dose received.

Amino aciduria is a sign of hepatic and renal injury, but there is no evidence that radiation injury in doses given in this study affects the function of the liver and kidney (1,4,7). In primate radiation injury it has been suggested that the amino aciduria may be due to failure of peripheral utilization of amino acids(7), but since little difference exists between the amino acid excretion in the irradiated monkey and the monkey fed identically with its irradiated self, dietary intake must be a factor as well. Very little evidence can be adduced that radiation injury *per se* in the lower lethal dose range causes the reported amino aciduria. So the increased values of urinary amino acids should be regarded properly as a reflection of the aberration of protein metabolism usually associated with the response of the healthy primate to fevers, infections and trauma(8,9).

Summary. The daily urine amino N excretion of 4 monkeys was followed before and after whole body irradiation with doses in the lower lethal range. In 2 non-survivors, there was little change in the daily quantity excreted until terminally. In 2 survivors given the same food intakes as in the irradiated study and sham irradiated, the daily urine amino N excretion during the first week differed but slightly from the values after irradiation, but in the second week more amino N was excreted after irradiation in one animal and less in the other. It would appear that amino aciduria in primates irradiated with doses in the lower lethal range is inseparable from the natural response of the overall protein metabolism associated with any injury.

1. Hempelman, L. H., Lisco, H., and Hoffman, J. G., *Ann. Int. Med.,* 1952, v36, 279.

2. Albanese, A. A., and Irby, V., *J. Biol. Chem.,* 1944, v153, 583.

3. Fawcett, J. K., *J. Med. Lab. Tech.,* 1954, v12, 1.

4. Dent, C. E., *Biochem. J.* (Proc.), 1946, v40, 44.

5. Corp, M. J., *Physics in Med. and Biol.,* 1957, v1, 377.

6. Haig, M. V., and Paterson, E., *Brit. Radiol.,* 1956, v29, 339.

7. Hasterlik, R. J., and Marinelli, L. D., *Biological Effects of Radiation UNO, Peaceful Uses of Atomic Energy,* 1956, p25.

8. Peters, J. P., and VanSlyke, D. D., *Quantitative Clinical Chemistry,* 2nd ed., 1946, v1, pp678, 809.

9. Wright, S., *Applied Physiology* IX ed., Oxford Medical Publications, London, 1952, p891.

Received September 19, 1957. P.S.E.B.M., 1957, v96.