RAPID ACTIVATION ANALYSIS OF TRACE VANADIUM IN TISSUE USING 3.8-MINUTE VANADIUM-52

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(Received January 7th, 1963)

The determination of trace quantities of vanadium has been handicapped by insensitive methods of analysis. Most of the recent methods are based on the formation of a color complex and quantitative spectrophotometric determination. Activation analysis, however, which holds great promise for trace analysis of biological systems,[1] offers a procedure which is sensitive to submicrogram amounts of vanadium. This is especially helpful in biological analysis where vanadium is present in amounts undetectable by standard procedures.

Because the facilities available at this laboratory permitted short irradiations in the Ford Nuclear Reactor of the University of Michigan, rapid chemical separation in the hot lab area, and continuous γ-ray spectral analysis with a 100-channel analyzer, considerable attention has been focused on short-lived radioisotopes. Utilizing activation analysis procedures with radiochemistry, BROWNLEE AND MEINKE[2] have determined the vanadium content in crude oils, while FUKAI AND MEINKE[3] analyzed ashes of marine organisms for their vanadium content. BENSON[4] has applied this technique to vanadium compounds pre-separated on chromatographic paper, but in doing so has acquired the problems of reagent blanks which are usually eliminated in activation analysis.

Spectroscopic investigations conducted by GUELBENZU *et al.*[5] placed the vanadium content of rat livers below the microgram level. The purpose of this investigation was to establish a lower limit for vanadium content in rat liver tissue or to determine it at the submicrogram level.

EXPERIMENTAL

Apparatus

Samples were irradiated in polyethylene screw-cap rabbits in the pneumatic tube system of the Ford Nuclear Reactor of the University of Michigan. This system permitted irradiations at thermal neutron fluxes of about 10^{12} n/cm^2/sec (when the reactor was operating at full power of 1 megawatt) and delivery to a hood in the neighboring Michigan Memorial Phoenix Laboratory within 3 sec after the end of irradiation. Samples were then worked up chemically and were measured by a 3" × 3" NaI(Tl) crystal coupled with a special 100-channel pulse-height analyzer with duplicate memories. This equipment has been described in detail elsewhere[6-8].

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Preparation of animals and tissue

Holtzman* albino male rats weighing between 350 and 400 g were used in this study. They were maintained on Rockland Rat Diet** and had free access to drinking water. The livers were surgically removed, weighed, allowed to air-dry at room temperature for 24 h and then reweighed. They were placed in envelopes prepared from 0.1 mm thick polyethylene film, which were closed by heat sealing. The sealed sample was then irradiated in a "rabbit" along with suitable monitoring foils for a period of 10 min at full power.

Radiochemical separation

While the sample was being irradiated a nickel crucible was prepared containing vanadium-48 tracer, 10 mg of vanadium carrier, and 10 mg of copper holdback carrier. Three sodium hydroxide pellets were added and the solution was heated almost to dryness. Two minutes before the end of the irradiation, 10 g of sodium peroxide were added to the crucible and melted.

The irradiated sample was then fused in this melt for 1 min (CAUTION). A cover must be used on the crucible since the reaction may be quite violent. The outside of the crucible was then cooled by dropping it into a beaker of cold water and the melt made to solidify in a thin readily dissolvable coating by manipulation of the crucible. The melt was then dissolved by immersion into 50 ml of water followed by the addition of 42 ml of concentrated hydrochloric acid. Ten grams of tartaric acid were added and H_2S gas was bubbled into the solution. The solution was filtered and 10 ml of cupferron solution (6% aqueous) were added to the filtrate. This filtrate was extracted with 10 ml of chloroform for 1 min. The organic layer was collected and counted with the 100-

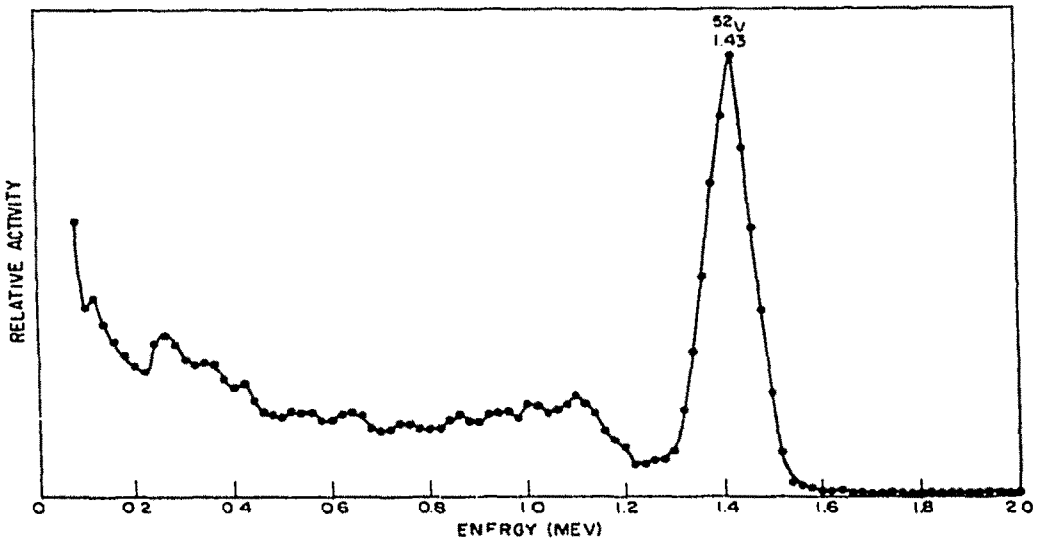

Fig. 1. γ-Ray spectrum of vanadium fraction separated from rat liver tissue (5 min after removal from reactor).

* Holtzman Company, Madison 4, Wisconsin.

** A product of Rockland Farms, New City, N.Y.; manufactured by A.E. Staley Manufacturing Company, Chicago 27, Illinois.

channel analyzer. The entire procedure could be completed in 5 min with an average vanadium recovery of about 40–45%.

Activity determination

Linearity of the measurement system was established by the use of ^{137}Cs and ^{60}Co standards. Spectra were obtained in the 0–2 MeV energy range as shown in Figs. 1 and 2. The amount of vanadium-52 was determined from the area under the 1.43 MeV photopeak, while the correction for chemical yield was made by measuring recovery of long-lived tracer vanadium-48 utilizing the 0.99 MeV photopeak after the shorter-lived peak had decayed out.

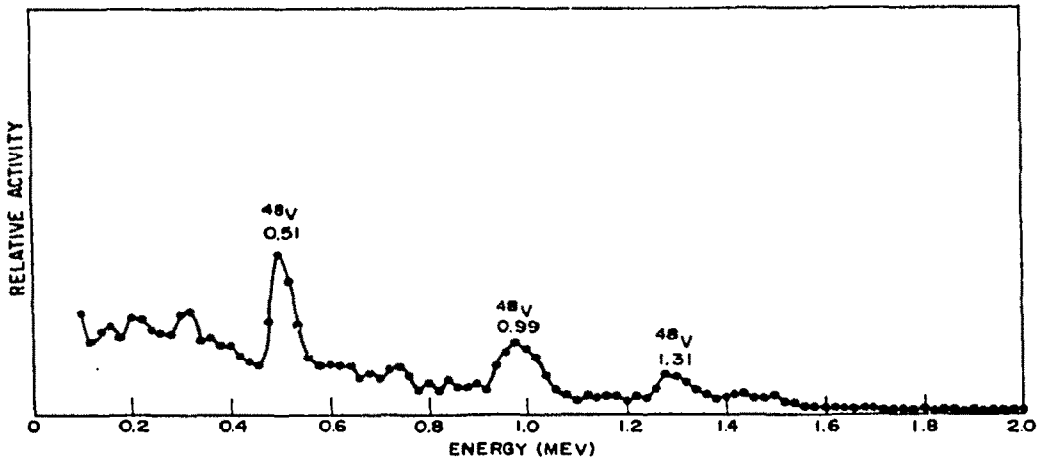

Fig. 2. γ-Ray spectrum of vanadium fraction separated from rat liver tissue (40 min after removal from reactor).

Monitoring procedures

Gold foils weighing between 0.2 and 0.3 mg were wrapped in plastic envelopes and taped to the inside of the cap of the rabbit. Following irradiation the foils were dissolved in 4 ml of aqua regia and diluted to 10 ml with distilled water. An aliquot of this solution was placed in a test tube and counted with a well-type scintillation counter. Comparison with other measurements made with calibrated gold foils permitted normalization of all irradiations to a neutron flux of $1 \cdot 10^{12}$.

RESULTS AND DISCUSSION

By this procedure, values for the normally occurring vanadium concentrations in rat liver tissue were obtained, as shown in Table I. Although these values show a variation of greater than a factor of two, they do establish an order of magnitude.

Most spectroscopic procedures are not sufficiently sensitive to detect these levels in complex biological systems. Utilization of activation analysis, *i.e.*, the short-lived vanadium-52, reduces the possibility of prolonged radioactive contamination, provides a sensitive analytical method and allows rapid analysis of numerous samples. The use of long-lived vanadium-48 for calculating chemical yield offers the added advantage of one procedure yielding two results, *e.g.*, amount of vanadium-52 and yield

TABLE I

ACTIVATION ANALYSIS FOR VANADIUM IN RAT LIVER TISSUE USING VANADIUM-52

Animal number	*Life weight (g)*	*Fresh liver weight (g)*	*Yield of separation procedure (%)*	*Vanadium found in livers 10^{-8} g/g fresh tissue*[a]
1	358.5	11.61	38.7	3.2
2	378.0	14.04	45.4	1.4
3	378.4	11.81	40.1	2.8
4	367.3	13.15	46.9	3.1
5	324.1	12.43	42.4	2.0

[a] Corrected to neutron flux of $1 \cdot 10^{12}$ n/cm²/sec.

correction from the isolated vanadium-48. Where possible it would be preferable to work with smaller samples because extreme care is required when a large irradiated sample is fused in sodium peroxide.

This work was supported in part by the Michigan Memorial Phoenix Project and the U.S. Atomic Energy Commission. Thanks are due to Professor H. J. GOMBERG, C. W. RICKER, and the staff of the Ford Nuclear Reactor for their help in making the irradiations.

SUMMARY

Submicrogram amounts of vanadium in rat liver tissue have been analyzed by rapid activation analysis. A 5-min radiochemical separation coupled with γ-ray spectrometry permitted utilization of the 3.8-min vanadium-52 radioisotope. With this procedure the lower limit of detection at a thermal neutron flux of 10^{12} n/cm²/sec was about $3 \cdot 10^{-9}$ g of vanadium.

RÉSUMÉ

De submicroquantités de vanadium ($3 \cdot 10^{-9}$ g) dans des tissus biologiques (foie de rat) ont pu être déterminées rapidement par activation. Une séparation radiochimique de 5 min, combinée avec une spectrométrie aux rayons-x, permet l'utilisation du radioisotope vanadium-52 (3.8 min).

ZUSAMMENFASSUNG

Vanadium wurde in Gewebe von Rattenlebern mit Hilfe der Neutronenaktivierungsanalyse bestimmt. Die Proben wurden kurze Zeit bestrahlt, innerhalb von 5 Minuten radiochemisch getrennt, und mit einem γ-Spektrometer die Aktivität des Vanadium-52 ($t_{1/2} = 3.8$ min) bestimmt. Die Nachweisgrenze lag bei einem thermischen Neutronenfluss von 10^{12}n/cm²/sec bei $3 \cdot 10^{-9}$ g Vanadium.

REFERENCES

1 B. A. LOVERIDGE AND A. A. SMALES, in *Methods of Biochemical Analysis*, Vol. V., Interscience, New York, 1957, p. 225.
2 J. L. BROWNLEE, JR. AND W. W. MEINKE, unpublished studies.
3 R. FUKAI AND W. W. MEINKE, *Limnol. Oceanog.*, 1 (1962) 186.
4 A. A. BENSON, *Proc. Second Intern. Conf. Peaceful Uses At. Energy, Geneva, 1958*, paper 858, p. 1–13.
5 M. D. GUELBENZU, J. M. L. AZCONA AND A. S. RUIZ, *Rev. Espan. Fisiol.*, 7 (1951) 74.
6 W. W. MEINKE, *Nucleonics*, 17/9 (1959) 86.
7 W. W. MEINKE, *U. S. At. Energy Comm. Rep. AECU 3641*, November 1957.
8 W. W. MEINKE, *U. S. At. Energy Comm. Rep. AECU 3887*, November 1958.