NUCLEAR FORENSICS – TECHNIQUES FOR CHARACTERIZATION (EXAMPLE OF NUCLEAR TECHNIQUE USE)

Marija Janković, PhD[1]
Jelena Krneta-Nikolić, PhD
Marija Šljivić-Ivanović, PhD
Nataša Sarap, PhD
Filip Veljković, PhD
Bojan Janković, PhD
Vinča Institute of Nuclear Sciences, National Institute of the Republic of Serbia
University of Belgrade, Serbia

INTRODUCTION

Nuclear forensics involves the examination of nuclear and other radioactive materials through various analytical techniques to determine their origin, history, and intended use. Key methodologies include gamma spectrometry, which qualitatively and quantitatively determines the individual radionuclide, and dosimetry, for measurement ambient dose equivalent rate. These techniques are critical in tracing the source of nuclear materials and supporting investigations related to security and non-proliferation. Legislation in the Republic of Serbia defines the methods and procedures for radioactivity control during import, export and transit. Radioactivity control is based on measurements performed by authorized legal persons and involves measurements of dose rate of ionizing radiation from the goods and examination of samples by gamma spectrometric method.

Many samples are too complex for all the radioactive isotopes present to be measured directly. By utilizing the differences in chemical properties of the elements, it is possible to devise schemes of chemical reactions to separate and purify elements, or groups of elements, to allow measurement of the isotopes present by radioactive counting methods, or mass spectrometry [IAEA, 2006]. Radiochemistry is especially important to allow measurement of isotopes that are present at low activity and are best measured by their alpha or beta emissions or by mass spectrometry. Radiochemistry, in combination with radioactive counting techniques and mass spectrometry, represents some of the most effective methods for precise radioactivity determination. These approaches allow for the separation, identification, and quantification of radionuclides in complex matrices.

Nuclear techniques, such as alpha, beta, and gamma spectrometry, enable measurement of radioactive decay emissions, providing information of the radionuclides present. On the other hand, mass spectrometry techniques offer high sensitivity and the ability to detect trace levels of radioactive elements.

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[1] marijam@vinca.rs

DOSIMETRY

There are various types of dosimetry measurements (passive and active, personal dosimetry, dose rate in air etc.). Dosimetry measurements are in wide use in radiation protection, aimed both at continuous control of medical instruments that are using the ionizing radiation sources and exposure of patients and staff operating those instruments [ISO 4037]. Also, dosimetric measurements are readily used to assess the exposure of population to ionizing radiation, as well as a part of nuclear forensic methods, via screening of various spaces and goods that are imported or in transit through the country [Official Gazette of RS, No. 95/18 and 10/19; Official Gazette of RS, No. 86/19]. This is done by measuring the ambient dose rate equivalent, $H^*(10)$, on the surface of the goods, in the transportation vehicle, or inside of the object of interest – ambient monitoring.

According to the legislation of the Republic of Serbia [Official Gazette of RS, No. 86/19], dosimetry measurements are mandatory at the state borders, whenever certain types of goods are imported or in transit. Also, fixed dosimetry portals [Dimović, S. et al. (2023)] are present as a type of early warning system that reacts in the presence of the ionizing radiation of whatever origin.

Dosimeters for ambient dose equivalent rate monitoring used in Radiation and Environmental Protection Department in the Vinča Institute of Nuclear Sciences, are readily exploited in the field work. The Radiation and Environmental Protection Department, Laboratory for the Radiation Measurements is accredited for method of ambient dose rate equivalent measurement according to the ISO 17025/2017 standard and as such, it is able to perform measurement that are both accurate and precise and the results are repeatable and thrust worthy. Most of the instruments used for this purpose are in-house made dosimeters based on compensated Geiger-Muller (GM) tubes as well as several commercially available instruments (such as Ionization chamber and scintillation dosimeter). The stability of the instruments is readily controlled as per requirements of the ISO 17025/2017 standard by measuring the dose of the known standard radioactive source.

In accordance with the Rulebook 86/19, goods that are listed therein have to be controlled by means of the dosimetry measurements. In the moment of the arrival of the goods at the state border, or at the customs point, a dosimetry measurement is performed using a certified instrument. The background ambiental dose rate is measured first, followed by the measurement of the dose rate at the goods and at the place of the transport driver and co-driver, if there is such.

The result of the measurement is then reported in the form of Report on Ambient Dose Rate Equivalent Measurement, which contains, among other relevant data, a conclusion pertaining to the presence of the contamination or source of ionizing radiation. In case of finding the contamination or source of ionizing radiation, the Directorate for the Radiation and Nuclear Safety and Security issues a report which bans the import/transit of the goods. The procedure of measurement and results reporting is defined in the Instructions for Ambient Dose Rate Equivalent Measurement, according to the ISO17025/2017.

Figures 1 – 4 show a typical situation of the dosimetry measurement on the state border.

Figure 1. *Measurement of the Background Dose Rate*

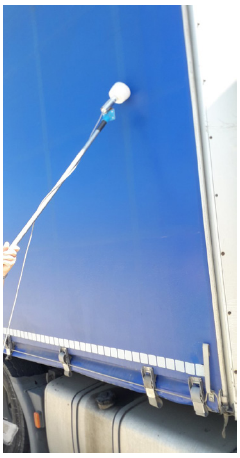


Figure 2. *Measurement of the Ambient Dose Rate Equivalent on the Side of the Truck Transporting the Goods*

Figure 3. *Ambient Dose Rate Equivalent Measurement on the Surface of the Goods*

Figure 4. *Ambient Dose Rate Equivalent Measurement on the Driver's Position*

In case of elevated ambient dose rate equivalent measurement results, further investigation is in order, namely gamma spectrometry, which enables us to determine which radioactive elements are present in the sample of the examined goods and draw more precise conclusions.

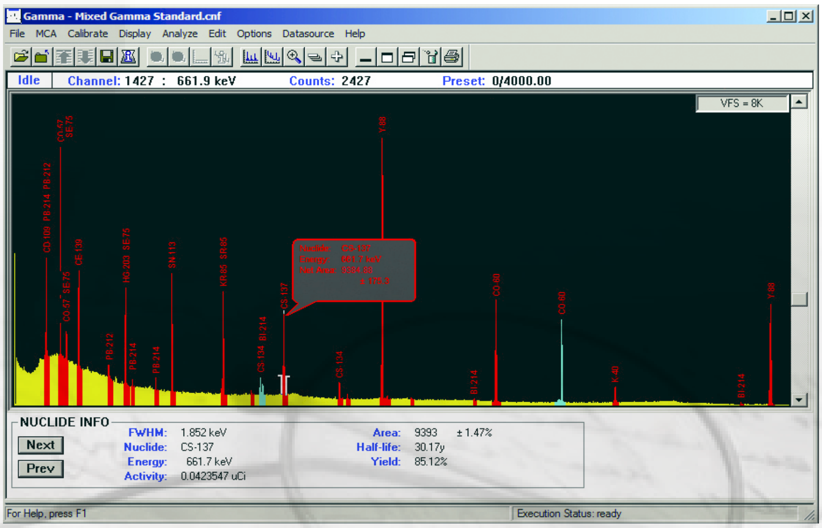
For example, results of ambient dose equivalent rate for goods that require dosimetric examination (waste of aluminum and waste of copper) [Official Gazette of RS, No. 86/19], measured by the Radiation and Environmental Protection Department, Vinča Institute of Nuclear Sciences, at one border crossing in the Republic of Serbia using digital radioactive radiation meter DMRZ-M15 show that the ambient dose equivalent rate were in range 0.057 to 0.070 µSv/h for waste of aluminum and from 0.065 to 0.072 µSv/h for waste of copper. No radioactive sources or contamination were found [Janković, M. et al. (2024)].

GAMMA SPECTROMETRY

Gamma spectrometry is a high resolution, non destructive measurement method which allows the identification and quantification of radionuclides present in the examined sample. It produces rapid results by analyzing the interaction of the gamma photons with the detector material. The samples can be measured in their native form or with minimal preparation which greatly reduces the time needed for the analysis and consumption of chemicals or other resources. Also, this method is convenient for the measurement of low concentrations of radioactive elements and as such, is useful in measurement of environmental samples. It enables us to analyze large number of samples taken in sequence or at particular points and could serve as a tool for determining the origin of the detected radionuclide in the environment from which the sample is taken.

According to the legislation, certain materials undergo mandatory gamma spectrometry measurements. These are various types of natural stones and granites, volcanic stones and ash, various waste products of the mining and metallurgy industry and raw materials that are consequently used in building materials [Janković, M. and Todorović, D. (2011); Janković, M. et al. (2011)]. These papers present the results of concentration of radioactivity content for natural radionuclides: ^{226}Ra, ^{232}Th, ^{40}K, ^{238}U, ^{235}U in coal, slag, and ash samples from coal-fired power plants in Serbia, as well as results of naturally occurred radionuclides in imported zircon samples. All the samples were measured by gamma spectrometry. Also food, feed and water undergoes mandatory control using this technique [Sarap, N. et al (2018)]. Air, soil, surface waters, precipitation and biota samples are readily measured as a part of planned monitoring of the environment in the country [Rajačić, M. et al (2016)]. This is of at most importance, since in the case of any incident or accident involving nuclear facilities, it provides rapid data for the rapid reaction.

Gamma spectrometry produces measurement results in the form of the spectrum. The spectrum consists of all impulses detected by the measurement system, which are a consequence of the interaction of gamma photons with the detector material. As it can be seen in the Figure 5, in the spectrum there are notable peaks which correspond to the detection of gamma photons with certain energy. According to the photon energy present in the spectrum, it is possible to identify which radionuclides are present in the sample. The height of the peaks corresponds to the number of detected photons and the number of the photons corresponds to the activity of the detected radionuclides. That is how we are able to conclude which gamma emitting radioactive elements are present in the investigated sample and in which quantity.



Channel: 1427 : 661.9 keV
Counts: 2427
Preset: 0/4000.00

Nuclide: CS-137
Energy: 661.7 keV
Net Area: 9384.88 ± 175.3

FWHM: 1.852 keV
Nuclide: CS-137
Energy: 661.7 keV
Activity: 0.0423547 uCi
Area: 9393 ± 1.47%
Half-life: 30.17y
Yield: 85.12%

For Help, press F1
Execution Status: ready

Figure 5. *Spectrum of the Commercial Calibration Standard*

For the completion of the gamma spectrometry measurement, certain steps should be performed. Firstly, an energy and efficiency callibration of the detector system should be performed by measuring a calibration source – an object with known presence of radinuclides and their activity. Knowing the radionuclides present in the calibration source enables us to correlate the position of the peaks with the known gamma photon energies. The number of detected photons, read from the spectrum, enables us to calculate a detection efficiency of the detector for known geometry and chemical composition of the source [Debertin, K., Helmer, R.G.,1988].

The next step should be providing a sample that is representative of the bulk material that needs to be measured. This is accomplished by following the sampling procedure in each particular situation [EPA/600/R-12/566]. Then, the sample has to be prepared in such a way that it provides the homegeinity of the material and enables placing into a geometry adequate for the measurement on the gamma detector. After that, the sample is placed on the detector and measured [IAEA, 1989].

The analysis of the spectrum resulting from the measurement gives the energies and number of detected photons and the activity of each radionuclide is calculated as follows:

$$A = \frac{N / t}{P_{\gamma} \varepsilon m} \tag{1}$$

where N is nuber of detected photons with a particular energy, t is measurement duration, P_{γ} is the probability of the emission of photons with a particular energy, ε is the detection efficiency for given energy and m is the mass of the sample.

The final result of the measurement is then given as the activity concentration (expessed in Bq/kg) for each detected radionuclide.

The obtained activities are then compared to the reference values defined in the Rulebooks and the conclusion is made whether the examined material is in accordance to the limits prescribed by the law.

In all materials that are examined according to the Rulebook on the Limits of Radionuclide Content in Drinking Water, Food, Feedstuf, Medicines, Items of General Use, Building Materials and Other Goods that are Put on the Market (Official Gazette of RS, No. 36/18) and Rulebook on Limits of Radioactive Contamination of People, Working and Living Environment and Ways of Performing Decontamination (Official Gazette of RS, No. 38/11), elements that can be present in samples are so called naturally occurring radionuclides and Cs-137 (a consequence of Chernobyl accident and nuclear trials). All other man made radionuclides should not be present in any material other than some specific materials intended for use in nuclear medicine or industry in which case, these materials and items should have a proper documentation [Official Gazette of RS, No. 25/11 and Official Gazette of RS, No. 50/18].

If however, the presence of man-made radionuclides is detected, licenced laboratory that made the measurement has to notify the Directorate on their findings and steps to discover the origin of those radionuclides have to be taken.

Legislative, presented in Rulebook on Radioactivity Control of Goods During the Import, Export and Transit (Official Gazette RS, 86/19), establishes the mandatory gamma spectrometry measurement of berries, while the limits of radionuclide content in these products is defined in the Rulebook on Limits of radionuclide content in drinking water, foodstuff, feedstuff, medicines, items of general use, building materials and other goods placed on the market (Official Gazette RS, 36/18). The Paper [Rajačić, M. et al. (2022)] deals with the examination of ^{137}Cs in imported or exported berries, measured in Radiation and Environmental Protection Department, Vinča Institute of Nuclear Sciences using gamma spectrometry.

GROSS ALPHA /GROSS BETA

Gross alpha and gross beta activity measurement is a screening analysis that is not intended to give an absolute determination of the activity concentration of all alpha and beta emitting radionuclides in a test sample, but to demonstrate whether a particular reference levels have not been exceeded. The method covers non-volatile radionuclides, since some gaseous or volatile radionuclides (e.g. Rn-222 and I-131) can be lost during the sample preparation. It is applicable to test samples of drinking water, rainwater, surface and ground water as well as cooling water, industrial water, domestic and industrial wastewater after proper sampling, sample handling, and test sample preparation [ISO 10704:2019, EPA, 1980]. Also it can be used for samples of aerosol, swipes, sediment and soil as well as samples of food and biota, after an appropriate preparation is performed.

This method is applicable to the measurement of alpha emitters having energies above 3.9 mega electron volts (MeV) and beta emitters having maximum energies above 0.1 MeV.

Given that the range of alpha particles is several cm in air and even less in a medium, the samples have to be prepared in such a way that ensures that after the preparation, all that is left is a thin layer of material that contains all alpha and beta emitters that were present in the sample in its native state. This means the evaporation of liquid samples or taking small aliquots of solid or powdery materials that are homogenized prior to the measurement. For some purposes, when a specific alpha or beta emitter is measured, some chemical preparation should be applied, so that other elements that could potentially interfere with the measurements are eliminated. That is why this method is not non-destructive and at times, can be time consuming [Janković, M. et al. (2015)].

The most used type of detector for gross alpha/gross beta measurements is gas - flow proportional counter. The principal on which it works is the ionisation of the gas present in the detector system, as a consequence of the interaction of alpha or beta particles with the gas. The number of pairs crated in these interactions is proportional to the energy of the particle emitted from the sample and thus the counting of detected particles as well as their discrimination to alpha or beta is enabled.

For absolute gross alpha and gross beta measurement, the detectors must be calibrated to obtain the ratio of count rate to disintegration rate. Americium - 241 (used for alpha activity in the collaborative test of this method) has higher alpha particle energy and Radium - 226 radionuclides but is close to the energy of the alpha particles emitted by naturally occurring radionuclides, trontium-90 and cesium-137 have both been used quite extensively as standards for gross beta activity. Sample weight stability is essential to gross alpha and gross beta measurements to ensure the accuracy of the self-absorption counting efficiency factor to be used for the samples. Strontium-90 in equilibrium with its daughter yttrium-90 is the prescribed radionuclide for gross beta calibrations.

The procedure for the measurement dictates the steps that need to be taken in order to assure valid measurement results. Firstly, the sample needs to be prepared (using evaporation or chemical separation or coprecipitation, or some other method depending on the type of the sample) so that thin layer of the aliquot of sample is obtained and placed in the measurement geometry. Then, the sample is put into the detector and the result of the count is used to calculate gross alpha and gross beta activity of all present radionuclides, following the equation

$$A_{\alpha\beta} = \frac{(R - R_0)}{\varepsilon \cdot m(V)} \tag{2}$$

where $A_{\alpha\beta}$ is the gross alpha or gross beta activity expressed in Bq/kg or Bq/l, R is the count of the sample, R_0 is the count in background, ε is the efficiency of the counting (obtained via calibration procedure) and $m(V)$ is mass or volume of the sample represented by the measured aliquot.

According to the legislation in Serbia, drinking water is subject to the mandatory gross alpha/gross beta measurement. The limits are set to 0.1 Bq/l for gross alpha and 1.0 Bq/l for gross beta activity. These limits provide that the ingestion dose from 730 l of water per year does not exceed 0.1 mSv/y [Official Gazette of RS, No. 36/18]

If the measurement results show values above these limits, an additional measurement aimed to identify the presence of individual radionuclides should be performed, usually by means of gamma spectrometry [Official Gazette of RS, No. 36/18].

The limit set for tritium which is so called soft beta emitter, is 100 Bq/l. Presence of tritium in various types of natural water such as river and lake water, can be used as an indicator of an accidental situation in a nuclear facility [M.Janković et al. (2024)]. Then further investigation should be performed by sampling the water bodies closing to the nuclear facility in order to determine whether there is a possible leakage and to determine, if possible, the precise position where the excess tritium entered a water body.

Planned and regulated monitoring of surface and ground water, precipitation and drinking water establishes the baseline for all other measurements conducted for the purpose of control.

MASS SPECTROMETRY

Mass spectrometry is an analytical technique for examining matter by forming ions in the gas phase that are detected and characterized based on the ratio of mass (m) and charge (z). Thanks to the fact that mass spectrometry provides both qualitative (identification and structure of compounds) and quantitative (molar mass and/or concentration of sample components) information about the investigated substance, it is used today in areas such as biochemistry for the analysis of proteins, peptides, oligonucleotides, lipids, steroids and polysaccharides; physical chemistry for studying the fundamental laws of the gas phase of ions, kinetics of reactions, as well as for determining thermodynamic parameters; inorganic analytical chemistry; atomic physics; pharmacy for the discovery of new drugs, pharmacokinetics and many others; clinical tests such as diagnosing various diseases, drug testing, hemoglobin analysis; geology for the study of isotopes or oil composition; forensics for identification of unknown samples; industry for process flow monitoring etc.

A modern mass spectrometer contains the following components: input for the substance being tested; ionization source, which is necessary to convert the neutrals of the tested substance into ions; one or more mass analyzers for ion separation (in modern instruments, analyzers are also used as fragmentation chambers); detector that registers the ions leaving the last analyzer. In some devices, individual components, such as neutral sources and ionization sources or/and mass analyzers and detectors, can be combined into unique units [De Hoffmann E., Stroobant V. (2007); Van Bramer S. E. (1998)].

Components such as the ion source, mass analyzer and detector in most mass spectrometers are located in a vacuum system, under high voltage, which allows the unhindered movement of ions to the detector without collision with molecules of other gases.

The following processes take place in each mass spectrometer:

- formation of ions from the sample in the ionization source;

- separation of the formed ions based on the ratio of mass and charge in the mass analyzer;
- fragmentation of the obtained ions and analysis of the fragments in the next analyzer;

Ionization Techniques

The ion source is a part of every mass spectrometer and its role is to convert neutral molecules or atoms of the analyte into charged particles, that is, ions. Thus, the ion source is part of the mass spectrometer in which the key process during the analysis, the ionization of the analyte, takes place [Arevalo Jr et al. (2019)]. Ionization is the most important stage in the analysis in mass spectrometry because the success of the analysis depends mostly on the way of converting a neutral compound into an ionic species in the gas phase. The ionization process is carried out by supplying energy to the molecule, resulting in the release or acceptance of electrons by the analyte, which creates a cation radical [M+•] (molecular ion radical) or anion radical [M-•], which represent the molecular mass of the given compound. In addition, the compound can accept or lose a proton, resulting in a protonated [M+H]+ or deprotonated [M-H]- molecular ion. Fragmentation also occurs during the ionization process. Fragmentation is a desirable process during analysis because the appearance of fragments in the spectrum indicates parts of which the molecule is composed. Which fragments will be formed depends primarily on the relative stability of the bonds in the molecular ion and on the stability of the resulting ions. A wide range of ionization techniques has been developed, and the choice of technique depends on the nature of the sample and the desired data. Some of the ionization techniques are [Chhabil Dass, (2007)]: Electron ionization (EI), Fast Atom Bombardment (FAB), Chemical ionization (CI), Atmospheric pressure chemical ionization (APCI), Atmospheric pressure photoionization (APPI), Matrix assisted laser desorption/ionization (MALDI), Electrospray ionization (ESI), Thermospray ionization (TSP), Field ionization/desorption (FDI) and Plasma desorption (PD).

The choice of technique depends on the nature of the sample, i.e. the aggregate state of the sample. Suitable ionization techniques for gas sample ionization are electronic ionization, chemical ionization, and desorption field ionization, therefore their use is limited to nonpolar to moderately polar, volatile, and thermostable analytes. If the sample is in a liquid aggregate state, the analyte is introduced into the ion source in the form of droplets, and this is achieved using thermospray ionization, electrospray ionization or chemical ionization at atmospheric pressure. Analytes analyzed from the liquid phase must be non-volatile, ionic or weakly polar. Solid-state samples where the analyte is polar, non-polar or ionic undergo preparation that involves dissolving the sample in a suitable solvent and mixing with the matrix, and are ionized by techniques such as plasma desorption, matrix-assisted laser desorption/ionization or fast atom bombardment [Gross, J. H. (2017)].

Ionization techniques can be divided depending on the amount of energy that is used, or rather delivered to the molecule. Based on this, “hard” and “soft” ionization techniques are distinguished. Hard techniques, such as electron ionization, are techniques that deliver more energy and lead to intense fragmentation of molecules, and are used for molecules that do not disintegrate easily, providing information about their structure. On the other hand, soft ionization techniques produce mostly ions of molecular species and this group includes chemical ionization, electrospray ionization, chemical ionization under atmospheric pressure, matrix-assisted laser desorption/ionization, fast atom bombardment, plasma desorption [Chhabil Dass, (2007); Gross, J. H. (2017)]. In the following, the most well-known ionization techniques that are applied for various researches and tests are described.

Matrix Assisted Laser Desorption/Ionization

Matrix Assisted Laser Desorption/Ionisation (MALDI) is considered one of the most powerful and sensitive soft ionization techniques [Veljković, F. et al. (2024)]. Michael Karas and Franz Hillenkamp made the biggest contribution in development of this simple technique, which significantly improved the capabilities of mass spectrometry. Thanks to sensitivity, high resolution and compatibility with high-throughput analysis, the technique has found great application for the analysis of non-volatile compounds of high molecular weight such as proteins, nucleic acids, saccharides, synthetic polymers and other organic molecules.

MALDI analysis begins with the preparation of a mixture of sample and matrix, which is deposited on a specially designed MALDI plate. The plate is made of metal and on it there are fields on which the prepared mixture is applied [https://www.creative-proteomics.com/technology/maldi-tof-mass-spectrometry.htm]. The preparation of the sample and matrix mixture is done in one of three possible ways. Some of the methods of preparation are: direct mixing of the sample and the matrix, applying the sample and then the matrix directly to the tile or the “sandwich” method where the matrix-sample-matrix is placed on the tile [Cho et al. (2015)].

The mixture applied to the plate is air-dried, and then the plate is inserted into the ion chamber under vacuum. The analyte is co-crystallized with a solid matrix that can absorb the wavelength of light emitted by the laser. MALDI directly ionizes and vaporizes the analyte from the solid phase, and the ions are produced by a pulsed laser [Glish, G. L. and Vachet, R. W. (2003)]. The laser pulse hits the crystal surface of the mixture and after a very short time the place heats up, the matrix absorbs the laser energy and desorption of analyte molecules from the surface of the field occurs, followed by ionization, i.e. translation of analyte molecules into the gas phase. During this process, analyte molecules are ionized by protonating or deprotonating with nearby matrix molecules [https://www.creative-proteomics.com/technology/maldi-tof-mass-spectrometry.htm]. The formation of individual protonated analytes by this technique is typical, the formation paths are numerous, but the mechanism by which they are formed is not completely clear. Due to the pulsed nature of the laser, ions are formed in discrete events, and MALDI achieves very high levels of sensitivity because very little sample is lost during analysis. Also, only singly charged ions are generated with this technique. The advantage of the technique is high tolerance to salts and buffers, the analyte can be dissolved in both polar and non-polar solvents. The method is fast and reproducible and provides a high level of accuracy in the detection of ionic species.

Lasers, which are the most important source of light, can be of ultraviolet (UV) or infrared (IR) wavelengths. The characteristic of UV lasers is the emission of pulses lasting 3-10 ns, while IR lasers emit 6-200 ns. UV lasers have a greater advantage because short pulses perform a sudden ablation of the sample layer, and thermal degradation of the analyte is avoided for a short period of ion generation.

MALDI matrices used for the desorption and ionization process of the sample are mostly weak organic acids. The correct choice of the matrix is the key to the success of the analysis. The MALDI matrix must efficiently absorb laser radiation, have a low vapor pressure and good stability in vacuum, preferably have a low molar mass in order to evaporate more easily and give a mass spectrum that is easy to interpret, have good solubility in solvents that are at the same time analyte solvents [Gross, J. H. (2017).]

The function of the matrix is to dilute and isolate the analyte molecules from each other, by absorbing the laser radiation, thereby protecting the molecule. The matrix absorbs laser energy, converts it into thermal energy and creates ions from large molecules without their fragmentation.

A TOF (Time Of Flight) analyzer or mass analyzer based on the flight of ions performs separation based on the time it takes for ions of different masses to reach the ion source to the detector. This separation method requires a precise definition of the conditions at the beginning of the ion movement, i.e. at the moment of leaving the ion source. The resulting ions are accelerated, towards the flight tube, by the potential difference between the electrode and the extraction mesh, and in this space all ions receive the same kinetic energy [De Hoffmann, E. and Stroobant, V. (2007); Van Bramer, S. E. (1998)].

The TOF analyzer uses an electric field that accelerates the ions at the same potential. As the mass resolution is proportional to the flight time and the ion path traveled, one of the solutions to increase the resolution of these analyzers is to extend the ion flight tube. However, tubes that are too long reduce its performance due to the loss of ions in collisions with gas molecules that may be found in the tube. The time of flight can be increased by decreasing the ion acceleration voltage, which can lead to a decrease in the sensitivity of the method. This can be avoided by introducing a reflective field, which ensures that ions of the same m/z ratio arrive at the detector at the same time [Mamyrin, B. A. (2001)]. The advantage of the TOF analyzer is that it is one of the fastest mass spectrometric analyzers, has high ion transmission and the highest mass range among mass analyzers, and is most suitable for combining with the MALDI technique [Janković et al. (2023)].

FOURIER TRANSFORM INFRARED (FT-IR) SPECTROSCOPY

The FT-IR has proven to be the valuable experimental tool for the forensic scientists at the macroscopic levels. In considered case, FT-IR represents a quick and non-destructive analysis method for analyzing the samples about 10 microns. This instrumental technique may have many applications for analysis of forensic samples, such as identification of illicit drugs, finger-prints, gun-shot residues, explosives, pharmaceuticals, etc. Recent advances have allowed this technology to be extended further, by using attenuated total reflection (ATR) sub-technique within FTIR spectroscopy. FT-IR is the analysis of infrared light interacting with molecules. It measures the amount of light the chemical bonds of sample absorbs. All molecules have specific frequencies that they vibrate at, caused by the specific energy levels, which are associated with bonds inside the molecule. In the current analysis, infrared radiation is transmitted through a sample. Some of the infrared radiation is absorbed by the sample, and some of infrared radiation is transmitted through the sample. The resulting signal at the detector is a spectrum representing a molecular "fingerprint" of the sample. Like a human fingerprint, no two different molecular structures produce exactly the same infrared spectrum (although some can be very similar). This FT-IR spectrum can be automatically compared to the library of the compounds, for a computer generated most-likely best fits. In that case, the forensic chemist must apply the knowledge of chemicals and chemical interactions, pertinent to the incident in order to definitively identify the compounds found in the spectra. Relying only on the computer generated best fit can leave the compound identification open to identification.

LASER-ABLATION-INDUCTIVELY-COUPLED PLASMA-MASS SPECTROMETRY (LA-ICP-MS)

The LA-ICP-MS is an elemental and isotopic microanalytical technique that is increasingly used for routine analysis in forensic laboratories around the world, due to its great discriminating power between samples and its capacity for analyzing solid evidence of the small sizes. LA-ICP-MS has quasi-non-destructive character that may advantage for samples analyzing in forensic sciencies. The main forensic applications of LA-ICP-MS have been developed for glass and paint samples. In the forensic field, the adjustment of instrumental analytical parameters, the study of interferences produced by fractionation and polyatomic ions, and statistical treatment of data are considered of the great importance. LA-ICP-MS is an effective technique for the direct analysis of solids without requiring their dissolution. LA-ICP-MS is especially recommended to overcome limitations regarding the sample size generally associated with forensic analysis. Its quasi-non-destructive nature also allows that samples already analyzed by LA-ICP-MS to be available for a complementary analysis. In addition, its excellent sensitivity, accuracy and precision, combined with its capacity for isotopic and multi-elemental identification, LA-ICP-MS acts as indispensable experimental technique in the forensics.

SCANNING ELECTRON MICROSCOPY (SEM)

Scanning electron microscopy (SEM) is a powerful imaging technique, where a focused beam of electrons is scanned over the surface of a sample for high-resolution analysis. SEM can offer detailed information regarding the morphology and the elemental composition of the surface. This can prove to be invaluable in the analysis of various types of evidence in forensic science. One of primary applications of SEM in forensic science is trace evidence analysis, a branch of forensic science that involves an examination, the comparison, and interpretation of small and microscopic materials. These materials can include fibers, hairs, glass fragments, paint chips, soil particles, plants, gunshot residue, etc. Trace evidence analysis aims to link these materials to specific sources or individuals, providing valuable information in the criminal investigations. Scanning electron microscopes play a crucial role in examining and characterizing trace evidence by providing key information about the sample including: a) High-resolution images - powerful imaging capability of SEM enables forensic scientists to visualize trace materials at high magnification and resolution. It facilitates a detailed examination of the surface morphology, texture, and structural features of trace evidence. By providing a closer look at the microtopography of fibers, particles, or other materials, SEM assists in their identification and differentiation, b) Elemental composition - Energy-Dispersive X-ray Spectroscopy (EDS) detects and analyzes the X-rays emitted when the sample is bombarded with electrons in the SEM. EDS can be used to perform elemental analysis enabling the identification and quantification of elements present in trace materials. This capability is useful in identifying unknown substances, such as drugs, explosives, or chemical residues, which can provide critical insights into the nature of a crime or aid in the identification of illicit substances. SEMs can analyze samples such as hair and fibers and paints, as well as particulates, entomological materials and gunshot residue. Namely, the gunshot residue analysis is specialized branch of the forensic science, which focuses on the trace evidence left by the suspect shooters. Discharged firearms release microscopic particles in the cloud of smoke which then deposit on surrounding surfaces, and the hands which pulled the trigger. The telltale signs that indicate when a firearm has been used in a crime, consist of particles with lead (Pb), barium (Ba), and antimony (Sb) traces, though some the lead-free primers, are rising in the abundance and becoming more relevant in investigations. Many components of weapon, such as primer ingredients, bullets, barrel, casing, and propellant (gunpowder), influence the elemental concentration of Pb, Sb, and Ba in particles.

X-Ray Diffraction (XRD) Analysis

The decisive advantage of X-ray diffraction (XRD) method in the forensic science is based on the unique character of the diffraction patterns of crystalline substances, the ability to distinguish between elements and their oxides, and possibility to identify chemical compounds, polymorphic forms, and mixed crystals by a non-destructive examination. This particular quality of X-ray diffraction examinations in the forensic science was verified by comparison of diffraction patterns from iron, from iron oxides, and from iron oxide hydroxide [Janković, B. et al. (2024)]. These substances are used as pigments in paints. In comparison with other methods, X-ray diffraction examinations in these cases provide additional information about the chemical and physical properties, and make it possible for further useful discrimination of materials. The type, the amount, and consistency of the suspected contact trace specimen, the involved contact trace carrier, and forensic questions raised by the criminal offense determine the diffraction method used, strategies of measurement applied, and the sample preparation technique selected. XRD as non-destructive technique requires minimum sample preparation prior to an analysis. The only requirement is that the sample should be homogeneous in nature, so as to provide uniform analysis results even if a small portion is analysed from the bulk quantity. Samples commonly received for forensic testing commonly include: a) building materials such as cement, concrete, steel rods, bricks, etc., b) drugs of abuse and other banned substances, c) residues from site of arson such as kerosene oil, gasoline or cotton lints, d) the explosive residues and splinters from sites, e) the assault samples such as torn garments, broken glass, cosmetic marks, etc., and f) theft and robbery site samples which include gunshot residues, tools, murder weapons, forged documents, blood residues, etc. Samples can be analyzed by the XRD if it exhibits a degree of crystallinity, even if the remaining content is the amorphous. The technique helps to establish the presence or absence of the particular material, through comparison against the reference of XRD data base.

X-Ray Fluorescence (XRF) Microscopy

The X-ray fluorescence (XRF) is a very useful and versatile tool to forensic scientists. It is a non-destructive technique, which is particularly advantageous if the sample is limited or further analysis is needed. Not only is it a non-destructive technique, it requires essentially no sample preparation. The first step in the analysis of the sample is the production of an X-ray, formed in a source which contains a wire (often tungsten) and an anode. A cathode releases electrons which accelerate towards the anode where they impact, resulting in to emission of an X-ray. When X-rays strike the material under the analysis, they can either be absorbed by atoms or scattered. If absorbed, the X-ray transfers all of its energy to the innermost electron of an atom. This is known as the photoelectric effect. Provided the X-ray has adequate energy, electrons are then ejected from the inner shells of the atom, thus creating vacancies in the electron orbitals. In order for the atom to return to an electronically stable state, outer electrons transfer to the inner shells, releasing X-rays in the process at wavelengths specific to the atoms present. This is known as X-ray fluorescence. These X-rays are characteristic to the particular element, and it is this feature that allows for elemental composition to be established. However as the atom is returning to its original stable state, it may sometimes transfer the excitation energy to one of the outer electrons, causing the electron to be ejected. This is referred to as an Auger electron, and can cause problems in detection in terms of competition. The emitted X-rays are then detected. XRF instruments are typically one of two types: Energy-dispersive (ED) or Wavelength-dispersive (WD). The EDXRF systems use the semi-conductor-like detectors which, having an received the emitted spectrum from the sample, transfer it into the histogram of numbers of counts against photon energy.

X-ray fluorescence is a particularly beneficial tool to forensic scientists, for a number of reasons: a) it is non-destructive technique and the sample is left intact after analysis, and b) particularly advantageous if the amount of sample is limited or further analysis is required. Naturally in the forensic science there may be only a limited amount of sample recovered. Compared to certain other analytical techniques it is fast, requiring very little sample preparation as material can often be analysed as is (perhaps requiring homogenisation first) and producing results in minutes, thus multiple analyses can be carried out in a shorter space of time.

The XRF instrument has a wide range of potential applications in the forensic science. It is particularly beneficial in the analysis of rocks, soils and other similar substances, allowing the analyst to compare the composition of these samples. This could be used in suggesting whether two similar-appearing soils are likely to have originated from the same source, or if they are different, based on the various elements detected. The inks and paints are particularly suitable for analysis by XRF technique, as they typically contain metallic pigments which can differ wildly between different brands and batches. The forensic analysis of writing and printer inks can be of particular importance in the investigation of questioned documents, for instance in aiming to establish whether two documents may have been written using the same ink. Furthermore, the identification of gunshot residues can also be achieved using the XRF technique. When a gun is discharged, minute particles from the firearm, bullet and gunpowder are propelled from the weapon, often onto the hands and clothing of the shooter. As the composition of gunshot residue is reasonably well documented and composed of compounds that are not typically present on the person’s hand, the XRF can be applied to a piece of clothing or even an individual’s body to ascertain, if such residues might be present, an indicating whether or not they could have fired a weapon recently.

CONCLUSION

Forensic examinations, and among them nuclear forensics as a special branch, are very specific examinations that can provide key information in particular investigations. Using isotopic, chemical and physical characteristics of nuclear and other material, together with related forensic evidence to include DNA, hair, fingerprints, tool marks and explosive residues, nuclear forensics can potentially link samples of interest to people, places and events. Nuclear forensics has a number of specific techniques and methods at its disposal, among which, dosimetry, gamma spectrometry, alpha and beta radiation measurements and mass spectrometry are one of the most prominent and useful. Dosimetry and gamma spectrometry are useful in case of some important considerations when it is necessary to collect and preserve evidence and properly sequence non-destructive ahead of destructive analysis in the laboratory. While dosimetry provides the most rapid information, gamma spectrometry has the advantage of providing radionuclide content information as well as activity concentration in the measured samples. Gross alpha/gross beta measurements are used for rapid screening, much as the dosimetry, but for alpha and beta emitting radionuclides that are of the special interest and often cannot be detected using dosimetry alone. For this type of measurement, the sample has to be properly prepared and therefore destroyed, but at the same time, gross alpha/gross beta measurements can be followed by gamma spectrometry if needed. Mass spectrometry in its variety of techniques, provides most precise and detailed results but the sample preparation is complicated, obtaining the results is time consuming and it is not applicable for field measurements.

All the methods available for the nuclear forensics can and should be used in combination, in proper sequence and in planned manner. In that case, the information gathered is both rapid and precise and can provide an invaluable insight in these specific investigations.

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