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DIRECT COUNTING OF CALCIUM-47 AND STRONTIUM-85 IN BIOLOGICAL SAMPLES

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The radioactivity of ^{47}Ca and ^{85}Sr was determined before and after thermal ashing of various biological specimens (carcass, skeleton, urine and fæces of rats). Using a two scintillation crystal assembly and making adequate adjustments to the volume of standards, the percentage of radioactivity retained in the body and excreted in urine and fæces was found to be the same (within statistical fluctuations) for both ashed and non-ashed specimens after single intraperitoneal, single oral and continous oral administration of the tracers.

It is well known that the measuring geometry, irregularity in shape and size of the sample, internal absorption and scattering properties of sample material have, or may have, an effect on the results of measurements in gamma-scintillation spectrometry (1). It is therefore general practice to ash the inhomogenous or irregularly shaped biological specimens in order to obtain optimal conditions i. e. samples identical in every detail except for their radioactivity. This is a long and tedious procedure, which sometimes involves losses of biological material. We have therefore attempted to measure radioactivity of various biological samples by a direct counting method, without any previous treatment of the samples. The results obtained by direct measurements were compared with the data evaluated on the same but ashed specimens. The purpose of this comparison was to obtain a quantitative picture of errors due to sample irregularities. The magnitude of this uncertainty also depends on the type of the counting system (2). In all direct counting measurements we therefore used a two scintillation crystal assembly in which a wide range of sample volumes can be measured with a fairly uniform counting efficiency.

The radioactivity of whole body or carcass and excreta of rats was measured after single intraperitoneal, single oral and continuous oral

administration of ^{85}Sr and ^{47}Ca in order to evaluate the possibility of using the direct counting method in different types of animal tracer experiments.

METHODS

For direct measurements we used a commercially available two-crystal assembly (Nuclear Chicago Corporation, »Tobor«). It is shielded from background radiation by a three-inch thick lead shield, encased in a quarter of an inch steel shell. The summed outputs of the two vertically opposed adjustable 3" × 3" cylindrical NaI(Tl) scintillation crystals were fed to a single-channel pulse height analyzer. In all measurements the two crystals were kept at the maximum distance so that the aluminum sample platform provided for positioning of the samples in the counting chamber was at 10,7 cm distance from the face of the upper crystal and at 4.6 cm distance from the lower crystal.

The experiments were performed on 8–9 week old female rats of on average weight of 150 g. Rats were kept in metabolic cages throughout the experiment and separate collections of urine and faeces were made from each animal. All animals were sacrificed at the end of the experiment by an overdose of ether.

For radioactivity measurements the whole body or carcass (body without the gastrointestinal tract) of the rat was put into an aluminum cage and placed on the sample platform between the two crystals. The urine samples were left in original containers and adjusted to the same volume by addition of water. Faecal samples collected in small polythene bags were spread on the sample platform and measured between the two crystals.

We measured ^{47}Ca by using the photopeak of its highest energy-gamma-ray i. e. 1.31 MeV within an energy interval of 170 KeV. ^{85}Sr was assayed by totalling the counts in a 102 KeV energy band centered on the 0.514 MeV photopeak and substracting therefrom the ^{47}Ca contribution in this range. Under these conditions the efficiency for ^{47}Ca was 1.5% and for ^{85}Sr 4%.

After direct measurements the specimens were ashed and the radioactivity of the samples determined in the usual way. The carcass or the skeleton was thermally ashed at 800 °C for 18 hrs. The ashed carcass or skeleton was dissolved in warm concentrated hydrochloric acid and made up to volume with distilled water. Faecal and urine samples were dried and ashed in a muffle furnace at 550 °C for 38 hrs. The radioactivity of the samples was determined in a 3" × 3" NaI(Tl) well-type gamma scintillation crystal, coupled to a single-channel pulse height analyzer.

RESULTS AND DISCUSSION

All results are expressed as percentage of the administered dose. Each figure in the tables represents the arithmetic mean of a group of animals and the probable error of the mean.

In Table 1 data obtained after a single intraperitoneal administration of ^{85}Sr are presented. The retention of ^{85}Sr was determined 72 hours after the application of the radioisotope, in whole animals and ashed skeletons. For direct counting the initial whole body radioactivity measured immediatelly after the intraperitoneal injection was considered as the 100% retention value. The movement of the animal within the cage during the counting was not found to affect the results of measurements. For ashed samples the administered dose diluted to a known volume was used as a 100% standard. Results show a good agreement between the data obtained in non-ashed and ashed specimens.

Table 1.

^{85}Sr retention in the whole-body of rats as compared to values obtained on the corresponding ashed skeletal samples 72 hours after intraperitoneal administration

Group	Number of rats	Percent retention	
		Whole-body (non-ashed)	Skeleton (ashed)
1	9	39.41 ± 0.88	39.04 ± 1.00
2	9	24.81 ± 1.09	23.54 ± 1.12

In Table 2 are presented mean retention and excretion values of ^{47}Ca and ^{85}Sr 72 hours after a single oral administration of the two radioisotopes by stomach tube. For direct counting of the carcass the retention value was determined by taking as 100% either the whole-body counting rate measured immediately after radioisotope application or the counting rate of the standard i.e. administered dose diluted up to 100 ml. For direct measurements of faeces we used a 25 ml standard in a 100 cm diameter and 1.5 cm high petri dish, whereas for urine the standard (adjusted to the volume of urine samples) was measured in the original urine containers.

Data obtained for non-ashed and ashed specimens of the carcass, urine and faeces were in good agreement.

The retention values of non-ashed carcass samples determined in the two ways were very similar, indicating that a 100 ml radioactive solution phantom may well substitute the initial whole body counting rate in rats.

Table 2

Mean values of ^{47}Ca and ^{85}Sr in ashed and non-ashed specimens of carcass, urine and fæces 72 hours after a single oral administration

Group	No. of rats	Percent retention carcass		Percent excretion urine		Percent excretion fæces		Percent recovery (carcass+urine+fæces)	
		non-ashed	ashed	non-ashed	ashed	non-ashed	ashed	non-ashed	ashed
		^{47}Calcium							
1	8	51.39 ± 1.53[1] 49.78 ± 1.77[2]	51.34 ± 1.65	1.45 ± 0.20	1.23 ± 0.16	45.98 ± 1.60	44.04 ± 1.48	98.82 ± 2.22	96.61 ± 2.35
2	8	46.15 ± 1.59 43.26 ± 1.68	44.53 ± 1.69	3.24 ± 0.31	2.78 ± 0.34	48.82 ± 1.98	46.68±1.83	98.21 ± 2.54	93.99 ± 2.51
3	8	53.58 ± 1.73 46.46 ± 2.47	52.62 ± 1.86	1.50 ± 0.16	1.39 ± 0.14	50.90 ± 3.02	47.31 ± 3.20	105.98 ± 3.47	101.38 ± 3.69
4	8	19.57 ± 1.23 20.89 ± 1.88	18.98 ± 1.26	0.73 ± 0.03	0.65 ± 0.02	79.09 ± 1.19	76.53 ± 1.28	99.40 ± 1.72	96.16 ± 1.80
		^{85}Strontium							
1	8	14.90 ± 0.83 14.42 ± 0.80	13.55 ± 2.11	2.68 ± 0.20	2.14 ± 0.19	83.30 ± 1.06	79.45 ± 0.95	100.88 ± 1.36	95.14 ± 2.33
2	8	4.13 ± 0.70 4.20 ± 0.14	3.91 ± 0.18	1.61 ± 0.06	1.37 ± 0.05	89.93 ± 1.37	88.39 ± 1.82	95.67 ± 1.48	93.67 ± 1.83
3	8	7.01 ± 0.58 8.22 ± 0.48	6.98 ± 0.65	2.39 ± 0.21	2.20 ± 0.17	91.79 ± 1.16	87.15 ± 1.16	101.19 ± 1.31	95.33 ± 1.34
4	8	7.01 ± 0.44 6.72 ± 0.47	6.22 ± 0.41	1.74 ± 0.20	1.26 ± 0.16	89.94 ± 0.91	85.93 ± 0.96	98.69 ± 1.08	93.41 ± 1.06

For direct counting of the carcass the retention value in each group of animals was determined by taking as 100 percent radioactivity either the whole-body counting rate of rats immediately after the radioisotope application (1), or the counting rate of the standard i. e. administered dose diluted up to 100 ml (2).

In Table 3 the results obtained after continuous two-day oral application of ^{85}Sr and ^{47}Ca in drinking water are given. Retention values of the carcass determined by direct counting were evaluated by using a 100 ml radioactive solution phantom. The 100% retention value for each rat was obtained by multiplying the counts/min/ml of the phantom solution by the number of milliliters consumed by the animal during the experiment.

Table 3.

Retention of ^{47}Ca and ^{85}Sr in ashed and non-ashed specimens of the carcass after continuous oral administration of the radioisotopes for 2 days

Group	Number of rats	Percent retention in carcass			
		^{47}Ca		^{85}Sr	
		non-ashed	ashed	non-ashed	ashed
1	9	36.23 ± 1.01	35.44 ± 1.69	11.63 ± 0.66	11.52 ± 0.45
2	9	33.31 ± 0.71	32.67 ± 0.50	5.16 ± 0.51	5.29 ± 0.18

The results obtained by the two methods seem to be equally reliable. The results of measurements on ashed samples show lower values than those obtained by direct counting. This is probably due to the fact that some losses are unavoidable during the process of preparation of the specimens for the counting.

References

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Sadržaj

DIREKTNO ODREĐIVANJE KALCIJA-47 I STRONCIJA-85 U BIOLOŠKIM UZORCIMA

Određivali smo radioaktivnost kalcija-47 i stroncija-85 u mineraliziranim i nemineraliziranim uzorcima biološkog materijala (tijelo, skelet, urin i fekalije štakora). Upotrebom uređaja sa dva kristalna scintilacijska detektora, uz adekvatno odabran volumen standarda, postotak radioaktivnosti u tijelu, urinu i fekalijama praktički je isti za mineralizirane i nemineralizirane uzorke. Pokusi su vršcni intraperitonealnom, jednokratnom oralnom i kontinuiranom oralnom primjenom radioaktivnog kalcija i stroncija u štakora.

Direktna metoda određivanja radioaktivnosti u biološkim uzorcima ima velike prednosti, jer se pri primjeni takve tehnike znatno skraćuje vrijeme potrebno za dobivanje rezultata i uklanjaju eventualne pogreške do kojih dolazi prilikom spaljivanja i otapanja uzoraka.

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