Short Communications

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Excretion of 1-aminocyclopentanecarboxylic acid in man and the rat

Recently we reported that 1-aminocyclopentanecarboxylic acid is almost completely reabsorbed by the kidney of the rat, so that only about 1 % of that present in the body is excreted in the urine per day[1]. The present study shows first-order kinetics for its urinary excretion in man and the rat, with half-times of 3.5 and 22 days, respectively. In the latter species about two-thirds of the dose, unexpectedly, is excreted into the large intestine. The results show that in both species a very adequate interval is available for the study of humoral or pathologic influences on the distribution and transport of this model amino acid.

Two men, ages 33 and 46, received in the deltoid muscle 1 and 2 mg, respectively ($16 \cdot 10^6$ and $32 \cdot 10^6$ counts/min) of carboxyl-labeled 1-aminocyclopentanecarboxylic acid[2] in aq. 0.9 % NaCl. Urine was collected in daily portions, and blood plasma collected at the end of each 24-h interval, until the radioactivities became rather low for accurate counting. The plasma was treated with 0.075 vol. of aq. trichloroacetic acid (100 g/100 ml). On removal of the precipitate the radioactivity of the filtrate was counted as previously described[1]. The findings are shown in Fig. 1.

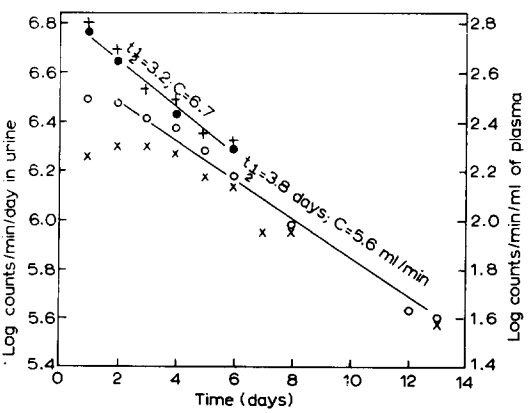

Fig. 1. Decline in the logarithm of plasma level (round points) and of the daily urinary-excretion rate (crosses) of injected 1-amino[1-^{14}C]cyclopentanecarboxylic acid in man.

For the person shown in the upper curve, the plasma level and the urinary-excretion rates declined in good correspondence with first-order kinetics. The second subject showed a similar behavior after the first 2 days. The relationships between the plasma and urinary levels represent renal clearances of 6.7 and 5.6 ml plasma per min. The two slopes correspond to half-times of 3.2 and 3.8 days. In the first 3 days the first subject had excreted in the urine 47 % of the injected dose, in a week 76 % of the injected dose, in good agreement with the decay curves. The corre-

spondence was similar in the other case. Accordingly excretion by other routes in man must be less than the analytical error of not over 5 %.

If we were to assume that all of the amino acid present in each human subject was contained in a solution having the concentration found for the plasma, these solutions would have volumes of 48 and 56 l, respectively, values that are very close to 60 % of the body weights of the subjects expressed in kilograms. Therefore we may assume that the amino acid is not concentrated into any tissue as abundant as muscle, and if it is concentrated into such organs as the liver, it must to a compensating degree be excluded from other portions of the body water. Greater overall accumulation occurs for α-aminoisobutyric acid into human tissues[3]. In the mouse in the course of 6 h the present test amino acid was concentrated by 2–10 times into liver, and by 1.2 times into skeletal muscle[1].

Male rats of about 150 g were injected subcutaneously in the scapular region with known amounts ($6.6 \cdot 10^6$ counts/min, either 0.5 or 10.5 mg) of the test amino acid. The daily urine output was then collected in a metabolism cage at regular intervals for 91 days. The collecting surface was washed down with hot water at the end of each collection interval, and the radioactivity determined by liquid scintillation counting, as described before[1].

Fig. 2 shows the decline in the logarithm of daily urinary excretion in the male rat, representing a half-life of 22 days through a 91-day interval. During this period, however, only one-third, rather than the predicted 94 % of the radioactivity had appeared in the urine. A similar discrepancy was seen in the mouse. Since volatile ^{14}C is not released into the environment of rats or mice receiving the labeled amino acid[1], the amino acid must either have been excreted by another route, or remain in the tissues. Analysis of liver and muscle showed that the tissue radioactivity had declined in good correspondence with the fall in the urinary-excretion rate. Accordingly the

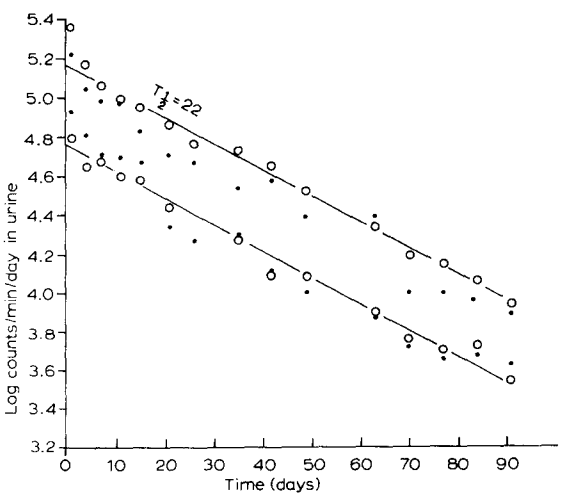

Fig. 2. Decline in the daily urinary-excretion rate of 1-aminocyclopentanecarboxylic acid injected into rats. The upper circles, describing the upper line, represent the urine of two rats receiving together 1.0 mg or $13.1 \cdot 10^6$ counts/min. The small points below this line represent the results with two rats receiving together 21 mg of $13.1 \cdot 10^6$ counts/min. The lower circles and points have the same significance, except that they refer to single animals receiving similar doses per animal.

feces were examined for ^{14}C, even though this route of excretion was insignificant for α-aminoisobutyric acid[4]. The animals were picked up at hourly intervals and exposed to ether vapors to cause urination, so that feces could be collected without contact with urine. The samples were extracted exhaustively with boiling water, and the extracts centrifuged after adding a little conc. trichloroacetic acid. The extract was added in 0.2-ml portions to the scintillator solution for counting as usual. During two 8-h intervals on the third and fourth days after injection, fecal radioactivity in 2 rats represented the daily excretion of 1.4 to 2.2 % (average, 1.7 %) of the injected dose. This excretion rate was double the urinary rate and hence accounts adequately for the radioactivity not found in the urine. During the preparation of this report, a paper by STERLING *et al.*[5] appeared, reporting the predominance of the fecal route in the slow elimination of the amino acid by the rat.

To determine the source of the fecal radioactivity we ligated the intestine in 3 rats at the ileosecal junction, just before injecting the labeled amino acid subcutaneously. During the next 24 h the contents of the large intestine came to contain 0.6–0.8 % of the injected radioactivity; a very much lower content was found in the small intestine above the ligature. Accordingly much of the excretion appears to be performed by the large intestine. Rather little of the amino acid appeared in the bile, and ligation of the bile duct did not interfere with excretion by the intestinal route. Because the amino acid is absorbed vigorously by the small intestine[2] one would not expect secretion into the upper tract to contribute much to the loss from the animal.

Renal tubular reabsorption of the test amino acid in the rat appeared to be nearly as complete at a dose of 70 mg as at a dose of 3.5 mg/kg body weight, since the excretion was only moderately accelerated at the higher dose (Fig. 2). Hence the agency mediating reabsorption was not yet saturated at this dosage. Transport of the amino acid probably occurs by a process serving for various neutral amino acids without distinguishing sharply among them. Therefore the effective load of amino acid handled by the renal process, as recorded by the tracer, was probably substantially larger than we might calculate for the test amino acid alone, and not greatly different at the two dosages. RIGGS AND WALKER[6] have shown that the distribution of α-aminoisobutyric acid in the rat is virtually independent of the dose, until the dose is large enough to increase substantially the plasma total amino acid level.

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Department of Biological Chemistry, HALVOR N. CHRISTENSEN
The University of Michigan, Ann Arbor, Mich. (U.S.A.) JANE A. CLIFFORD

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