

Chapter 16

Biomarkers of Radiation and Risk Assessment by Ionizing Radiation, Countermeasures for Radiation Protection of Environment, Workers and Public



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Abstract Institute of Applied Nuclear Physics (IANP) is main responsible institution in country for managing of methodologies and techniques used for environmental/epidemiological samples collected for levels of radiation monitoring and radionuclide identification activities. In Albania, was well established the legal framework of legislation and regulations for using of radioactive materials: Law No. 8025, date 9.11.1995 “*On Ionizing Radiation Protection*”, amended No. 9973, July 28th 2008. Also, the process of establishment of more laws, regulations, code of practices in radiology, nuclear medicine etc., started based on IAEA documents, elements of Joint Convention, as well as part of Interregional Projects & EU Directives, about all issues related to the policy issues for the application of biomarkers in the field of human health. Both types, of environmental and human body samples, are as indicators of biological markers, signaling events in biological systems and those are classifying into three types: [a] those of exposure, [b] biological effects, and [c] susceptibility. Once exposure has occurred, a continuum of biological events may to be detected. These events may serve as markers of the initial exposure, internal dose, biologically effective dose and some other parameters for evaluation of exposures. Even, before exposure occurs, there may be biological differences, between humans that cause some individuals to be more susceptible to environmentally induced disease. Biomarkers, therefore, are robust tools that can be used to clarify the relationship between ionizing exposure radiation & environment health impairment. The Institute studied in collaboration with Working Hygiene and Epidemiological Research Department in Institute of Public Health (IPH) in Albania, the effects of radiation caused by Chernobyl & Fukushima NPP accidents, carried out: radiation level monitoring, radionuclide identification activities, analyses of environmental human body laboratory samples to the individual workers and public exposed to radiation. Also, was assessed the ambient monitoring using chemical or physical analyses of food, air, water, soil etc., coupled with measurement for

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M. D. Wood et al. (eds.), *Biomarkers of Radiation in the Environment*, NATO

Science for Peace and Security Series A: Chemistry and Biology,

https://doi.org/10.1007/978-94-024-2101-9_16

estimation of actual human intake at these areas, and by biomarkers of exposure was study the effects in body fluids such as: blood, urine, saliva, or some limited samples for reproductive and developmental systems, follicular fluids, cells, and semen. It is known, the thyroid cancers attributed to ^{131}I radioisotope exposure, as well as by other radionuclide's which have contaminated the environment, and for that reason it was important to evaluate patterns of excess absolute and relative risks by external/internal irradiation over time.

Keywords Radiation exposures monitoring · Biomarkers for human health · Biological effect · Environment contamination

16.1 Introduction

16.1.1 *Challenges for Establishment of Albanian Legislation and Regulations According EU*

In the Republic of Albania is well established the legal framework with laws and regulations involving practices and applications of using radioactive materials or devices in medicine, researches, agriculture, industries, environment protection/control and education for the safety, security and radiation protection from ionizing exposure radiation. Based at the main Law No. 8025, date 9.11.1995 “*On Ionizing Radiation Protection*”, amended No.9973, July 28th 2008; some other important regulations are approved by Albanian institutions:

- Regulation on “Safe management radioactive waste in Republic of Albania”, Decision No. 08, date 07 January 2010 of Council of Ministers.
- Regulation on “Categorization of radioactive sources in the Republic of Albania”, Decision No. 09, date 07 January 2010 of Council of Ministers
- Regulations on “Licensing and inspection of activities with sources of ionizing radiation” Decision No.10, Date 07 January 2010 of Council of Ministers.
- Regulation on “Safe transport of radioactive materials”, Decision No. 488, date 23 June 2010 of Council of Ministers.
- Regulation on “Safe handling with ionizing radiation sources”, Decision No. 543, Date 7 July 2010 of Council of Ministers.
- Regulation on “Physical protection of the radioactive materials in the Republic of Albania”, Decision No 344, date 29 April 2011 of Council of Ministers.
- Regulation on “Protection of the employees professionally exposed to ionizing radiation sources” Decision No 590 date 18 August 2011 of Council of Ministers.
- Regulation on “The permitted levels of the radon concentration on buildings and water, guide levels of radionuclide’s building materials, as well as permitted levels of radionuclide’s in food and cosmetic products”, Decision No 591 date 18 August 2011 of Council of Ministers.

- Regulation on “Public protection from the discharges in the environment, determination of sampling, regions and frequency of measurement” Decision No 313, date 09 May 2012 of Council of Ministers.
- Regulation on “Public safety to exposures caused from ionizing radiation sources” Decision No 481, date 25 July 2012 of Council of Ministers.
- Regulation on “Safety on Medical exposure with ionizing radiations” No 229 date 20 March 2013

The process of establishment of more laws, regulations, code of conduct, code of practices in radiology, nuclear medicine etc., started based on IAEA documents, elements of Joint Convention, as well as part of Interregional Projects & EU Directives, about all issues related to the policy issues for the application of radioactive materials including biomarkers in the fields of environment and human health monitoring. So, for instance at the Regulation No. 481, date 25.7.2012 “*Public protection from exposures by ionizing radiation devices*”, was improved our understanding to the diseases, providing new knowledge of disease mechanisms and processes providing a means for improved health management through the earlier diagnosis of disease and the delivery of more efficacious and safer therapies. In fact our above mentioned laws and regulations needed to be in accordance with the directives of the European Parliament Commission, as well as with the European Economic and Social Committee and the Committee of the regions documents, especially: “Safe, Innovative and Accessible Medicines” which as the main focus have on the developing technologies and tools for gathering information on various classes of biomolecules, biomarkers and understanding relationships among them, including the related regulatory mechanisms.

16.1.2 Ionizing Radiation Exposures and Determining Risk Health

Ionizing radiation is a known carcinogen but the magnitude of health risk at low doses and dose-rates, for instance below 100 mSv or 0.1 mSv min⁻¹, remains controversial due to a lack of direct human evidence. Epidemiological studies of radiation exposed populations can provide evidence of risk. Much more information is needed for interpreting ionizing radiation measurements for determining health risks, because several factors should be considered before making qualitative or quantitative evaluation of exposures by ionizing radiation. For example, the concentration, duration and the time of exposure, and physicochemical nature of the radioactive agents are all relevant to the selection of an appropriate marker of the ionizing radiation exposure. Specialists have proposed that there are two key factors governing interpretation of radiation measurements: [1] measurements have no meaning until interpreted and [2] measurements only have meaning in terms of how they are interpreted. Thus, recorded or reported radiation measurements have no inherent meaning by themselves, they are just numbers.

Radiation safety specialists have the advantage for interpreting radiation measurements based on knowledge of comparative readings from background and other sources. Most people without this specialized knowledge do not know that we live in a sea of radiation, which surrounds us all the time. Furthermore, screaming dosimeter instrument may sound alarming but radiation risks depend on many other factors, such as the type of radiation and the duration of exposure (Jonson, 2014).

16.1.3 General Considerations on Biomarkers for Use in Epidemiological Studies

Biomarkers allow new ways of understanding disease processes and the ways in which medicines work to counteract disease. Within the practice of evidence-based medicine this knowledge can be used to improve disease diagnosis, to improve the safety and efficacy of existing medicines and to develop new medicines and targeted therapies. A biomarker has been defined as “*any measurement reflecting an interaction between a biological system and an environmental agent, which may be chemical, physical or biological agent*”. Biomarkers can be used for multiple purposes in epidemiological investigation included: [a] estimation or validation of received dose, thus improving the validity of a correlation between exposure and biological responses; [b] investigation of individual susceptibility; and [c] early detection of a radiation induced health effect (Committee on Biological Markers of the National Research Council, 1987).

Biomarkers are robust tools that can be used to address many different issues confronting environmental health scientists. Biomarkers that indicate the occurrence of an internal dose, a biologically effective dose, or the presence of an incipient disease can be useful in hazard identification, for example, as the qualitative step that causally associates an environmental agent with an adverse effect (Fowle, 1984). Biomarkers can also be used to determine dose-response relationships and to estimate risk, especially at the low doses relevant to most environmental samples. Another major role of biomarkers is clarification of the extent of exposure in human populations. Methods of direct or indirect measurement of total exposure through analysis of body fluids (e.g. IAEA, 1969) are far more likely to be of value in epidemiological studies than are most of the modeling and ambient monitoring approaches now in use. Biomarkers of exposure also hold the promise of demonstrating, which individuals in a potentially affected population (e.g., residents or workers in the neighborhood of a hazardous radioactive wastes facility) have inordinate levels of exposure. Developments in the field of biomarkers are also likely to lead to a more accurate determination of the proportion of highly susceptible people within the population and of the results of human or public exposure (Ashford, 1986). For present purposes, the effects on, or responses of, an organism to an exposure are considered in the context of the relationship of exposure to health impairment or the probability of health impairment. An effect is defined as: an actual

health impairment or (by general consensus) recognized disease; an early precursor of a disease process that indicates a potential for impairment of health; or an event peripheral to any disease process but correlated with it and thus predictive of development of impaired health. A biological marker of an effect or response, then, can be any change that is qualitatively or quantitatively predictive of health impairment or potential impairment resulting from exposure.

16.1.4 Collection and Use of Biological Samples in Epidemiological Studies

Differences among species and individual variations in physiological characteristics such as sex, age, and health status can significantly affect the absorption and distribution of the chemical and its metabolites. Individual response to environmental temperature, such as the ingestion of large quantities of water, also may affect absorbed dose. Blood flow, capillary permeability, transport into an organ or tissue, the number of receptor sites, and route of administration (which determines the path of the parent compound or its metabolites in the body) can all influence internal or biologically effective dose. Exposures to environmental agents are classically evaluated by mathematical modeling based upon assumptions concerning emission sources, environmental fate, and the location of individuals in space and time. Exposures are also evaluated by ambient monitoring using chemical or physical analyses of food, air, water, or soil, coupled with measurement or estimation of actual human intake of these media, and by biological markers of exposure including measurements in body fluids such as blood, urine, saliva, cerebral spinal fluid for reproductive and developmental systems, follicular fluids, amniotic fluids and cells, and semen. Examination of other biological samples, such as hair, feces, or teeth, may prove useful. The use of such biological markers is a more preferable means for accurately estimating exposure than are the more indirect approaches of modeling or environment monitoring (Ashford, 1986; Fowle, 1984).

16.2 Material and Methods of Sampling Analysis in IANP

In IANP exist some different methods and techniques for evaluation and determination of the alpha, beta, gamma nuclides level contents in natural samples or aquatic discharges by research Labs in the country. We are describing our simple methods used, in order to realize the cooperation between above-mentioned institutions, promoting environmental safety and security in natural resources management. The methods used for measurements of the background level, in order to determine the activity/concentration of the component elements (nuclides) by alpha or gamma spectrometry, beta-gamma total measurements, as well as by radiochemistry

separation analysis to the specific nuclides, are consolidated in IANP already (Suomela, 1993b; Suomela et al., 1993; LMRI-CEA, 2006).

16.2.1 Epidemiological Liquid and Aqueous Samples Analyses

The introduction of the European Commission's Water Framework Directive (WFD; 2000/60/EC) established a new era in environmental risk assessment. In addition to incorporating the compliance of chemical quality standards, the key objective of the WFD is the general protection of the aquatic environment in its entirety. This new approach emphasizes the need for an integrated environmental risk assessment and offers the potential for the incorporation of biological effects measures, including the use of biomarkers in this process. A variety of biological samples can be used for biomarker measurements in epidemiological studies, given appropriate ethics approvals and informed consent and do depend on the nature of the internal/external radiation exposure. These include aqueous or fluid biological samples like: urine, blood, saliva and semen or solid biological samples like: faeces, hair, hair follicle cells, and nail clippings. The fluid or aqueous samples are submitted the measurements of gamma nuclides procedures in order to determine their radioactivity (concentration) in epidemiological Lab of IPH. To provide a suitable and repeatedly geometry of the detection (measurements) are used the Marinelli beakers with 500 ml volume for aqueous environmental samples and Laboratory tubes for epidemiological samples. The "Anal-Spec" (system) equipment with NaI (TI) detector ($\text{\O} = 2 \times 2$ inch), as well as the Lab tubes or Marinelli beakers with volumes 5, 10, 50–500 ml volume, putted into the lead shielding place. This apparatus performs the determination of natural/artificial radionuclide, gamma radiation measurement. The system was provided with digital suite (set) for the gamma spectrometry. The device has the multi-channel analyzer, its spectroscopic amplificatory, the high voltage system, memory with a scintillation integral detector. The device has in its structure a standard NaI (TI) detector ($\text{\O} = 1\text{--}2$ inch), and another G-M detector. "Anal-Spec" apparatus was connected with PC system storing in its memory over 74 specters at 1024 channels that it has. The spectra are processed by its TMCA software.

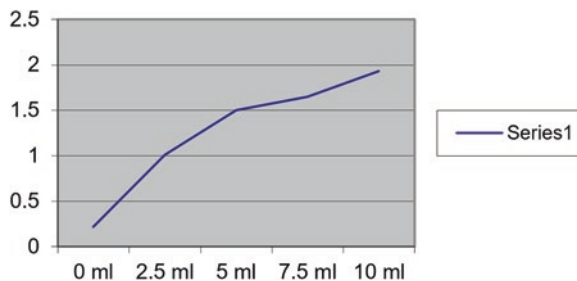
Technical Parameters "Anal-Spec" system are:

- Radionuclide identification and spectrum analysis;
- Multi-channel analyzer its spectroscopic amplificatory, the high voltage system;
- NaI (TI) detector ($\text{\O} = 1\text{--}2$ inch), as well NaI (TI) detector with tungsten protection support etc.

Some other specifications "Anal-Spec" are as below:

- Some other detectors may be used: NaI, BGO, CdWo, CdZnTe, Plastic;
- Selected High – Voltage (HV): 50–1275 volt diapason; Type/Shape: digital filter

Fig. 16.1 Calibration curve established for the results comparison of the aqueous samples



- The energy ranges: NaI (TI) detector 20 KeV–2,5 MeV; Geiger-Muller 60 KeV–1,6 MeV.
- Sensitivity: ^{137}Cs > 10 imp/sec for NaI (1 × 2 inch) detector.

Calibration curve: In beginning of detection of each gamma nuclides was established the calibration curve (Fig. 16.1), using the ^{137}Cs standard solution (LMRI-CEA, 2006) with data as below: *Standard solution:* ^{137}Cs , *Type:* ELE-1, *Vials number:* 228/27703, *Produced by:* LMRI-CEA France on 15 April 2006, *Specific activity:* $A_s = 93, 76 \text{ Bq/g}$ (2%) and $C_c = 0,999 \text{ gr/cm}^3$ (20%).

16.2.2 Environmental Samples Analysis

In cases of a radiological/nuclear a release of short-lived fission and activation products to the environment can be expected. Some of these radionuclides, mainly lanthanides and actinides, will form with HDEHP di(2-ethyl-hexyl) phosphoric acid and consequently interfere with the subsequent counting of the Cerenkov radiation from yttrium-90 (^{90}Y). According to the standard procedure, interfering nuclides such as uranium, thorium, radium and their decay products, as well as isotopes cesium, potassium and strontium are separate from the samples by an extraction with HDEHP. The determination of strontium-90 (^{90}Sr) in equilibrium with yttrium-90, is accomplished by monitoring Cerenkov radiation of high energetic beta particles (2,27 MeV) from yttrium-90 in a liquid scintillator counter. Yttrium-90 is the decay product of strontium-90. The chemical yield of yttrium-90 is determined by adding a known amount of inactive yttrium carrier. The amount of yttrium recovered is determined by acidimetric titration of the sample in the scintillation vial with "Titriplex III". In IANP are in use two methods of determination of strontium-90 in food and environmental samples in the absence and presence of short-lived activation and fission products. By using the above-mentioned procedures and a low-level liquid scintillator, a lower limit of detection of 10 MBq/sample can be reached for beta nuclides.

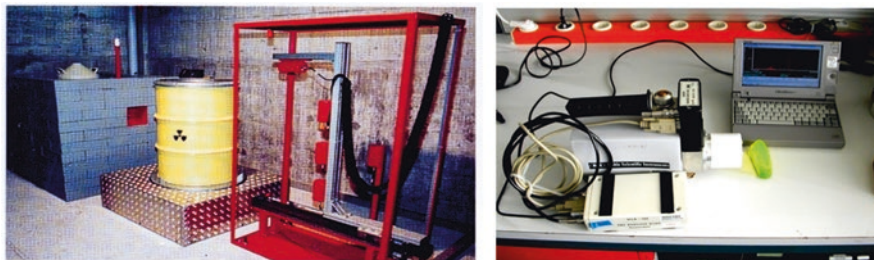


Fig. 16.2 Facilities and equipment used for sample analysis

16.2.3 *Transuranic Environmental Samples Analysis*

The radioactivity of the uranium, thorium, radium isotopes released in the environment from discharges of radiochemistry division at the IANP, or by contamination of NPP accidents was performed by electrodeposition of coprecipitation method with an ammoniac alkaline earth phosphate precipitate, adding a known amount of uran-232 marker solution to a sample 500 ml. The purification of uranium by the ion exchange procedure and electrodeposition on stainless steel disc, when measured by alpha spectrometry, normally gives alpha peaks with a frequency wave measured of about 50 keV. This means that the peaks from the different uranium isotopes are well separated and easily identified and qualified. The radioactivity of the uranium isotopes deposited on the stainless steel disc is measured by counting the alpha particles of uranium isotopes in an alpha spectrometer. The minimum detectable activity for a counting time of 1000 min, is about 10 MBq for each of the uranium isotopes present in the sample (Fig. 16.2).

16.3 Results

In this study are represented some results of analysis performed by our Laboratory teams for environmental samples, in order to probate the validity of our measurements and methodology of measures, and based at the data received by some of aqueous samples was established respectively the table results for the standard samples with known concentration of the ^{137}Cs nuclide, as well as for the environmental samples collected very close with resources that supply the Shkodra Lake (Farkas, 1980; HASL, 1983). As well, we have determined the lower limit of detection of concentration of the epidemiological or environmental samples for alpha, beta and gamma particles (Suomela, 1993a, 1993b). The calibration curve for the standard samples was established, while for the environmental aqueous samples made the mean (average) of the measurement for each of the resource samples and later all the measurement are compared with the average measure of the standard sample. Mean value $\approx 0,23$ Bq is background (Table 16.1).

Table 16.1 The measurements of the ^{137}Cs standard radioactive solutions

| No | Marinelli Beaker | Volume of standard solution | Volume (500 ml) | Measurements (Bq) | Mean value (Bq) |
|----|------------------|-----------------------------|-----------------|-------------------------|-----------------|
| 1 | Blank | 0 ml | 500 | 0,02; 0,75; 0,02; 0,27; | $\approx 0,23$ |
| 2 | Vessel no. 1 | 2,5 ml | 497,2 + 2,5 | 2,03; 9,35; 0,78; 5,77, | $\approx 1,01$ |
| 3 | Vessel no. 2 | 5 ml | 495 + 5 | 0,43; 2, 07; 1,45; 1,15 | $\approx 1,50$ |
| 4 | Vessel no. 3 | 7,5 ml | 492,5 + 7,5 | 4,13; 0,12; 0,02; 0,47; | $\approx 1,65$ |
| 5 | Vessel no. 4 | 10 ml | 490 + 10 | 0,67; 2,68; 0,87; 3,67; | $\approx 1,93$ |

Table 16.2 The measurements of the aqueous samples resources supplier of Shkodra Lake

| Aqueous sample | Resources | Measurements (Bq) | Mean value (Bq) |
|----------------|-----------------|-------------------------------|-----------------|
| Vessel no. 1 | Syri i Vrakes | 0,00; 0,00; 0,00; 1,87; 0,08 | 0,760 (zero) |
| Vessel no. 2 | Syri i Sumajve | 3,60; 6,51; 4,07; 0,00; 0,05 | 3,34 |
| Vessel no. 3 | Syri i Gjonit | 0,00; 0,00; 1,13; 1,52; 2,48 | 1,49 |
| Vessel no. 4 | Syri i Virit | 0,00; 0,00; 0,00; 0,00; 0,00 | 0 (zero) |
| Vessel no. 5 | Syri i Vakes | 0,05; 0,03; 0,02; 0,00; 0,00 | 0,033 (zero) |
| Vessel no. 6 | Syri i Zi | 2,82; 2,55; 5,05; 1,95; 8,25 | 3,80 |
| Vessel no. 7 | Vija e Mullirit | 0,02; 0,025; 0,13; 0,32; 1,72 | 0,27 (zero) |

The results received in Table 16.2, by samples of resources: Syrit te Zi = 3,80 Bq; Syri Sumajve = 3,34 Bq and Syri i Gjonit = 1,49 Bq; shown that concentration of nuclides (gamma total) in the supplying resources of the Shkodra Lake is $\cong 5$ –15 times above the background.

The measurements were carried out by rapid method for determination of activity (concentration) of gamma total nuclides by environmental samples, but underlining: by above mentioned method, was impossible to determine the effects and role of specific nuclides. So, for that reason confirm the necessity to realize the measurements of the aqueous samples resources using other systems and contemporary methods: [a] Instrumental method of the gamma spectrometry analysis; [b] Instrumental method of the liquid scintillates or X-ray fluorescence method; [c] Alpha spectrometry for determination of the uranium, thorium, plutonium and americium nuclides.

Table 16.3 shows the data which given: the collective effective dose takes by patients (calculated and measured by TLD-100 chips; how is used the technetium ^{99m}Tc nuclide, marked by phosphon for bones scintigraphy, and how was evolved the patients numbers during 2010–2015 based at the examinations performed in NML in University Hospital “Mother Theresa” in Tirana.

Article 2 of the national regulation for treatment/management of liquid radioactive waste discharged to the environment (Albanian Government, 1996) are

Table 16.3 The collective effective dose using the ^{99m}Tc nuclide marked with phosphon

| Years | No. Exam (s) bones scintigraphy | Examination number % | Collective effective dose SE = man Sv |
|-------|---------------------------------|----------------------|--|
| 2010 | 247 | 19,5 | 3,22 |
| 2013 | 355 | 27,9 | 3,47 |
| 2015 | 478 | 37,6 | 3,89 |

Table 16.4 Concentration limits & total radioactivity for nuclides at the discharged liquid radioactive waste

| No | Radiotoxicity group | Total activity (Bq/day) | Concentration (Bq/l) |
|----|---------------------|-------------------------|----------------------|
| 1 | Very higher | 4×10^4 | 4×10^3 |
| 2 | Higher | 4×10^5 | 4×10^4 |
| 3 | Medium | 4×10^6 | 4×10^5 |
| 4 | Lower | 4×10^7 | 4×10^6 |

foreseen the level of radioactivity for specific nuclides, which shown in Table 16.4 are in accordance with WAC recommended by IAEA, EC.

16.3.1 Interpretation of Measurements and Content Validity for Risk Assessment

Measurements are one of the principal building blocks of quantitative risk assessment. If measurements are invalid, it is likely that the risk assessments constructed from those measurements will also be invalid. Measurement validity characterizes the extent to which a biomarker of a phenomenon has *content validity*: for instance, pertains to the underlying phenomenon; or *construct validity*: for instance, correlates with other relevant characteristics of the underlying phenomenon; and *criterion validity*: for instance, to predicts some component of the underlying phenomenon. These three components of measurement validity are best assessed in terms of the extent or degree to which they apply to the underlying phenomenon. While, the content validity is the extent to which a marker “incorporates the domain of the phenomenon under study”. For instance, a biomarker of internal dose will have content validity if it reflects the dose contributed by all routes of exposure. A biomarker of effect will have content validity, if it encompasses the essential characteristics of the disease it represents. In other words, the marker must pertain to the appropriate target organ, or its relationship to the natural history of the disease in question must be unambiguous. To properly assess content validity, one must consider the extent to which the marker pertains to the phenomenon (exposure, effect) of interest or, the extent to which the marker represents a relevant feature of that phenomenon. However, it is possible to strengthen determinations of content validity if judgments are made by a group of experts. The focus of such judgments should be the degree to which the marker represents the underlying phenomenon.

16.4 Conclusions

1. Adaptability of the national legal framework and its integration for using of a number of scientific, economic, regulatory and governance challenges need to be addressed if biomarker applications must be incorporated into clinical practices for health innovation.
2. Moreover, much more research studies in these areas are still needed to understand the mechanism of the relationship between exposure by ionizing radiation and health effect. Biomarkers can be used to gain insight into these mechanisms, as well as to describe the empirical associations between exposures by ionizing radiation and results.
3. The framework presented in my research paper may serve as a basis for evaluating the validity of biomarkers for research and for quantitative risk assessments. At present, there are few valid biological markers that can be used to conduct quantitative risk assessments. Before a marker is useful in risk assessment, it should be shown to have content, criterion, and construct validity, and it should be shown to be reliable. More other studies should be performed by Albanian institutions, in order to establish background levels, the range of normal, confounding factors, and optimize collection and analytical techniques.
4. If studies are to be useful in risk assessment, they must be generalized but, more importantly, they must be internally valid. If separate studies are conducted for use in risk assessments, efforts should be made to use similar markers and to pay attention to confounding factors.
5. The results received in our study, by rapid method of the measurements carried out for environmental epidemiological liquid or aqueous samples, and all other measured samples, reflect our first attempt for applying of such method in alpha, beta, gamma, and nuclides determination, taking in consideration factors of risk assessment.
6. The guidance levels that we have in use were taken from IAEA Basic Safety Standards (BSS) for different procedures of safety and security of public and environment control and protection and are adapted in accordance with issues of National Regulations and Code of Practice that exist in Albania already.

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