Neutron Activation Analysis and its Application to the Analysis of Food Products[1]

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Abstract

High-flux thermal-neutron activation analysis (NAA) is an extremely sensitive means of quantitatively determining most of the elements of the periodic system. The usual limits of detection, for 75 of the elements, in the absence of appreciable interferences, range from picograms (for some elements) to as high as micrograms (for less sensitive elements). A typical element can be determined down to as low as a nanogram. This high sensitivity enables one to analyze food products, for example, for numerous trace-level elements that may be present: whether natural or added beneficial essential trace elements, or deleterious elements introduced from pesticide residues (such as Br, As and Hg), or from processing (such as Cr, Sn, Sb and Cu). Studies to be reported include the nondestructive determination of Hg in foodstuffs down to levels as low as 0.01 ppm, and of Br in foodstuffs down to about 0.1 ppm. With radiochemical separations, these detection limits can both be extended to 0.001 ppm, if needed. By combination with paper chromatographic or solvent extraction techniques, phosphorus- and halogen-containing pesticides can be sensitively determined. The NAA method can also be used to advantage at element levels much higher than trace levels, and in such cases the very high neutron flux of a nuclear reactor may not be necessary. For example, even with a small 14 Mev neutron generator, the nitrogen content of foodstuffs can be determined nondestructively, rapidly and accurately, down to levels of about 100 ppm. These determinations can also be made on-line, in food processing plants.

Introduction

HIGH-FLUX THERMAL-NEUTRON activation analysis (NAA) is an ultra-sensitive method of elemental analysis. At high thermal-neutron fluxes, it is capable of quantitatively determining lower concentrations of a majority of the elements of the periodic system than any other known method of elemental analysis. Under the conditions commonly employed, the limits of detection for 75 of the elements range from as low as picograms (10^{-12} g), for a few extremely sensitive elements, to as high as micrograms (10^{-6} g), for a few rather insensitive elements. A median, or more typical, limit of detection is about one nanogram (10^{-9} g). Except at levels very close to the limit of detection of that element, the amount and concentration of an element can usually be determined to a precision and absolute accuracy of about $\pm$2–3% of the value, and at levels ranging all the way from trace levels up to major constituent levels. Sample sizes can range from micrograms all the way up to tens of grams. For a 1-g sample, the μg limit of detection is numerically the same as the parts per million (ppm) concentration limit of detection (i.e., 1 μg/g = 1 ppm = 0.0001%). The method is completely independent of the chemical form (or forms) of the elements present. In many cases the analyses can be performed purely instrumentally and nondestructively, via multichannel γ-ray spectrometry of the activated sample. In other cases, post-irradiation radiochemical separations of the elements of interest, with carriers, must be employed. A given sample can be analyzed for a single element of interest, a few or several elements or a large number of elements, as desired. Where the element of interest forms a very short-lived induced activity (half life of seconds to minutes), samples can be analyzed very rapidly, instrumentally, one at a time. Where the element of interest forms a longer-lived induced activity (half life of hours, days, or longer), a longer irradiation may be desirable, but then many samples can be activated simultaneously, then counted one or a few at a time, at appropriate decay times.

For a number of years, the author and his colleagues have been applying the NAA method to many fields of investigation (e.g., chemistry, metallurgy, geochemistry, biology and medicine, crime investigation, pesticide residues, etc.). In this paper, attention is devoted almost entirely to the useful applications of the method that have been developed for the determination of certain pesticide residues in foodstuffs, and certain trace elements in foodstuffs.

Nature of the Method

Typically, a small weighed sample of the material to be analyzed is placed in a small polyethylene vial, then exposed in a nuclear reactor (research type) to a rather high thermal-neutron flux (10^{12}–10^{13} n/cm^2-sec) for an appropriate period of time (usually in the range of seconds to hours), then counted, after an appropriate decay time, on a multichannel γ-ray spectrometer.

With thermal neutrons, for practical purposes, carbon, nitrogen and hydrogen are not activated at all, and oxygen very little. Thus, typical foodstuffs represent a rather ideal matrix, if one desires to detect and measure the amounts of various other elements present in foodstuffs as minor constituents or trace impurities. Almost all other elements are readily activated (though to various degrees) by bombardment with thermal (slow) neutrons. They become activated by a neutron-capture nuclear reaction, usually called an (n,γ) reaction. For example, if a sample containing some fluorine is exposed to a flux of thermal neutrons, some of the stable F^{19} nuclei will undergo a neutron-capture reaction, $F^{19} + n^1 \rightarrow F^{20} + \gamma$, or, more simply, $F^{19}(n,\gamma)F^{20}$, to form a radionuclide of fluorine, F^{20}. Fluorine-20 is radioactive, and decays with a half life of 11.6 seconds, emitting β particles of a certain maximum energy (E_{max} = 5.41 Mev) and monoenergetic γ-rays with an energy of 1.63 Mev (Mev = million electron volts).

Twenty of the elements in nature are monoisotopic, i.e., they occur only as one stable nuclide. Except in those cases where neutron capture forms a metastable isomer, these elements each form only one radionuclide, by (n,γ) reaction. The remaining ele-

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ments occur in nature as mixtures of 2 or more (in a few instances, up to 10) different stable nuclides. Many of these elements, by (n,γ) reaction, form two or more radionuclide products. For example, copper in nature consists of 69.09% Cu^{63} and 30.91% Cu^{65} (stable nuclides). With thermal neutrons, copper forms two radioactive products: 12.8-hr Cu^{64} and 5.10-min Cu^{66}. Copper-64 decays in a branched fashion (38% by pure β^- emission, 19% by pure β^+ emission, and 43% by simple electron capture), whereas Cu^{66} decays in a simpler fashion (91% by pure β^- emission, and 9% by β^- emission plus a 1.04 Mev γ-ray).

The amount of a particular radionuclide activity (A_o, expressed in disintegrations per second, dps) generated in a given weight (w) of a particular element depends upon the isotopic abundance (a) of the "target" nuclide in that element, the atomic weight (AW) of the element, the thermal-neutron flux (Ø) to which the sample is exposed, the isotopic cross section (σ) of that stable nuclide toward capture of a thermal neutron, the duration of the irradiation period (t_i), and the half life (T) of the radionuclide formed:

$$A_o = N\emptyset\sigma S, \quad [1]$$

where $$N = 6.02 \times 10^{23}\ aw/AW, \quad [2]$$

and $$S = 1 - e^{-0.693\ t_i/T} \quad [3]$$

In an analysis, of course, w is the unknown quantity. The quantity, S, is the so-called "saturation" term. It is a dimensionless quantity, with values that range only from 0 to 1. At t_i/T values of 1, 2, 3, 4, ... , S has values of ½, ¾, ⅞, $^{15}/_{16}$, ... If an irradiation is continued for a period of time that is several times the half life of a particular induced activity, the level of that activity in the sample will asymptotically approach a limiting value, since, as rapidly as new nuclei of that species are formed by (n,γ) reaction, ones formed earlier will be disappearing by radioactive decay. The S term is the only time-dependent quantity in Equation [1], other than the resulting value, A_o. As a result, when one is looking for elements that form very short-lived products, one uses only a short irradiation period, followed rapidly by counting for a short time. Where longer-lived activities are the ones of interest, one usually employs a longer irradiation period (t_i), in order to increase S, then waits for a while after the end of the irradiation (to allow the interfering shorter-lived activities to decay out) before counting; for improved sensitivity, one may also wish to employ a longer counting period.

When the irradiation is stopped, each induced activity decays according to the equation:

$$A_t = A_o\, e^{-0.693\ t/T}, \quad [4]$$

where t is the decay period since the end of the irradiation (t_o), and T is the half life of the radionuclide. In practice, one seldom employs the absolute form of the method, directly substituting values in Equations [1] through [4], but instead employs the comparator form. This usually involves the irradiation and counting of known (standard) samples of each element of interest, in exactly the same way as the unknown samples. For any particular induced activity, one can then write the appropriate equation (based upon Equations [1]–[4], and including the counting efficiency (ϵ) for that radionuclide) for both unknown and standard. Dividing one equation by the other, the terms, a, AW, Ø, σ, S, and ϵ all cancel out, since they are exactly the same for both unknown and standard. The final equation is thus very simple:

$$\frac{w\ (unknown)}{w\ (standard)} = \frac{A'_o\ (unknown)}{A'_o\ (standard)}. \quad [5]$$

The A'_o terms represent radionuclide counting rates (counts per second) rather than disintegrations rates: $A'_o = \epsilon A_o$. It is equally valid for the counting rates (of unknown and standard) at any particular decay time, i.e., not only at t_o.

Forms of the Method

There are two forms of the NAA method that can be employed: the instrumental form and the radiochemical-separation form. Usually, the instrumental (nondestructive) form is preferred—if it is applicable—since it requires less of a chemist's time. It is based upon multichannel γ-ray spectrometry, using a thallium-activated sodium iodide (NaI(Tl)) scintillation detector or a lithium-drifted germanium (Ge(Li)) semiconductor detector. Both of these detector materials are good absorbers for x-radiation and γ-radiation—the efficiency of detection and efficiency of total absorption (as opposed to only Compton scattering) increasing with increasing size of the detector, especially thickness. Upon interaction with a γ-ray photon, each kind of detector produces an electrical output pulse (in the case of the NaI(Tl) detector, via a coupled photomultiplier tube) whose magnitude is directly proportional to the γ-ray energy absorbed by the detector in that event. Each detector output pulse is linearly amplified and its size then measured by the pulse-height analyzer, and a count then stored in the appropriate channel of the analyzer's memory. Available NaI(Tl) detectors are very efficient, but rather poor in resolution—resulting in rather broad total absorption peaks (photopeaks)—hence a 400-channel pulse-height analyzer is quite adequate. At present, only rather small Ge(Li) detectors are available, and hence their detection efficiencies are rather low, at least for higher-energy γ-rays. However, their energy resolution is very good, typically being from 10–20 times better than that of a NaI(Tl) detector. In order to retain the advantage of this higher resolution (which results in the complete separation of many photopeaks that are close enough in energy to one another to partially overlap one another with a NaI(Tl) detector), a more complicated multichannel analyzer is used, usually one with 4,096 analysis and storage channels.

With either type of detector or analyzer, the pulse-height spectrum of an activated sample consists of counts per channel, as the ordinate, versus channel number, as the abscissa. The abscissa scale is a calibrated, linear, energy scale (typically set at 7.5 kev/channel with a NaI(Tl) detector and 400-channel analyzer, or at 0.5 kev/channel with a Ge(Li) detector and 4,096-channel analyzer). The observed spectrum, accumulated during a counting period, perhaps, of 1 min, is then a 400-point, or 4,096-point spectrum. The general shape is that of a continuum, decreasing in magnitude with increasing γ-ray energy (increasing channel number), superimposed upon which are Gaussian-shaped photopeaks of various sizes, at various locations on the energy (channel number) scale. The channel number of a photopeak thus indicates the γ-ray energy and hence the radionuclide, and hence the element. The net photopeak counting

rate (above the Compton continuum), compared with that of the corresponding standard of that element, corrected to the same decay time, is a quantitative measure of the amount of that element present in the sample. For a given sample, irradiated at some particular thermal-neutron flux, the detailed shape of the resulting pulse-height spectrum is dependent upon the irradiation time (t_i) and the decay period before counting (because the different contributing radionuclides have different half lives, T), the type and size of detector used and the distance between the activated sample and the detector. In simple cases, all the calculations can be done readily by hand. In more complicated cases, such as those involving many induced activities, and ones involving overlapping photopeaks, data processing by computer is desirable and is routinely employed now in many activation analysis laboratories, such as the author's. Where an element is not detected, a firm (3σ) upper limit for the possible amount present in the sample can be calculated from the pulse-height data. For multi-element scanning of samples, the author's group has developed, and uses, a particular schedule of irradiation, decay, and counting times, plus a computer program that gives a readout for 65 elements: a firm value for each one detected, and a firm upper limit for each undetected one.

The instrumental form of the method requires that the induced activity of interest emit gamma rays, reasonably energetic x-rays, or positrons (which undergo annihilation, to emit 0.511 Mev γ-ray photons). If, by (n,γ) reaction, the element of interest forms a radionuclide that decays only by β^- emission or the emission of low-energy x-rays, or if much higher levels of other induced activities, emitting γ rays of higher energy than those of the activity of interest, obscure the detection of the activity of interest, one must resort to post-irradiation separation of the species of interest. An example where radiochemical separation is necessary is that of determining phosphorus by NAA with thermal neutrons, since the only product it forms is 14.3-day P^{32}, a pure β^- emitter. In such cases, the activated sample is dissolved (usually by wet-ashing) in the presence of an accurately measured macro carrier amount of the element of interest (e.g., 10.0 mg of phosphorus in some suitable chemical form), chemically equilibrated with the carrier, then separated and purified from the interfering activities. Once adequately purified (checked for radiochemical purity by regular techniques), the separated activity can be counted on a simple β^- (or γ-ray) detector. The recovery of the added carrier is then measured by any standard procedure (gravimetric, volumetric, etc.), since it is present at a high, easily-measured level, and the counting results then normalized to a 100% recovery. For trace, and ultra-trace, concentration levels, this is a very powerful technique, since it eliminates all of the usual problems of microconcentration elemental analysis by more conventional methods (losses, reagent contamination, reagent blank correction, etc.). It is, of course, more tedious than the instrumental form of the NAA method, and hence is usually only employed where it is really necessary. In the author's laboratory, a new Swedish automated radiochemical-separation apparatus is now being tested. It shows considerable promise.

Sensitivity of the Method

As shown in Table I, at a typical research reactor thermal-neutron flux (10^{13} n/cm^2-sec), and a maximum sample irradiation time of 1 hr, the limits of detection for 75 elements range from as low as about 10^{-7} μg (dysprosium) to as high as about 10 μg (iron). A typical element can be detected down to about 0.001 μg. These sensitivities can all be improved further, if necessary, by employing an even higher neutron flux. Also, the sensitivities for about half of the elements listed in the Table (those forming radionuclides with half lives of several hours or longer) can be improved further, if necessary, by employing longer irradiation times and longer-than-normal counting periods.

TABLE I

Interference-Free Limits of Detection for 75 Elements by Neutron Activation Analysis at a Thermal-Neutron Flux of 10^{13} n/cm^2-sec (for 1 hr maximum)

Median sensitivity = 0.001 μg	
Limit of Detection, μg	Elements
$1-3\times10^{-7}$	Dy
$4-9\times10^{-7}$	Eu
$1-3\times10^{-6}$	
$4-9\times10^{-6}$	Mn, In, Lu
$1-3\times10^{-5}$	Co, Rh, Ir
$4-9\times10^{-5}$	Br, Sm, Ho, Re, Au
$1-3\times10^{-4}$	Ar, V, Cu, Ga, As, Pd, Ag, I, Pr, W
$4-9\times10^{-4}$	Na, Ge, Sr, Nb, Sb, Cs, La, Er, Yb, U
$1-3\times10^{-3}$	Al, Cl, K, Sc, Se, Kr, Y, Ru, Gd, Tm, Hg
$4-9\times10^{-3}$	Si, Ni, Rb, Cd, Te, Ba, Tb, Hf, Ta, Os, Pt, Th
$1-3\times10^{-2}$	P, Ti, Zn, Mo, Sn, Xe, Ce, Nd
$4-9\times10^{-2}$	Mg, Ca, Tl, Bi
$1-3\times10^{-1}$	F, Cr, Zr
$4-9\times10^{-1}$	Ne
1 — 3	S, Pb
4 — 9	Fe

Pesticide-Residue Applications

The NAA method has been used effectively, to date, to determine bromine, chlorine, arsenic and mercury pesticide residues in various types of foodstuffs. Some illustrative cases are cited below.

The first NAA studies of pesticide residues in foodstuffs were carried out by the author and J. C. Potter, in 1960–1961, and were published in 1962 (1). This study was concerned with the instrumental NAA determination of bromine pesticide residues in a variety of crops, resulting from the application to the soil, in various amounts, of the bromine-containing nematocide, 1,2-dibromo-3-chloropropane. In the various kinds of crops studied, the Br levels found ranged from as low as 20 ppm to as high as 440 ppm, the level being dependent upon the type of crop, the extent of nematocide treatment of the soil, and the time interval since treatment. Check samples of various crops, grown in untreated soil, in most cases contained in the range of 1–10 ppm of natural bromine. Analyses were carried out both with thermal-neutron activations at the modest (10^8 n/cm^2-sec) neutron flux from a 3 Mev electron Van de Graaff accelerator—detecting the 0.511 Mev (β^+ annihilation) and 0.618 Mev γ-rays of 17.6-min Br^{80}, and at the higher (10^{12}) neutron flux of a research-type nuclear reactor—detecting the 0.554–0.777 Mev γ-rays of 35.3-hr Br^{82}, in both cases using a NaI(Tl) γ-ray spectrometer.

Since the time of these first experiments, the technique has been refined further, and is now used quite routinely for the determination of bromine pesticide residues in many kinds of crops and foodstuffs. The present technique typically involves the simultaneous activation of up to 40 samples (0.5–1 g), and a Br standard, for 30 min in the rotary specimen rack of the 250 kw TRIGA Mark I reactor (where the

thermal-neutron flux is 1.8×10^{12} n/cm^2-sec), followed by NaI(Tl) γ-ray spectrometry after a decay period of 3 days, to decrease the interference from 15.0-hr Na^{24}. Calculations are based upon the net photopeak area of 35.3-hr Br^{82} in the 0.554 Mev or 0.777 Mev region of the pulse-height spectrum. A counting period of 5 min is usually sufficient. Bromine levels down to 1 ppm, or somewhat lower, can thus be detected instrumentally (under the same irradiation and counting conditions, Br levels as low as 0.01 ppm can be determined, if the Br^{82} is radiochemically separated before counting).

Bromine pesticide residues of interest, and concern, also occur in the methyl bromide fumigation of stored grains, flour and certain other food products. If the fumigations are excessive, or are repeated frequently, the stored material can accumulate quite considerable levels of bromide residue, up to several hundred ppm. This application was first investigated by D. L. Lindgren et al. (2), the analyses being performed in the author's laboratory. Many such samples are now routinely analyzed for Br residues in the author's laboratory (same procedure as that described above for nematocide Br residues).

In a recent study of an inter-laboratory comparison sample of flour lightly fumigated with CH_3Br, supplied by the International Atomic Energy Agency (IAEA), NAA measurements carried out by D. M. Fleishman and V. P. Guinn gave a mean value of 8.0 $\pm$ 0.6 ppm Br (total range of 23 values obtained on three aliquots: 7.1–9.1 ppm Br). The 23 measurements included measurements with three different detectors—solid and well-type NaI(Tl) crystals, and a 15 cm^3 Ge(Li) detector—at five different decay times, and via the 0.554 and 0.777 Mev peaks of Br^{82} separately. Nine NaI(Tl) measurements, based on the 0.777 Mev peak, gave a mean value of 8.0 $\pm$ 0.8 ppm Br (from measurements at three different decay times). Fourteen Ge(Li) measurements gave a mean value of 8.0 $\pm$ 0.5 ppm Br (8.1 $\pm$ 0.5 from the 0.554 Mev peak; 7.9 $\pm$ 0.6 from the 0.777 Mev peak) from measurements at three different decay times. In the 0.544–0.828 Mev region, Br^{82} has major peaks at 0.554 Mev and 0.777 Mev, and lesser peaks at 0.619 Mev, 0.698 Mev and 0.828 Mev. With the intrinsically poorer resolution of the NaI(Tl) detectors, there is so much overlapping that only two broad peaks are observed, peaking at about 0.55 Mev and 0.78 Mev. With the much better resolution of the Ge(Li) type of detector, as shown in Figs. 1 and 2, all five of these Br^{82} peaks are completely resolved from one another. On this same IAEA sample, unofficial results from five other laboratories, that also used the instrumental NAA technique, resulted in mean values of 8.3, 8.6, 8.0, 8.1 and 8.6 ppm Br. Three other laboratories, using NAA followed by radiochemical separation, obtained mean values of 5.7, 7.6 and 8.8 ppm Br. One laboratory, using a chemical method (not NAA), obtained a mean value of 6.5 ppm Br.

Chlorine pesticide residues can also be sensitively determined by NAA, as reported, for example, by R. A. Schmitt and G. Zweig (3), but this is not so attractive an application of the method as the application to the determination of bromine residues. The reason, of course, is that all plant and food materials contain relatively high levels of inorganic chloride. In order to determine the much smaller amount of organic chlorine possibly present in the material (e.g., in the form of DDT), one must perform an organic solvent extraction of the sample, followed by NAA of the extract for chlorine. Since plant materials contain very little natural organic chlorine, any Cl found in the extract can, in almost

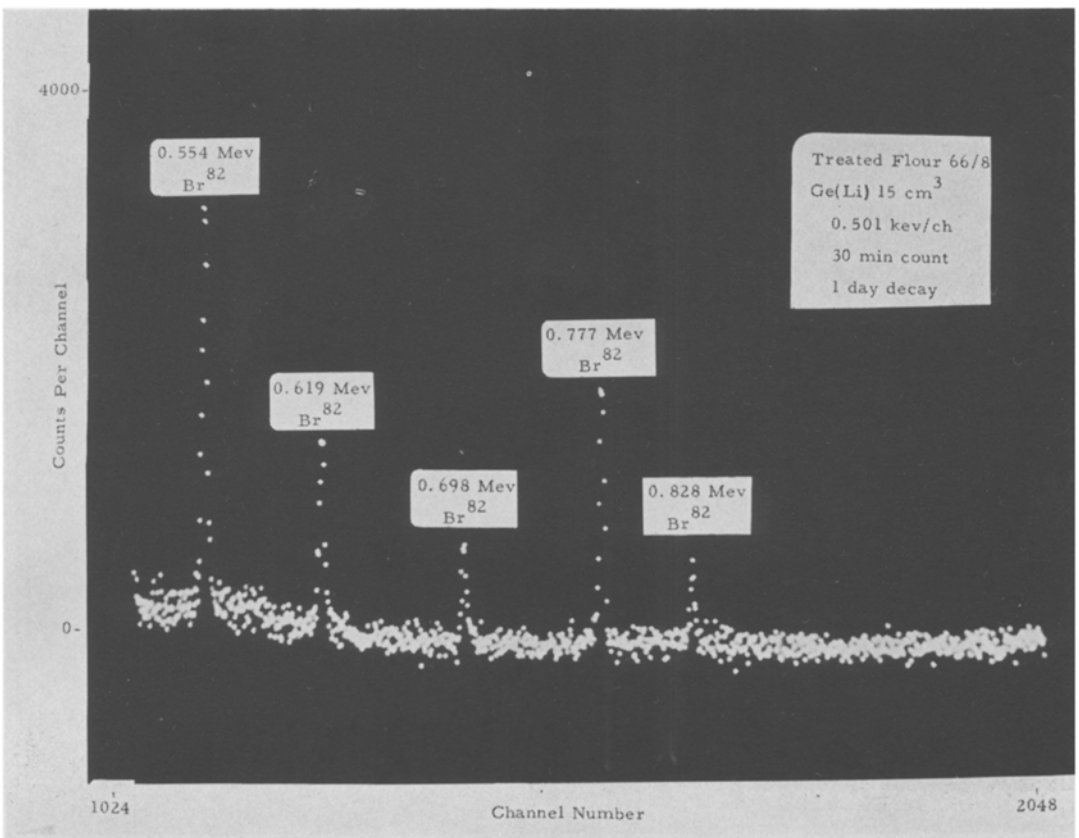

FIG. 1. Ge(Li) pulse-height spectrum of a neutron-activated sample of flour containing 8 ppm bromine.

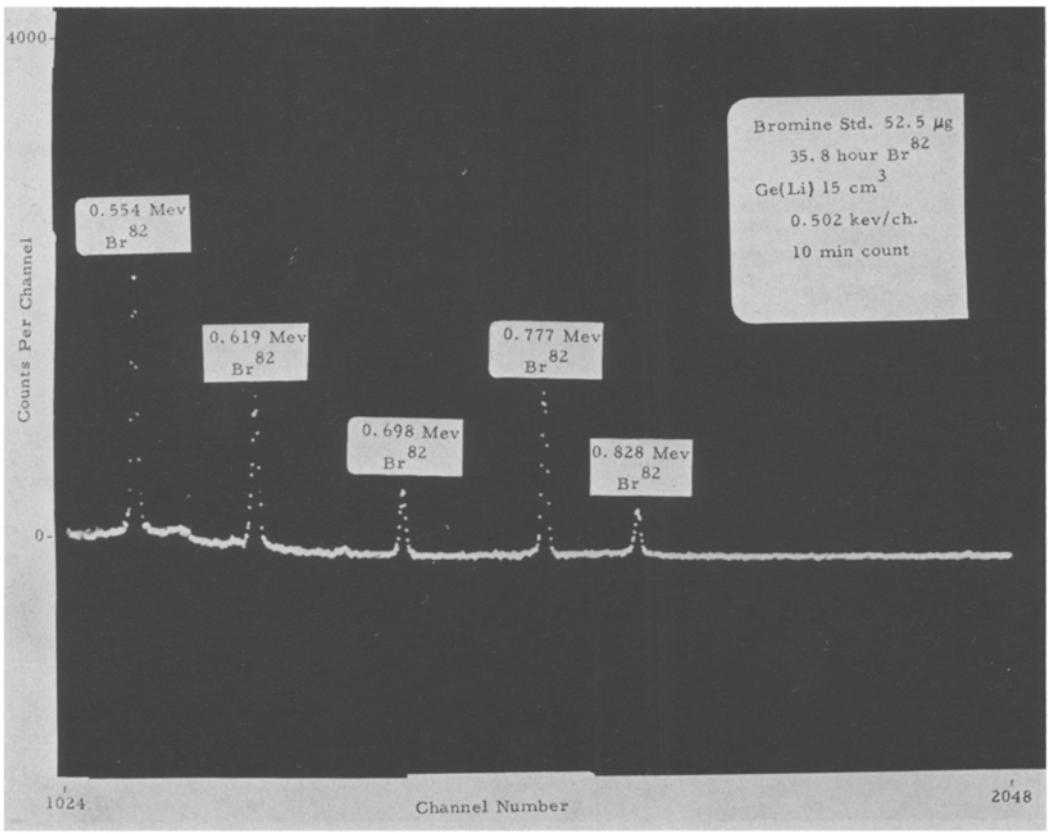

FIG. 2. Ge(Li) pulse-height spectrum of a neutron-activated aqueous bromide standard.

all cases, be legitimately assumed to be present as chloro insecticide. However, once the organic extraction has been performed, it is usually much easier, and just as sensitive and quantitative, to analyze the extract by gas chromatography. This latter techniques also identifies the particular chloro insecticide species (one or more) that are present.

Interest in the determination of mercury pesticide residues in foodstuffs and animal tissues has increased markedly during the past few years. Part of this increased interest has stemmed from some major instances of mercurial poisoning in Japan (in some cases fatal) that resulted from concentration of mercury in fish that were exposed to water accidentally contaminated with mercury from waste storage ponds, followed by consumption of the fish by humans. Some of the increased interest has also come from studies in Sweden, which have shown dangerously high levels of mercury in fish from some Swedish freshwater lakes (apparently largely due to contamination of streams and lakes by mercury-containing effluents from pulp and paper plants), and concentration of mercury in both grain-eating and fish-eating birds. Although the toxicity of mercury has been shown to be strongly dependent upon the chemical form of the mercury (alkyl Hg compounds being much more toxic, for example, than aromatic or inorganic Hg compounds), an analysis for total mercury is, itself, of considerable value.

Various studies on mercury in water, foodstuffs, and various animal species (fish, birds, mammals)—summarized in Amsterdam in May of 1967, at the IAEA Expert's Meeting on Mercury in the Biosphere—have shown, for example, that many common, uncontaminated, foodstuffs contain from 0.005 to 0.05 ppm Hg, some even containing as much as 0.1–0.3 ppm Hg. It has been shown that one can readily detect Hg in such materials, by purely instrumental NAA (at reactor thermal-neutron fluxes in the range of 10^{12}–10^{13} n/cm^2-sec), down to levels of about 0.05 ppm. With radiochemical separation of the induced 65-hr Hg^{197}, levels as low as 0.001 ppm Hg can be determined.

Recently, the author's laboratory undertook the analysis of two interlaboratory comparison samples of flour, distributed by the IAEA. One was flour from grain containing no deliberately-added mercury. The other was flour from grain containing some mercurial fungicide-treated seed grain. Thermal-neutron activation of mercury produces several radionuclides of mercury, but, of these, the one that provides the best sensitivity of detection is the induced 65-hr Hg^{197} activity. Three representative, ~4 cm^3 bulk volume, ~2.3 g aliquots were taken of each of the two flour samples. They were activated for 30 min in the 250 kw TRIGA Mark I reactor at a thermal-neutron flux of 1.8×10^{12} n/cm^2-sec, along with a carefully-prepared aqueous standard solution of Hg^{++}. The various samples and the standard were then counted, at decay times of 1 day and 7 days, by multichannel γ-ray spectrometry. Three kinds of detectors were employed in the spectrometry: a $3 \times$ 3-in. solid NaI(Tl) detector, a 3×3-in. well-type NaI(Tl) detector, and a 15 cm^3 (10 mm depth) planar Ge(Li) detector. The results obtained on the mercurial-pesticide containing sample were essentially the same regardless of the type of detector used, or of the decay time at which the measurements were made. The mean value of the 12 results obtained was 5.10 ± 0.16 ppm Hg (5.13 ± 0.15 ppm from three Ge(Li) measurements; $5.09 + 0.17$ ppm from nine NaI(Tl) measurements).

It is interesting to note that, in its decay, Hg^{197} emits essentially equal numbers of 77.6 kev γ-ray photons and 68.8 kev gold K x-rays. With a NaI(Tl) detector, these are so close together in energy that

they appear in the pulse-height spectrum as a single photopeak, just slightly broader than one would expect (from the resolution of the detector) for a single peak in this energy region. However, as can be seen in Figs. 3 and 4, the two peaks are completely resolved with the Ge(Li) detector.

The mean value obtained (5.10 ± 0.16 ppm Hg) is in good agreement with the results obtained in 16 other laboratories on this same material, using the NAA procedure (4 instrumental, 12 with radiochemical-separation). The mean value from all 17 laboratories (excluding one grossly high value) was 4.86 ± 1.25 ppm Hg.

In the other IAEA sample, which contained no deliberately introduced mercury, no mercury could be detected by means of purely instrumental NAA. However, from the counting data in the region of the γ-ray spectra where the Hg^{197} peak (or peaks) should appear, a firm upper limit could be set for the possible concentration of mercury in the sample: $\leq$0.04 ppm Hg. Radiochemical separations were not attempted on the sample, in the author's laboratory, but were carried out in 14 of the other participating laboratories. The mean of all their determinations on this sample was 0.044 ± 0.014 ppm Hg.

Arsenic, from arsenic-compound pesticide residues, is also readily determined by means of high-flux NAA with thermal neutrons (4). By (n,γ) reaction, arsenic forms 26.4-hr As^{76}. This emits a 0.559 Mev γ-ray photon in 43% of its disintegrations. In a 1-hr irradiation at a thermal-neutron flux of 10^{13} n/cm^2-sec, the As^{76} photopeak counting rate, per gram of arsenic, is about 1.7×10^{10} cpm (2 cm from a $3 \times$ 3-in. NaI(Tl) detector). Thus, the defined limit of instrumental detection for arsenic, in the absence of appreciable interferences, is about 6×10^{-10} g, or 0.0006 μg, i.e., 0.0006 ppm in a 1 g sample. With typical food samples to be analyzed for arsenic (apple peels, for example), Compton pulses from other induced activities of comparable half life, but larger γ-ray energies (e.g., 15.0-hr Na^{24}) increase the practically attainable limit of detection appreciably above this interference-free value, but very good sensitivity is still attainable. If samples contain relatively appreciable concentrations of other elements whose (n,γ) products emit γ-rays very close in energy to the 0.559 Mev As^{76} peak, such as copper and antimony, for example, the various photopeaks can readily be resolved—with some sacrifice in sensitivity, of course—by means of a Ge(Li) detector. If all else fails, one can still attain the interference-free limit of detection by employing a radiochemical separation of the As^{76} activity, with arsenic carrier.

Toxic elements, other than ones coming from pesticides, can end up in food products as a result of corrosion and wear in food-processing equipment, or from inadvertant additions of chemicals. Contamination with low, but possibly toxic, amounts of Cu, Ni, Cr, Sn, and Sb can happen. High-flux NAA can be a very useful analytical method in these cases also.

Trace-Element Determinations

In contrast to the interest in determining ppm levels of certain toxic elements in foodstuffs, occurring as pesticide residues, there is also much interest in determining ppm levels of certain elements known to be beneficial trace elements (to fowl, animals, and man), or possibly to be beneficial, from the nutritional standpoint. For example, it is known that the trace elements, Zn, Cu, Mn, Mo, I, Co and Se, are essential to man (5), and there is some evidence,

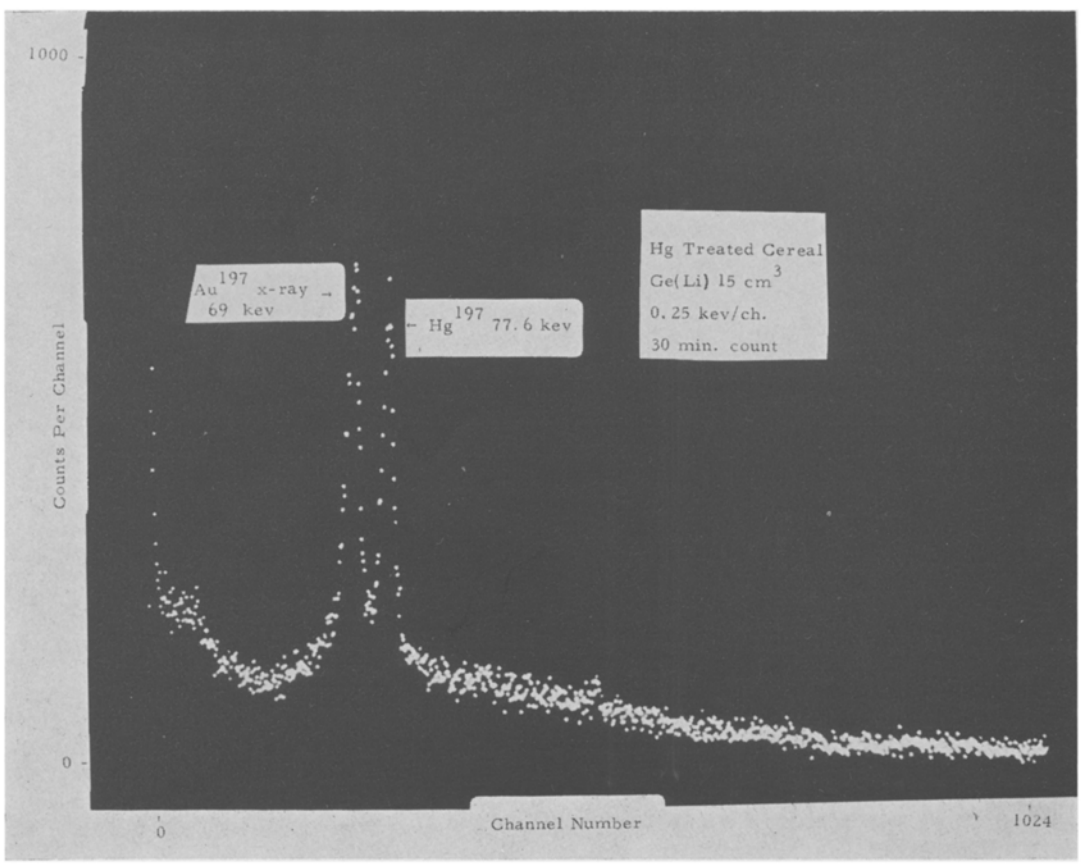

FIG. 3. Ge(Li) pulse-height spectrum of a neutron-activated sample of flour containing 5 ppm mercury.

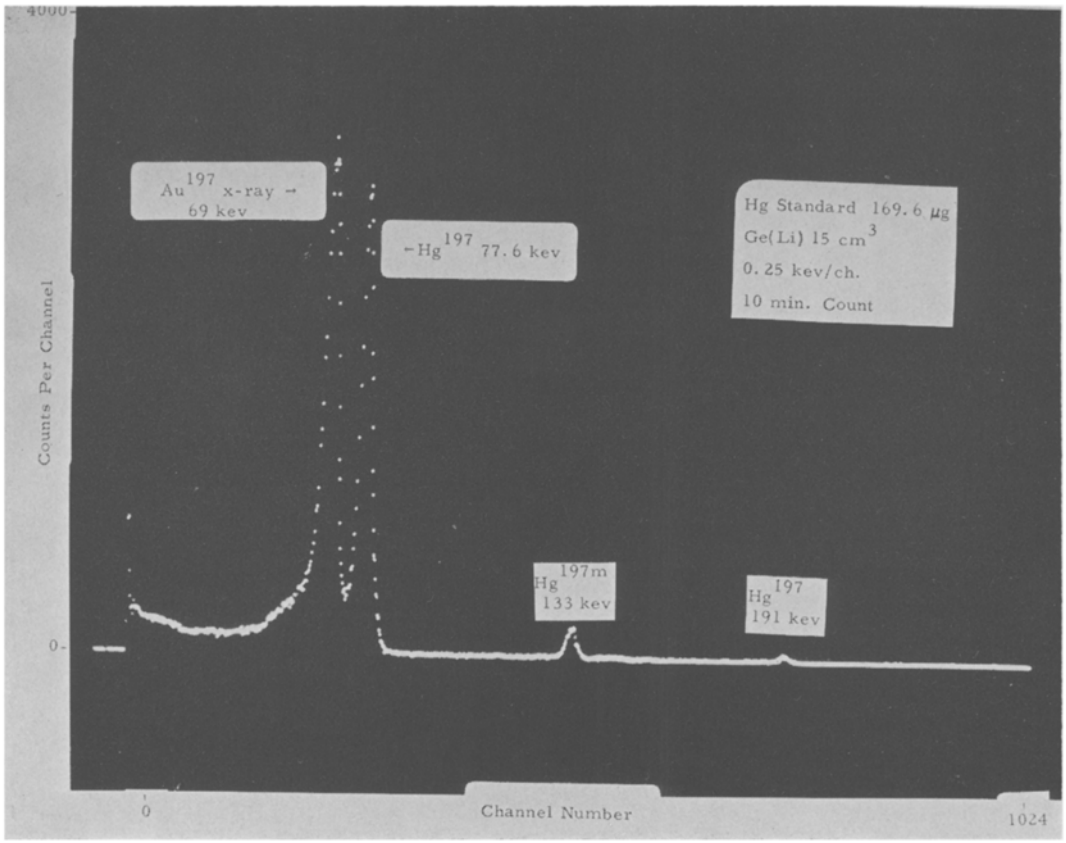

FIG. 4. Ge(Li) pulse-height spectrum of a neutron-activated aqueous mercury standard.

though not conclusive, that the trace elements, Cr, F, Ni, Si, V, Cd, Al and Sn, may also play essential roles in human biochemistry (5). As may be gleaned from the sensitivities listed in Table I, all of these 15 elements can be sensitively determined by means of high-flux NAA with thermal neutrons:

NAA Limit of Detection	Elements
$4-9\times10^{-6}$ μg	Mn
$1-3\times10^{-5}$	Co
$4-9\times10^{-5}$	
$1-3\times10^{-4}$	V, Cu, I
$4-9\times10^{-4}$	
$1-3\times10^{-3}$	Al, Se
$4-9\times10^{-3}$	Si, Ni, Cd
$1-3\times10^{-2}$	Zn, Mo, Sn
$4-9\times10^{-2}$	
$1-3\times10^{-1}$	F, Cr

Excluding the stable inert gases (He, Ne, Ar, Kr, Xe), and the appreciably radioactive natural elements (Po, At, Rn, Fr, Ra, Ac, Th, Pa and U), one notes that the earth's crust consists of 76 elements. Of these 76 elements, 10 occur in the human body as main structural elements (O, C, H, N, Ca, P, K, S, Na and Cl, arranged in descending order of percentage in the body), and 2 more occur at near-trace levels (Mg and Fe) (5). The remaining 64 elements occur in the body at trace levels, or may occur there at trace levels. Of these 64 trace elements, as mentioned above, 7 are definitely known to be present in the body as essential trace elements, and 8 more are known to be present probably as essential trace elements. But, of the remaining 49 trace elements, extremely little is known (6). For most of these, we do not even know at what levels they commonly occur in the body, or in various particular tissues or organs of the body, nor the normal ranges of their concentrations. Certainly, for these 49 trace elements, we do not know which ones, if any, perform essential biochemical roles (usually, as co-factors in enzyme reactions). It seems very likely that a number of these essentially unstudied trace elements, upon careful study, will later prove also to be essential trace elements. Since these elements all occur in the body at levels lower than 1 ppm, sensitive analytical techniques are obviously needed in such studies. High-flux NAA is, for most elements, outstanding with respect to sensitivity, especially quantitative concentration sensitivity, so it is frequently either the method of choice, or the only applicable method. For this reason, this technique is being used in a number of laboratories in the United States and abroad, including the author's laboratory, to further extend our knowledge of trace elements in biomedical systems.

It should be noted that, to attain the kinds of limits of detection shown in Table I, in biological samples (vegetable or animal), it is for most elements necessary to employ post-irradiation radiochemical separations, to remove higher level interfering activities. Such separations can be rather tedious if one is endeavoring to detect and measure quite a number of trace elements in each sample. Recently, a very promising automated radiochemical separation apparatus has been developed (7), capable of automatically separating an activated, wet-ashed, biological sample into 16 different groups of trace elements, representing a total of 50 elements. These 50 elements are distributed amongst 16 different ion-exchange and chromatographic columns and solutions, each one essentially containing only 1–3 elements,

except for one column that contains lanthanum and all the rare-earth elements. The contents of each of the 16 columns and flasks can them be counted on a γ-ray spectrometer. The separation time, per sample, is about 2 hr. An apparatus of this type, obtained from Sweden, is now being tested and evaluated in the author's laboratory. The results to date are quite promising.

Major and Minor Elements in Foods

All of the attention in this paper, up to this point, has been devoted to the use of high-flux NAA for the determination of trace levels of certain elements in foods or other biological materials: pesticide residues, other toxic element contaminants, essential trace elements and other possibly essential trace elements. Another type of application of NAA to problems concerning foodstuffs is that of determining nitrogen. At the percentage nitrogen levels found in many protein-containing foods, the nitrogen content can be determined quite accurately in a few minutes per sample. The analysis is based upon the formation of 9.96-min N^{13}, a pure positron emitter, via the $N^{14}(n, 2n)N^{13}$ reaction. This is not a thermal-neutron reaction, but rather is a fast-neutron reaction, requiring neutrons of at least 10.5 Mev energy to cause the reaction to take place, since it is endoergic to this extent. The most convenient source of fast neutrons is a small Cockcroft-Walton deuteron accelerator, which forms 14 Mev neutrons by bombarding a tritium target (typically 5 c/sq in.) with a beam of 150 kev deuterons (typically 1–2 milliamperes). The target reaction is the highly exoergic (17.6 Mev) $H^3(d, n)He^4$ reaction. Such small generators, capable of producing, isotropically, about 2×10^{11} n/sec (corresponding to an average flux at a 10-cm^3 sample location of about 10^9 n/cm^2-sec), cost about $20,000. The lower limit of detection of nitrogen by this method is about 100 ppm (0.01%).

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