

## Chapter 2

# Methods and Materials of Analysis

The research work has been carried out in Greece, mainly in the region of Northern Greece as well as its neighboring geographical regions. The research work aforesaid has been made during the period 1989–1998.

Analyses have been made in the laboratory of Applied Physics at the Alexander Technological Educational Institute of Thessaloniki, which is under the control of the Greek Atomic Energy Committee (GAEC) as well as in the laboratory of the Environmental Radioactivity Measurements, “Demokritos” NRC, Athens.

A great deal of information from data and found correlations, obtained in the world laboratories, has been used in the research work.

- *Object of Research.* Air, soil, plants, food.
- *Methods of Work and Analyses.* Samples are taken and processed according to international accredited methods, ensuring of their state and contents when being studied in the laboratory.

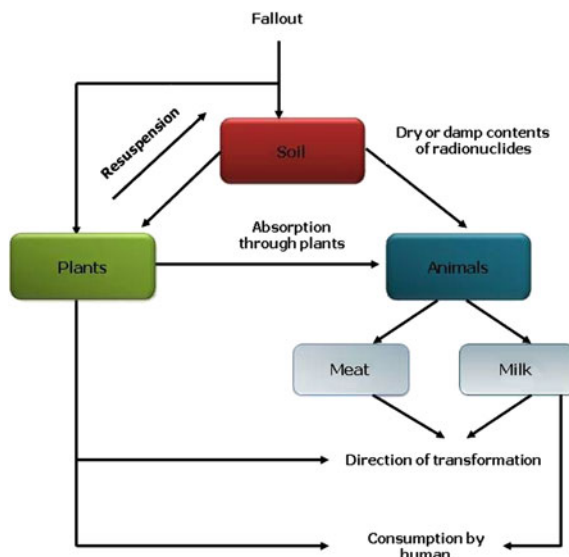
A great number of the airborne radioactivity measurements in Northern Greece for the period 1981–1991 have been obtained. On the basis of the obtained data from studies of the basic physical parameters—temperature, relative humidity, wind speed, atmospheric pressure of the air and rain, a comparison to previous periods as well as anticipations about the future state are made by application of mathematical models.

For estimation of the radioactivity level in a given region, a systematic method has been used, where the distribution is estimated by measurement of the radioactivity on the surface soil Fig. 2.1.

### 2.1 Methods for Analyses of Atmospheric Radioactivity

Atmospheric radioactivity and the airborne nuclides have been studied by measuring the radioactivity of the dust falling on the earth through filters (IAEA 1989). A constant flow rate air suction pump of AEG, operating at  $1.1 \text{ m}^3\text{h}^{-1}$ , equipped

**Fig. 2.1** Contamination principal transfer pathways in the environment



with paper filters “Whatman—41”, mounted at air inlet position, was used as the radionuclide collection device in Thessaloniki (40.5°N, 22.9°E). The filters are usually compressed to provide a standard counting geometry and are measured by gamma-spectroscopy. The spectroscopy system consisted of a high purity coaxial Germanium detector *p*-type (CP—2100 Tennelec). The sample chamber was a cylinder 12 cm in diameter and 25 cm in height and it was shielded by 5 cm of lead and 0.5 cm of copper in order to minimize the background. The full width at half maximum (FWHM) of the system was found to be 1.95 KeV at 1332 keV of  $^{60}\text{Co}$ . The linearity of the detector was checked with a  $^{152}\text{Eu}$  source and a simple regression analysis gave a straight line with a correlation coefficient of 0.999 [7, 76, 141, 182].

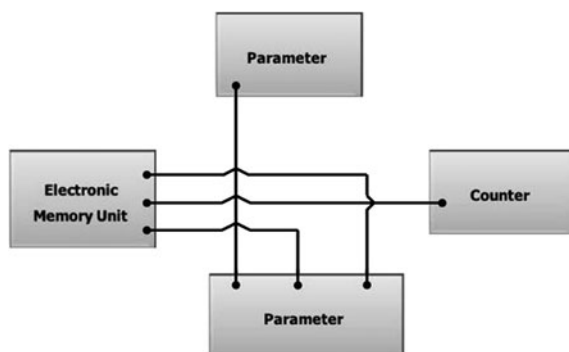
A radioactivity measuring device was also used and it has been collecting data permanently since 1981 for an industrial area where no nuclear stations exist. The study of the radioactivity transfer is complied with the illustrated device in Fig. 2.2.

### 2.1.1 Diffuse Radioactivity Measurements

The diffuse radioactivity measurements and the radionuclides identification is made by gamma-ray spectroscopy for identification of  $^{103}\text{Ru}$ ,  $^{106}\text{Ru}$ ,  $^{134}\text{Cs}$ ,  $^{137}\text{Cs}$  and  $^{131}\text{I}$  presence in the atmosphere 10 days after the arrival of the cloud, in regions endangered by contamination to a greater extent (Sindos—a suburb of Thessaloniki).

Two Geiger-Muller detectors—one for  $\alpha$ ,  $\beta$ ,  $\gamma$ , and the other for  $\beta$ ,  $\gamma$  radiation were used with a Leybold Heraeus counter-amplifier system. The detectors were positioned at 3.5 m height from ground. Measurements were made hourly on a 24 h basis and were being stored into an electronic memory unit connected to the

**Fig. 2.2** Diffuse radioactivity measuring facility



measuring equipment (Fig. 2.2). Recorded values were in counts per minute (cpm) and were used to estimate the mean daily values for the years 1981–1987.

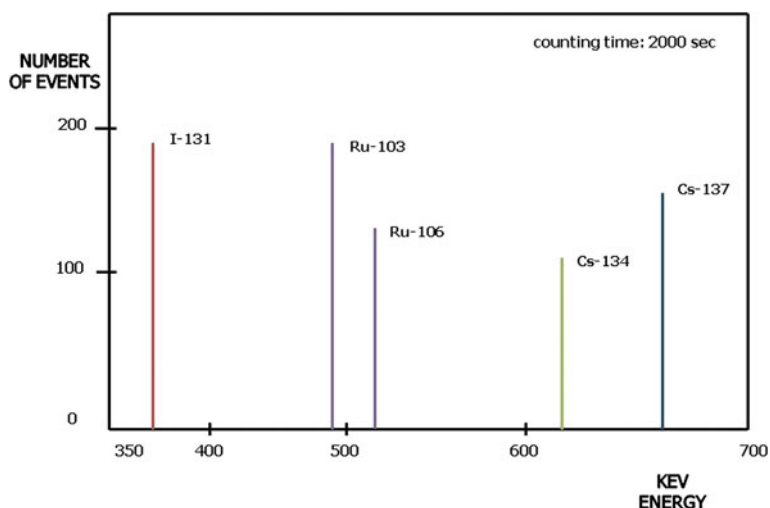
### ***2.1.2 Radionuclides Detection and Identification***

A suction pump of AEG, equipped with paper filters “Whatman–41”, mounted at air inlet position, was used as the radionuclide collection device. A time switch circuit was used with the pump to ensure an 8/43 working time over cease time ratio. The suction rate was measured accurately and found equal to  $1.1 \text{ m}^3\text{h}^{-1}$ .

Whatman filters have been analyzed using the  $\gamma$ -ray spectroscopy technique. The associated facility consisted of a Pb-shield, NaI crystal, a photomultiplier tube, a Caberra multichannel analyzer, a preamplifier and a linear amplifying unit, a computer and a printer. Computer printouts contained energy peaks, each peak being associated with a specific radionuclide (Fig. 2.3). The location of each discrete peak on the KeV energy scale, defines the energy of the specific gamma that has created the peak in question. This gamma is characteristic of a particular radionuclide, which is then identified by referring to tables of gamma ray spectroscopy. Finally the total number of events for each peak enables the estimation of the concentration of each radionuclide in air by making use of an empirical relationship as shown in the discussion [190].

### ***2.1.3 Correlation with Meteorological Parameters***

A complete meteorological station has also been working since 1981 alongside the radioactivity measuring facility. A data bank of parameters such as ambient temperature, relative humidity, atmospheric pressure and wind were elaborated with the radioactivity data bank to give corresponding mean daily values for the years 1981–1987. A computer program has been used to correlate radioactivity and meteorological parameters on the basis of a linear variation [191].



**Fig. 2.3** Spectrograph, showing the existence of radionuclides in air

A spectrograph, showing the existence of radionuclides in air has been used (Fig. 2.3).

Anticipation of the environmental contamination level is made on the basis of data of the meteorological conditions and the sources of contamination. On the basis of that transitory and lasting forecasts have been developed.

The total penetration and transfer of the air contaminants is found by simultaneous measurement of the concentration of  $\text{SO}_2$ ,  $\text{O}_3$ ,  $\text{NO}_x$  in the air in the industrial area of Thessaloniki, the meteorological parameters and the concentration  $C(o)$  of  $\text{SO}_2$  on the ground surface for a definite point of the region [182].

A spectrophotometer BREWER MARK IV has been used for measuring the intensity of solar ultraviolet radiation in regions with wavelength in which the absorbing spectra of  $\text{SO}_2$ ,  $\text{O}_3$ , and  $\text{NO}_x$  have a special configuration. For calculations of the total amount through the measured intensities, there has been used database of A. W. Brewer [7] and the atmospheric contamination team in Canada [15].

## 2.2 Methods for Analysis of the Impact of Atmospheric Contamination on the Food Chain (Soil, Plants, Foods)

### 2.2.1 Analysis of Soils and Plants

Diluting the elements in water and then mixing them with the ground surface study the relative uptake of various radionuclides from soils. Various concentrations of elements which were present in the tested crops were examined before and after

soil contamination, as follows: strongly concentrated 10–1,000; slightly concentrated 1–100; non-concentrated 0.1–10; slightly excluded 0.01–1; strongly excluded <0.01 [20, 138].

For determination of the aqueous radioactive contamination, the so-called concentration factor was used, i.e. the ratio between the concentration of a given element in a certain organism and its concentration in water. On the basis of a large informational material, there have been worked out tables for concentration factors (CF) of various classes of fresh water organisms.

Natural radioactivity in the soils in Northern Greece is studied from soil samples, taken from 0 to 20 cm in depth, by special metallic samplers. Samples have been dried, pulverized and sifted through a 2 mm sieve. They were then sealed in small cylindrical pots, made from urea. Samples stayed thus for at least 3 weeks, in order that radioactive equilibrium was obtained between the studied isotopes  $^{226}\text{Ra}$  and  $^{222}\text{Rn}$ . A highly resolution gamma-spectrometer for measurements was used with a detector HpGe (Tennelec Coaxial Ge Detector System) with relative efficiency of 20% and resolution (FWHM) 1.8 keV of  $^{60}\text{Co}$ . The detector was connected with a 4,096-channel analyzer. The measuring time for soil samples was usually 76,000 s. Activities are calculated in  $\text{Bq kg}^{-1}$ , by using data from the spectrographs. Depositions were calculated in  $\text{Bq m}^{-2}$  using activities, soil densities and hypothetical depth of 1 cm. The absorbed external dose rates in air were calculated by using the conversion factors [119].

Statistical analysis uses the methods of descriptive statistics as well as these of deductive statistics. And especially the average values, the standard deviations, maximum and minimum values were found for Macedonia and Thrace, as well as for each country separately. Then there were made tests of single groups for estimating the statistical differences of mean levels of activity concentrations.

The level of  $^{137}\text{Cs}$  in the soils in Greece one decade after the Chernobyl accident was measured during the period January 1993–May 1995 on 380 soil samples from the soil surface of 5 cm in depth (Fig. 2.4).

All the samples were sealed in containers for at least 15 days in order that equilibrium of isotopes  $^{226}\text{Ra}$  and  $^{222}\text{Rn}$  was obtained [225, 226].

The taken soil samples after being dried were weighed and measured in Marinelli beakers with gamma-spectroscopy system, consisting of a coaxial Germanium detector of high purity, *p*-type (CP 2100, Tennelec). The sample chamber was a cylinder 12 cm in diameter and 25 cm in height and it was shielded by 5 cm of lead and 0.5 cm of copper, ensuring lowered background. The full width of a half maximum (FWHM) of the system was found 1.95 keV at 1,332 keV of  $^{60}\text{Co}$ . The linearity of the detector was checked with  $^{152}\text{Eu}$  source and a simple regression analysis gave a straight line with a correlation coefficient of 0.999. The radionuclides used were supplied by “The Nucleus”, Oak Ridge, USA. The measuring time was 86,400 s for all soil samples, in order for the time to be sufficient to measure almost any level of radioactive contamination (from 0.1  $\text{kBq/m}^2$ ).

Firstly, the activity per unit mass is evaluated for each sample. The activity per unit area ( $\text{Bq/m}^2$ ) was calculated afterwards by assuming the soil sample volume

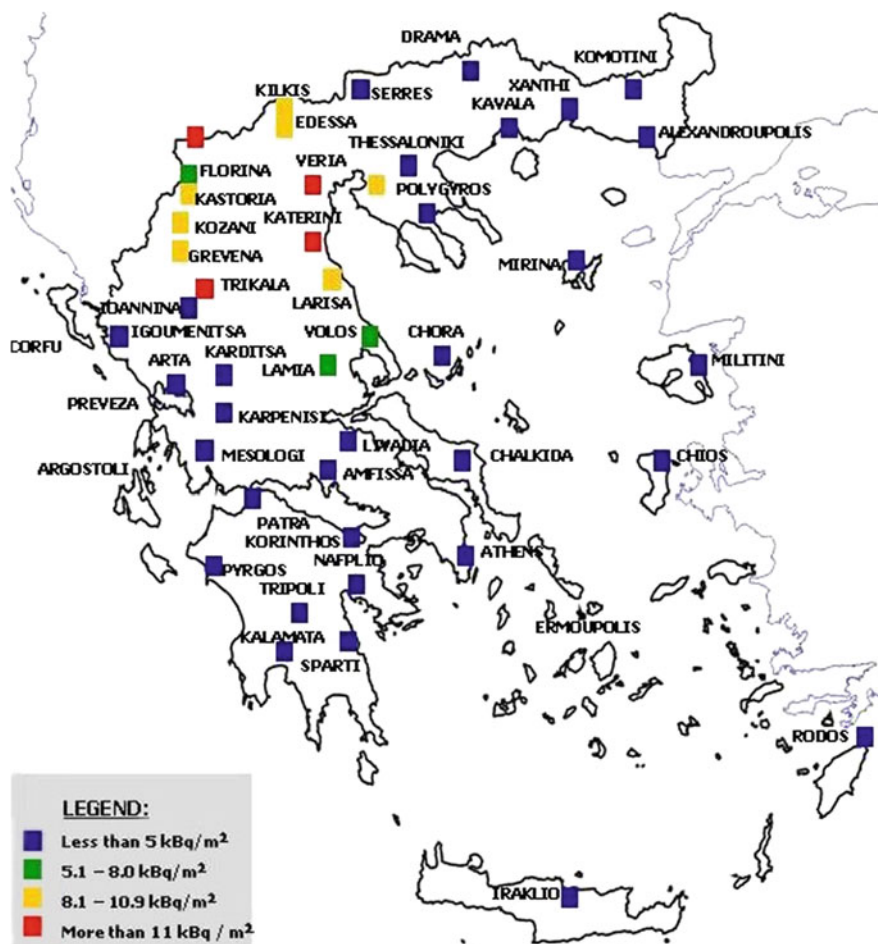


Fig. 2.4 Map of radioactive contamination in Greece

equal to the surface of a layer with depth 1 cm. The mean value and the standard deviation of the density of the 380 samples was  $1.43 \pm 0.23$  Kg/L.

The soil contamination in Greece during the period January 1993 to June 1997 was studied. For that purpose, 742 samples were taken from soil surface (0–5 cm) and from deeper soil layers (5–50 cm), in which the expected mobility of Cs was slight ( $0.2 \text{ y}^{-1}$ ). The sampling of surface soil of about  $500 \text{ cm}^3$  each, were taken from geographic divisions of Greece with emphasis to those where in 1986 serious depositions of  $^{137}\text{Cs}$  (from  $15 \text{ kBq/m}^2$  and more) were observed [20, 138, 181].

Samples were studied according to the aforesaid method.

For comparison of the levels of soil contamination according to data during 1986 and 10 years after the Chernobyl accident, the level of  $^{40}\text{K}$  (0.0118% of natural potassium in the same soils) was studied [225].

The levels of  $^{137}\text{Cs}$  and  $^{40}\text{K}$  were compared in the grown plants in 1986 as well as in the cultivated crops during the radioactive fallout. The measuring system was calibrated for the energy using standard sources of  $^{137}\text{Cs}$  and  $^{60}\text{Co}$ , supplied by “Nucleus”, Oak Ridge, USA. The ratio channel-energy was linear, with proportionality coefficient 0.99999.

The activity was calibrated with standards dry soil and dry plants of a known activity (Bq), supplied by the National Research Center in Greece (Demokritos).

In September 1998 samples were taken from corn and clover and soil samples from cultivated land (root depth) in the area of Thessaloniki. Plants were separated from their roots and grain, thoroughly washed, air-dried and oven-dried (80°C). Then they were weighed (dry plant mass) and burned in another oven (Nabertherm) at 500°C for at least 24 h to get carbon white ash. This was done because Cs in a certain volume of ash is 8–9 times more than the Cs in the same volume of dry plant. It should be mentioned that Cs is not lost by oven drying at 500°C (boiling point of Cs and K, 687°C and >750°C). Plant ash was filled in the standard geometry white cup (cylindrical, 7 cm diameter and 2 cm height), recommended for measurement by the Greek National Research Center–Demokritos).

Soil samples were also air-dried, oven-dried (80°C), pulverized, sifted (2 mm sieve) and weighed (mass dry soil). Then they were put to fill the white standard cups. Measuring method: gamma-ray spectroscopy [141].

The concentrations of  $^{137}\text{Cs}$  and  $^{40}\text{K}$  were measured in  $\text{kBq/m}^2$  dry plants. Soil samples were prepared according in the aforesaid manner and their concentrations were measured in  $\text{kBq/m}^2$ .

For the study of  $^{137}\text{Cs}$  and  $^{40}\text{K}$  transfer soil-plant in the areas Drama (Eastern Macedonia) and Thessaloniki (Central Macedonia), the corn was taken for its being the main food of cows in the country [10].

Samples were taken in November 1998–1915 plants in total. Each plant was taken out carefully so that it remained intact (roots, plant, grain), as well as the soil round its roots. Plants were gathered when ripe and prepared for analysis according to the manner aforesaid [226].

### 2.2.2 Foods Analysis

Studies of radioactive contamination of foods have been made: of cow milk, milk products, cheese, curd and yogurt and also typical representatives of freshwater fish [186, 187, 188, 189].

The determination of the radioactivity of cow milk to cheese curd, cheese whey, mytzhithra, and lactose serum was made by application of gamma-spectroscopy. The spectroscopic system consisted of:

- *Detector.* NaI (TI) scintillator  $2'' \times 2''$ .
- *Shield.* 50 mm Pb + 5 mm Cu cylinder.
- *Analyzer.* Canberra 8100 (USA), multi-channel analyzer with 4,096 channels.

- *Resolution (FWHM)*. 70 keV approximately for  $^{137}\text{Cs}$  or 10.5%.
- *Geometry of the chamber*. cylindrical, dimensions 120 × 250 mm.

The efficiency of NaI (TI) was determined by a set of gamma-ray standard sources ( $^{152}\text{Eu}$ ,  $^{134}\text{Cs}$ ,  $^{137}\text{Cs}$ ), supplied by Amersham. For the energy calibration the following radionuclides were used:

$^{131}\text{I}$	364.5 keV
$^{137}\text{Cs}$	661.6 keV
$^{134}\text{Cs}$	604.7, 795.8 keV
$^{152}\text{Eu}$	122, 245, 344, 427.5, 779, 964, 1,099, 1,408 keV

The radionuclides used were supplied by “The Nucleus”, Oak Ridge, USA.

Fats determination in the milk, the cheese whey and the lactose serum were done according to Gerber’s method (standard 1/1963 IDF). The Shmid Bondzinski process (standard 3/1963 IDF) was used for the curd and the mytzithra. The Kjeldahl method (standard 20/1962 IDF) [86, 97] was applied for determination of the total amount of protein and non-protein nitrogen (N. P. N.). The proteins content was calculated by using a nitrogen conversion factor of 6.38. The spectroscopic method was used for the determination of lactose [113, 136]. The ashes as well as the total solids in the milk, in the cheese whey and in the lactose serum were determined according to standard methods (British standard 1741/1963, standard 21/1962 IDF). In the case of cheese curd and mizithra, the ash and the total solids and NaCl were determined by standard methods (standard 27/1964 IDF, 4/1963 IDF, standard 17/1961 IDF).

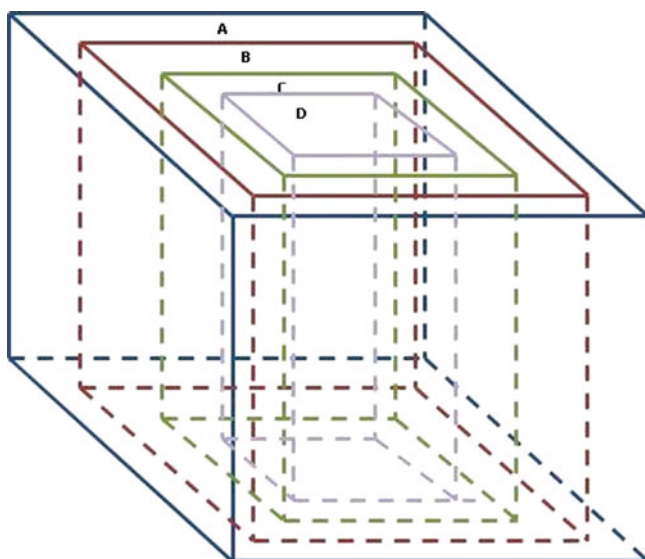
The fresh chilled raw milk was supplied by the experimental farm of the Technological Institution of Thessaloniki. The method used for the production of “Feta” cheese was done according to Kosikowski’s procedure [104]. Yogurt from the market was used as a starter and the curdling was accomplished by adding artificial rennet (Rennilase 150 L, Novo, Denmark). Mytzithra was traditionally produced according to the following procedure: A mixture from cheese whey and milk, in a 9:1 ratio is heated for 45 min at 90°C, pH being fixed to 5.4 by adding 10% solution of citric acid. The obtained curd is heated for about 20 min until it reaches 90°C and then it is pressed for 16 h at room temperature.

Two experiments have been carried out for determining  $^{131}\text{I}$  transfer from “Feta cheese” to clear water or brine and conversely.

The first experiment includes “Feta cheese”, which had been made in the experimental laboratory of the Alexander Technological Educational Institution of Thessaloniki from contaminated with  $^{131}\text{I}$  cow milk (10–15 kBq/kg).

Portion of the cheese, 6 × 6 × 6 cm sized were divided into 4 zones and immersed in different solutions of NaCl, KCl and in tap water. The immersion time varies from 5 to 48 h. According to the immersion, there have been outlined four zones of the cheese, 0.75 cm thick from external to inner surface (see Fig. 2.5). These four zones have been separately removed and homogenized in a blender and the radioactivity was measured into 50 g samples for 500 s.





**Fig. 2.5** Section of the cheese layers

The second experiment was carried out with “Feta” cheese, which had been bought from the market. The cheese pieces used were of the same size as in the first experiment. Three zones, 1 cm thick were formed after immersing the cheese in solutions of NaCl, KCl and tap water, previously contaminated with  $^{131}\text{I}$  ( $10\text{--}12\text{ kBq kg}^{-1}$ ). The cheese samples were examined with respect to their radioactivity as in the first experiment.

$^{131}\text{I}$  impact on the lactic acid microflora of the yogurt was examined in two varieties of yogurts: the set yogurt and the strained yogurt. Milk was standardized with respect to its fats content (3.9% for cow milk and 6.00% for sheep milk), it had been boiled for 5–10 min, cooled to  $45^\circ\text{C}$  and inoculated with 2% starter culture from *Thermophilus* and *bulgaricus* [37, 38, 151]. Milk had been inoculated for 3 h at  $45^\circ\text{C}$  temperature. The fermented milk had been consequently stored in refrigerator at  $4\text{--}5^\circ\text{C}$  for 12 h. The yogurt variety was obtained by straining the tissue matter, upon which a part of the serum was retransferred [16, 37, 38, 151].

For decontamination of milk from radionuclides ( $^{131}\text{I}$ ,  $^{134}\text{Cs}$ ,  $^{137}\text{Cs}$ ), ion-exchange resins for the unit chosen for this purpose were used. The elements of this decontamination unit has been designed according to the calculations for a capacity of about 5,000 l milk per day at constant flow [197].

The selection of the resin was made taking into account:

- The avoidance of the entrance of harmful substances in milk
- The avoidance of organoleptic changes, and
- The capability of ion-retainment.

Amberlite IRA 120 Plus and Amberlite IRA 904 as cationic and anionic exchangers were used respectively. Their grain size of the resin was chosen so that to allow the liquid pass freely through the columns, without causing changes in the pressure, which had not been foreseen upon designing the unit. After running out, the resin should be regenerated according to the manufacturer's instructions. Regeneration of the cationic resins was obtained through their passing in a diluted solution of HCl (about 6%), and the anionic resins with respective solution of NaCl.

A fixed bed anion-cation removal method has been used for the contamination, which uses columns with ion-exchanger for the accomplishment of the final purpose.

For the laboratory tests glass columns were used with the respective ion-exchangers, as well as cow milk with the following chemical analysis:

Total acidity	17.00°D
Proteins	3.06%
pH	6.62
Fats	3.56%
Lactose	5.52%

Similarly, for long-life skim milk which was also used, the characteristics were: total acidity: 15.5 (°D); fats: 1.5%; proteins: 3.5%.

The experimental procedure followed the steps:

- Adding solutions of radioactive iodine ( $^{131}\text{I}$ ) and radioactive cesium ( $^{137}\text{Cs}$ ) in ionic form in the milk. The concentration of the radioactive substances added was about 1,000 times higher than it was expected to be in order to control the effectiveness of the method. The manufacturer of  $^{131}\text{I}$  was N.R.C. Demokritos (Greece) and the one of  $^{137}\text{Cs}$ —the Nucleus Company (USA).
- Heating to about 30°C.
- Passing through the column of the anionic resin ( $^{131}\text{I}$  removal).
- Acidifying with a diluted solution of citric acid to pH 5.3. The avoidance of excess of this value is very important in order to avoid the formation of colloids, which impede the normal flow of the milk through the column, but also deterioration of the milk, as it has been observed during the experimental work.
- Passing of a cationic ion-exchanger through the column (removal of  $^{134}, ^{137}\text{Cs}$ ).
- Neutralization of milk by diluted solution of KOH (pH 6.5)
- Radioactivity control
- Quality control of milk.

Comparative studies were carried out in two cultured fresh-water fish, *Cyprinus carpio* (carp) and *Anguilla anguilla* (eel), to determine their tolerance in the uptake of  $^{137}\text{Cs}$  (3,000 Bq/l) [280].

The histological studies were concentrated in muscular tissues, livers, kidneys and gills. The symptoms observed include hypothermia, and gradual degeneration of liver and kidney tissues. The physiology and anatomy of each species played an important role in the accumulation process of  $^{137}\text{Cs}$ .

The determination of  $^{137}\text{Cs}$  was made by  $\gamma$ -spectroscopic system, which was described previously. Fish were cultured in small water tanks, artificially contaminated with radioactive  $^{137}\text{Cs}$ . The species *Anguilla anguilla* were collected from artificial ponds 2 days before the experiment. The fish *Cyprinus carpio* were collected from a local lake. They were placed in a 200 l tank, supplied with good aeration and a continuous through flow of tap water, which had been dechlorinated by active carbon. The dimensions of the water tanks used were 79 cm in length, 35 cm in width and 50 cm in height.

The fish were sacrificed everyone or 2 weeks, weighed, their length was measured and their overall conditions were compared to the control. These observations were recorded and the radioactivity of a few organs was measured. The samples of the organs were fixed with 10% formaldehyde solution, embedded in paraffin wax and sections of 5–10  $\mu\text{m}$  thickness stained with Erlich's hematoxylin-eosin, to be examined under the microscope.

The results were processed by modern computer technologies. On the ground of that, condition models were obtained for forecasting the changes in radioactive contamination and the radioactivity transfer through the chain air-soil-plants-foods. The results and the found correlations are presented in tables and figures.



<http://www.springer.com/978-3-642-28740-4>

Radioactivity Transfer in Environment and Food

Vosniakos, F.K.

2012, XVI, 148 p., Hardcover

ISBN: 978-3-642-28740-4