Determination of Fallout Cesium-137 in Animal and Plant Tissues

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Cesium-137 is one of the principal fallout contaminants in agricultural products. A method is reported for its determination in plant and animal tissues. Cesium-137 is separated from ashed samples by coprecipitation with cobaltous cobalticyanide and measured with a single-channel γ-ray spectrometer. An alkaline earth fraction suitable for Sr^{90} analysis is available. The method uses conventional equipment and is adaptable to parallel determinations on large numbers of samples.

Cesium-137 is one of the principal, long-lived nuclear fission products which are incorporated into the tissues of plants and animals. It is thus one of the fallout contaminants of interest in agricultural products. Fallout Cs^{137} in agricultural products has been measured by bulk counting, using large scintillation crystals and multichannel pulse-height analyzers (*1, 2, 9*). This method, aside from requiring very specialized instrumentation, is subject to interference from γ-rays of $RuRh^{103, 106}$ and $ZrNb^{95}$, if they are present in a high ratio to Cs^{137}. A method of analysis involving chemical separation of Cs^{137} would permit the use of conventional instrumentation and eliminate interferences from gamma emitters of similar energies.

Radioactive cesium has been separated by ion exchange from other fission products (*6, 8*) and from water (*4, 10*). It has been separated from fission product mixtures by coprecipitation with thallous salts (*11*), cobaltous cobalticyanide (*5*), and potassium sodium cobaltinitrite (*4*). Precipitation as the chloroplatinate (*3*), tungstophosphate (*7*), and molybdophosphate (*4*) has also been reported, as have combinations of the procedures.

A procedure for determination of Cs^{137} in agricultural products should use conventional instrumentation; give good decontamination, especially from $RuRh^{103, 106}$ and $ZrNb^{95}$; give quantitative recovery of cesium, so as to eliminate the necessity for weighing of carrier to determine chemical yield through the procedure; be adaptable to parallel determinations in large numbers of samples; and be compatible with procedures for other fallout contaminants. Coprecipitation of cesium with cobaltous cobalticyanide as reported by Landford (*5*) gave adequate decontamination and quantitative recovery of cesium. Various other methods were rejected after preliminary investigations or on the basis of literature concerning them.

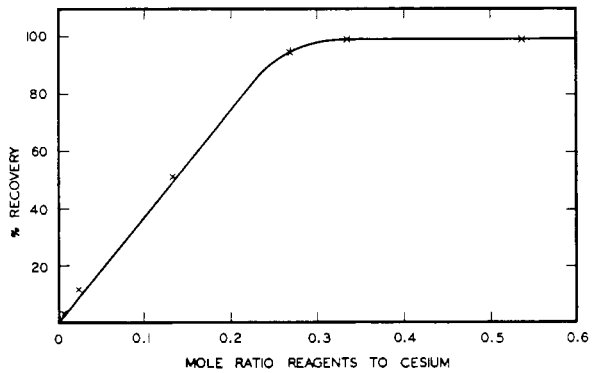

Figure 1. Effect of mole ratio of cobaltous chloride and potassium cobalticyanide to cesium on recovery

Mole ratio of cobaltous chloride to potassium cobalticyanide 1 in all cases. Each point represents average of at least three experiments

Reagents and Equipment. All reagents were of highest available purity and not further purified. Potassium cobalticyanide (K and K Laboratories, Inc., Long Island City, N. Y.) was used. Radiation measurement equipment consisted of a well-type scintillation counter [NaI(Tl) crystal 4.76 cm. in diameter, 5.71 cm. thick] in a 2-inch lead shield, linear amplifier, single-channel pulse-height analyzer, and decade scaler. For Cs^{137} the 0.662-m.e.v. γ-ray of the daughter Ba^{137} was counted and for Cs^{134}, the 0.601-m.e.v. γ-ray was used. In both cases the equipment was set to count a range of γ-ray energies ± 5 k.e.v. from the photopeak energy.

General Procedure. The sample for analysis was placed in a glass 50-ml. centrifuge tube. It was acidified, cobaltous nitrate was added, and the volume was adjusted to 25 to 35 ml. After heating in boiling water, potassium cobalticyanide was added and on cooling the precipitate was centrifuged and washed with 2*N* nitric acid. The steps of this general procedure were studied and are reported below. Ten milligrams of cesium carrier as cesium chloride and 0.1 microcurie of Cs^{134} as a tracer were used in all studies.

Concentration of Cobaltous and Cobalticyanide Ions. The recovery of Cs^{134} was studied as a function of cobaltous and cobalticyanide concentrations. Solutions of cobaltous nitrate and potassium cobalticyanide (0.2*M*) were added in varying amounts, using 2.1 ml. of concentrated sulfuric acid as recommended by Landford (*5*). It was initially found that the yield was limited by the ion present in least concentration and thereafter equimolar amounts were added (Figure 1). A mole ratio of each reagent to cesium of about 0.33 was necessary for quantitative recovery of cesium in the precipitate. This is somewhat higher than the mole ratio of 0.267 used by Landford (*5*). Since excess reagent was not objectionable, it was decided to use a mole ratio of 0.53, which corresponds to 2 ml. each of 0.2*M* cobaltous nitrate and 0.2*M* potassium cobalticyanide with 10 mg. of cesium carrier.

Acidification. In the final reaction volume of 30 ml., a sulfuric acid concentration of 0.14*M* gave reduced yield, as did a concentration of 7.2*M* (Table I).

Table I. Effect of Acidity on Recovery of Cs^{134}

H_2SO_4 Concn., M	Recovery, %
0.14	94
1.5	99
3.6	100
7.2	86

Table II. Effect of Acid Anion on Recovery

Material	Recovery, % H_2SO_4	Recovery, % HCl
Synthetic solution	100.0	100.0
Wood ash, 0.5 gram	100.9	99.9
Muscle ash, 5 grams	99.9	101.4
Bone ash, 5 grams	72.1	100.2

Table III. Separation of Strontium and Cesium

Method	Recovery, % Sr	Recovery, % Cs
Sr (10 mg.) pptd. as $SrCO_3$	99	97
Supernatant from Cs ppt.	99	101
Ca (2.7 grams) and Sr (10 mg.) pptd. as carbonates	101	50–80

Table IV. Recovery of Cs^{134} from Agricultural Materials

Material	Ash Wt., G.	Replications	Recovery, %
Bone	5	8	100.2
Muscle	5	7	101.4
Wood	0.5	6	100.2
Alfalfa	5	3	99.9

In the range of 1 to 4*M* the yield was essentially quantitative. It was desired to determine cesium in the presence of large amounts of calcium, as would be found in bone and milk samples. The use of sulfuric acid resulted in copious calcium sulfate precipitation which entrained some of the cesium (Table II). As hydrochloric acid could be substituted for sulfuric acid without harm to the yield (Table II), 3 ml. of concentrated hydrochloric acid were used as the acidifying agent.

Precipitation Conditions. The yield of Cs^{134} was not increased by digesting the precipitate in a boiling water bath for 30 minutes or by prolonging the time allowed for the precipitate to form beyond 30 minutes. Very slow addition of potassium cobalticyanide was advisable to obtain an easily sedimented precipitate.

Interferences with Chemical Yield. All elements normally present in appreciable quantities in plant or animal ash, as well as all micronutrients expected in such ashes, were checked for their effect on chemical yield. When the following ions were tested in the weights indicated —Na (10 mmoles), K (10 mmoles), Ca (53 mmoles), Cl (50 mmoles), phosphate (87 mmoles), Cu (20 μg.), Mo (80 μg.), Co (0.7 μg.), Mn (200 μg.), Fe (0.4 μg.), and Zn (40 μg.)—none of these materials reduced the chemical yield of cesium. The quantity of potassium used did not increase the count rate at 0.662 m.e.v. due to the naturally occurring K^{40}.

Nitric Acid Wash. Six washings of the coprecipitate of cesium with cobaltous cobalticyanide with 2*N* nitric acid did not cause detectable loss of cesium.

Recovery of Strontium. Sr^{89} tracer with 20 mg. of strontium carrier and 2.7 grams of calcium was quantitatively recovered in the supernatant after cesium precipitation when hydrochloric acid was used as the acidifying agent. However, considerable variable losses of cesium occurred, probably by entrainment, when 2.7 grams of calcium containing strontium carrier and tracer were precipitated as either the carbonate or the phosphate prior to cesium determination. Strontium (20 mg. labeled with Sr^{89}) could, however, be quantitatively recovered without loss of cesium by precipitation as the carbonate. Thus, the small amount of alkaline earths present in soft tissues may be precipitated with strontium carrier without loss of either Sr^{90} or Cs^{137} (Table III).

Procedure

Sample Preparation. An amount of plant or animal tissue, including bone or milk, expected to contain at least 25 picocuries (micromicrocuries) of Cs^{137} was dry-ashed at 550° C. for 12 hours. The ash was transferred to a beaker and moistened with concentrated nitric acid. After the nitric acid was evaporated on a hot plate, the sample was placed in a furnace at 550° C. for 2 hours. The nitric acid evaporation followed by dry ashing was repeated until the ash was free of carbon and of a constant color. (Two nitric acid treatments are usually required, except for soft animal tissues which often require six.) The ash was then moistened with perchloric acid, and the acid was evaporated on a hot plate, and placed in a furnace at 550° C. for 2 hours, to volatilize ruthenium (*5*, *6*). The ash was then repeatedly extracted with small volumes of hydrochloric acid and transferred to a 50-ml. glass centrifuge tube. The acidity was adjusted to the equivalent of a total addition of 4 ml. of concentrated hydrochloric acid, 10 mg. of cesium carrier was added, and the insoluble material was separated centrifugally and washed with water. The total volume of the supernatant and washings should be 25 to 35 ml.

Cesium Precipitation and Measurement. To the ash solution prepared above 2 ml. of 0.2*M* cobaltous nitrate were added and the tubes were heated in a boiling water bath for 5 minutes. Two milliliters of 0.2*M* potassium cobalticyanide were added slowly, and the contents were mixed. After standing at room temperature for 30 minutes, the tubes were centrifuged and the supernatant was rejected or reserved for strontium-90 separation. The precipitate was transferred, by using 5 ml. of 2*N* nitric acid, to a plastic test tube that will fit a well-type scintillation detector and centrifuged, and the supernatant was rejected. The cesium-137 was measured with a well-type scintillation counter, single-channel pulse-height analyzer, and a scaler.

Optional Recovery of Strontium. In all samples other than bone or milk, strontium may be precipitated prior to cesium separation. After the ash had been transferred to a centrifuge tube with hydrochloric acid, the solution was centrifuged and the insoluble material was rejected after washing with water. Twenty milligrams of strontium carrier were added, the solution was neutralized with 6*N* sodium hydroxide, and strontium carbonate was precipitated by the addition of 2 ml. of 10% sodium carbonate. After standing at room temperature for 0.5 hour, the precipitate was separated centrifugally and washed with water. Strontium-90 may be determined in the precipitate by other procedures and the supernatant and wash water carried through the balance of the procedure for Cs^{137} determination.

Recovery. Recovery studies were made by the procedure outlined above, using Cs^{134} as a tracer (Table IV). When animal tissues from a tracer experiment with Cs^{134} were used, the recovery was determined by comparison with the Cs^{134} measured by direct counting of the tissues. For liver and muscle the mean recovery through the procedure as given was 99%. These data indicate a quantitative recovery of cesium and hence obviate the weighing of cesium carrier for determination of chemical yield.

Discussion

Landford (*5*) in his study of the determination of radiocesium in fission products by cobaltous cobalticyanide coprecipitation demonstrated very high decontamination ratios for most probable contaminants. It was not felt necessary to duplicate this study. The only fission products with interfering γ-ray energies that were not adequately removed by cobaltous cobalticyanide coprecipitation of cesium were ruthenium isotopes. In this procedure ruthenium is volatilized in the ashing procedure after oxidation to RuO_4 by perchloric acid (*5*, *6*). Perchloric acid was chosen as the oxidizing agent because of its wide use in agricultural analytical laboratories.

The procedure as outlined above is adaptable to parallel determinations in a large number of samples, the principal factors limiting the size of a given run being centrifuge and furnace capacities. The author routinely makes eight

determinations in parallel. Since no unusual equipment is required, the fallout cesium separation can be made in a general-purpose laboratory. It is expedient, however, not to conduct any part of the procedure in a laboratory used for radiotracer studies or to use glassware used for radioactive materials. The counting equipment used is commercially available. The size of the initial sample is set only by the Cs^{137} level expected and the precision of measurement desired. A sample containing 25 picocuries of Cs^{137} was necessary to give a count rate equal to background, when the equipment previously described was used and an energy width of ± 5 k.e.v. was counted.

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COTTONSEED MEAL IN POULTRY FEED

Collaborative Study of the AGU Method of Grading Cottonseed Meals for Laying Rations

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The results of a collaborative study of the available gossypol unit (AGU) method of grading cottonseed meals for laying rations indicate that significant differences exist in AGU values among eggs, birds, and meals. The correlation between the AGU values of cottonseed meals and coloration in yolks of stored shell eggs produced by the meals is virtually zero. The AGU method may not be relied upon for grading cottonseed meals for laying rations.

A LARGE proportion of yolks from shell eggs from hens on cottonseed meal-containing diets develop a brown coloration when the eggs are stored under refrigeration conditions. Cottonseed meals vary widely in their ability to induce brown coloration in yolks of stored shell eggs. It was presumed by earlier workers that coloration develops because of the "free" gossypol present in the meals (*6, 7, 10, 11*).

Apparently cottonseed meals cannot be graded for laying rations on the basis of their free gossypol contents, however, since the correlation between intensity of coloration in yolks and free gossypol content of the meals fed is poor (*3*). A method for grading cottonseed meals for laying rations proposed by Grau (*4, 5*) depends upon a greater concentration in the yolks from eggs laid by cottonseed meal-fed hens of some acetone-hexane soluble material than that occurring in yolks from hens fed other rations. This acetone-hexane soluble material has an absorption maximum at 440 mμ. The absorption at this wave length was ascribed by Grau to a condensation product of gossypol and cephalin. The increased absorptivity of the preparation obtained from cottonseed meal-produced yolk over that obtained with control yolk was proposed by him as a measure of the available gossypol in the cottonseed meal fed. More specifically, the AGU of a cottonseed meal was defined by the relationship:

$$\text{AGU} = \frac{[A'_{400} - A'_{450}] - [A''_{400} - A''_{450}]}{\% \text{ of material tested in ration}} \times 100$$

where A' has reference to the absorbance of the extract from cottonseed meal-produced egg and A'' has reference to the absorbance of the extract from the control egg. Subscripts refer to the wave lengths at which the absorptivities are measured. A cottonseed meal having an AGU of 0.30 or less was reported by Grau to be suitable for laying rations in amounts up to 10% of the total ration.

Large quantities of cottonseed meals have been used in laying rations for hens during the past 2 years, where the meals incorporated into the rations were selected on the basis of the AGU testing method. The eggs produced were sold on the fresh egg market. A collaborative test of the AGU method became imperative because of the wide interest engendered by this use of cottonseed meals. These collaborating in the test were: V. P. Entwistle, Calif. Dept. Agr., Sacramento, Calif.; C. R. Grau, Univ. of Calif., Davis, Calif.; A. A.

Table I. Per Cent Composition of Rations for Laying Hens

Constituent	Amount, %
Cottonseed meal	10
Soybean meal	15
Ground yellow corn	15
Ground milo	49.4
Alfalfa meal, dehydrated	2.5
Ground limestone	3.5
Bone meal	3.5
Manganized salt	0.5
Source of riboflavin equivalent to 500 unit/gram	0.3
Source of vitamin A, 2,250 units; and vitamin D, 300 units/gram	0.3