SIGNIFICANCE OF USING SCANNING ELECTRON MICROSCOPY WITH ENERGY DISPERSIVE X-RAY SPECTROMETRY (SEM-EDX) FOR ANALYSIS OF EVIDENCE MATERIAL IN FORENSIC SCIENCES

Abstract

Forensic applications of Scanning Electron Microscopy (SEM) are found mostly in areas where there is a need for good imaging at relatively high magnifications.SEM enables the forensic scientist to examine specimens at much higher magnification than those possible with optical microscopy and without the difficulties of specimen preparation associated with conventional electron microscope. Physicochemical examinations of gunshot residues, called also chemical ballistics, are helpful, e.g. in identification of damages and injuries as the effect of the use of firearms (with indicating the entrance and exit of projectile), estimation of the shooting distance and also establishing, whether a person has used a firearm.It is discussed in details that Scanning Electron Microscopy with Energy Dispersive X-Ray Spectroscopy (SEM-EDX) can serve as a powerful tool for forensic scientists to classify and discriminate evidence material because they can simultaneously examine the morphology and the elemental composition of objects.

Keywords: gunshot residues, elemental analysis, Scanning Electron Microscopy, forensic science

1. Introduction

"The electron microscope uses electron beams to generate an image of the sample, unlike the compound light microscope, which uses light to image the sample. Two important parameters of any microscope are the resolution and magnification. The electron microscope has a much greater resolution capability than the light microscope because the wavelength of the electrons is about 10^5 times smaller than the wavelength of light. The best resolution with the light microscope is around 200 nm whereas with electron microscope it is less than 1 nm. Thus, the resolution with electron microscope is ~100 times higher than the light microscope. Similarly, the magnification of electron microscope is almost 300 times more than the optical microscope".(nanoscience.com, 2024)

"Scanning Electron Microscopy (SEM) has been an important forensic tool since the 1970s"(Pilkington, 2022). "SEM enables the forensic scientist to examine specimens at much higher magnification than those possible with optical microscopy and without the difficulties of specimen preparation associated with conventional electron microscopy" (Taylor, 1973). "One of the most striking properties of SEM is its ability to combine imaging with the elemental analysis

together with its suitability for digitization and automation of complete tasks. SEM can also be a useful tool for solving the forgeries, and to interpret the damage of textile materials"(Vermeijetal, 2009).

"Forensic applications of SEM are found mostly in areas where there is a need for good imaging at relatively high magnifications in combination with elemental analysis" (Yashoda etal, 2021)."The concept of trace evidence in forensic science originates from Locard's exchange principle, which states that "every contact leaves a trace." The trace evidence is typically in the form of particles of skin, hair, fibres, clothing, soil, paint, and glass, among other materials" (Mummery, 2021). "Fragments of various materials such as glass, paints, fibres and gunshot residues (GSR) are frequently present at the scene of such events as car accidents, burglaries, fights or crimes committed with the use of fire arms. These materials can be recovered in trace amount from the hair, hands, clothing and shoes of the victims and witnesses in a crime scene and can be provided as an evidence at a court" (Zadora &Brożek-Mucha, 2003).These trace materials are of relatively heterogenic character and with a complex composition. Thus, sensitive analytical methods like SEM are required in order to obtain satisfactory results from small amounts of sample.

"When SEM is combined with energy dispersive X-ray spectrometry it is known as SEM–EDX.It is a powerful tool for forensic scientists to classify and discriminate evidence material because they can simultaneously examine the morphology and the elemental composition of objects. Moreover, the obtained results could be enhanced using some methods of chemometric analysislike Locally weighted regression (LWR), Multiple linear regression (MLR), Neural networks (NNs), Artificial neural networks (ANN), Partial least squares (PLS), Principal component regression (PCR)"(Zadora &Brożek-Mucha, 2003)(Biancolillo& Marini, 2018)

2. Experimental details of SEM-EDX

SEMis a technique that produces significantlymagnified images by using electrons beams. A schematic diagram of the SEM is shown in figure 1. "Firstly, a beam of electrons is produced at the top of the microscope by an electron gun mounted in an enclosure and maintained at high vacuum. The electron beam follows a vertical path through the microscope. The beam travels through electromagnetic fields and lenses, which focus the beam towards the sample. There are three main signals that occur when an electron beam meets the sample (figure 1). Secondary electrons, backscattered electrons, and X-rays. These signals are produced from interactions between electrons and the sample material, and each signal provide different information about the sample. Secondary electrons emanate from atoms on the surface of the sample material and recording them enables SEMs to output detailed, topographic imagery with high spatial resolution. Backscattered electrons are refracted incident electrons that have penetrated below the sample surface and interacted with atoms inside the material. Atoms of more massive elements in the sample bounce incident electrons further away, and these scatter patterns can reveal information about the sample's internal elemental makeup" (Pilkington, 2022).

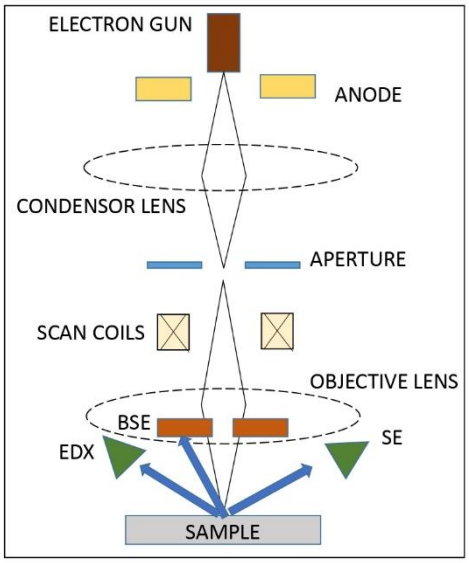

Figure 1:*Schematic diagram of SEM-EDX. A beam of electrons is produced at the top of the microscope by an electron gun mounted in an enclosure and maintained at high vacuum. The beam travels through electromagnetic fields and lenses, which focus the beam towards the sample. All electromagnetic lens and detector are maintained at high vacuum. There are three main signals that occur when an electron beam meets the sample.*

BSE: Back Scattered Electrons
SE: Scattered Electrons

"X-rays are produced when electrons in the electron beam replace electrons from atoms in the sample. Designated detectors mounted in vacuum collect these X-rays, backscattered electrons, and secondary electrons and convert them into a signal that is sent to a screen. This produces the final image"(Han etal, 2018)."The wavelengths of X-rays produced in SEM are related to the elements that electrons have interacted with. Energy-dispersive spectrometry (EDX) is combined with SEM in SEM-EDX to record this information. EDX can be used for both qualitative and quantitative analysis, enabling users to identify both the type of elements that are present as well as the percentage of each element's concentration within the sample"(Scimeca etal, 2018)

"The first scanning electron microscope with very high resolution came in 1937 by Manfred von Ardennes" (Von Ardenneetal, 1921).It is worth mentioning that SEMs can achieve magnification higher than 100,000x with a resolution below 1 nm. The samples need to be either conductive themselves or be coated with a conductive material such as gold-plated nanoparticles before inserting it in SEM. SEM-EDX is considered a relatively rapid and non-destructive approach to surface analysis. So, the sample may be reused for other analyses. This is particularly helpful in the case of forensic investigation as often the sample obtained is of micro or nanoscale range.

3. Result and discussion: SEM-EDX in forensic analysis

A) Gunshot Residue:

In 1968, research for detection of Gun Shot Residue (GSR) elements was done using SEM/EDX first carried out in England (Ward, 1982). GSR, which is also sometimes called Firearm Discharge Residue, can be collected from various surfaces such as parts of the human body, clothing, parts of an automobile, or the vicinity of the suspected gun discharge. The efficiency of collecting the samples will greatly affect the detection and analysis. A simple tape lift-off method is the most common method for collecting samples for SEM analysis and is used to collect GSR from various surfaces. A SEM aluminium stub with a carbon adhesive is used to collect the samples (nanoscience.com).

"Anywhere between 0 and 1,000 particles, or even in excess of 10,000 particles, can be found on a GSR sample. It depends largely on the circumstances, such as which gun has been used, which ammunition, and which location (whether outdoors or indoors)"(azom.com). While suspects are the main source of samples, it is still common practice to take samples from victims. In instances of potential suicide, it is possible to gather evidence of whether or not the victim has used the firearm in order to take their own life. Significantly, more GSR particles will be present on the victim as compared to cases where another individual has fired the gun. It is hard to determine exactly what a positive GSR outcome means as it is generally accepted that GSR particles do not only come from firearms. They may, for instance, be picked up via secondary processes. Shaking the hand of a person who has shot a firearm recently may deposit GSR particles on a person's hand.

The presence of GSR alone is not sufficient – it is always presented as supplementary evidence. For instance, a person is killed via gun-shot and during investigation, the investigators locate GSR-particles on a neighbour's hand, then the neighbour can be called in for questioning, but merely finding gunshot residue in the hand cannot be conclusive evidence for convicting a person (azom.com). French et. al, (2013) have reported that "while varying between runs, over 100 particles were transferred via a handshake in one instance, and it was found that even very large particles (60–100+ µm) were transferred from the shooter to the second individual via a handshake. The findings have implications for forensic investigations, including highlighting the need to sample from individuals who might have been involved in transfers and underscoring the importance of achieving accurate particle counts using the SEM-EDX method. Most importantly, the findings suggest that the presence of GSR (especially in small quantities) may not always indicate that a person discharged a firearm and that the possibility for misidentification of the shooter exists, as does the potential to distinguish shooters from those who have acquired GSR through secondary transfer".

"The gunshot residue is mainly of two types, Organic and Inorganic. Organic residues originate from the propellant: unburned and partially burned gun powder particles, some products of their transformation and also particles of lubricants. Inorganic residues, mostly metallic, originate from the primer as well as from metallic parts of cartridge and the weapon itself" (Gunaratnam, Himberg 1994). "Among them only particles originating from the primer reveal specific chemical

content and characteristic morphology. Their morphology reflects the kinetics of the processes undergoing during a gunshot, especially rapid cooling of droplets of the existing molten metals in the expanding plum of products during the primer detonation and the propellant combustion. The analysis of gunshot residue of inorganic nature, is always preferable using scanning electron microscopy coupled with the X-ray microanalysis (SEM/EDX)" (Brozek-Mucha, 2014; Taudte et al., 2014), because "it is nondestructive and allows the chemical and morphological identification of mineral particles" (DeGaetano & Siegel, 1990; Romolo & Margot, 2001).

Neutron Activation Analysis (NAA) assess with high sensitivity the traces of antimony (Sb), Barium (Ba), Bromine (Br) and iron (Fe) on the graphite sampling support. But NAA requires a relatively large sample size and a nuclear reactor. So, NAA may not be suitable for GSR analysis, as most of the times, the sample size available for analysis is in trace amounts. In addition, thetechnique is unable to decide whether these elements come from a GSR or another source of pollution.

"Atomic absorbance spectroscopy (AAS) detects Pb, Ba, and Sb in trace samples but is an expensive and destructive technique. Inductively coupled plasma (ICP) combined with mass spectrometry (MS) can rapidly detect various elements, but is also a destructive technique. All these techniques are slow, lack spatial specificity, and require highly trained personnel to run the instruments and interpret the results" (Hellmissetal, 1987).

"Total Reflection X-Ray Fluorescence Analysis (TXRF)is an Energy Dispersive XRay Fluorescence (EDXRF) technique, utilizing the total external reflection of X-rays on the smooth plane surface of a reflector material, e.g. polished quartz. TXRF is used for ultra-trace analysis of residues, particles, and impurities on smooth surfaces. It's a powerful analytical tool for micro and trace multi-elemental analysis and can detect a wide range of elements simultaneously"(Strelietal, 2017). "TXRF is highly sensitive and can detect elements at ppb levels. However, XRF can be affected by several factors that can introduce errors or uncertainties in the results. These factors include: matrix effects, interferences, background noise, calibration standards, and instrument performance"(Badla et al, 2020). "Muratsu et al used Synchrotron Radiation Total Reflection X-Ray Fluorescence Analysis (SR-TXRF) for detection of trace elements in drugs of abuse like methamphetamine, amphetamine, 3,4-methylenedioxymethamphetamine, cocaine, and heroin" (Muratsuetal, 2002).

"The non-destructive nature of the SEM-EDX permits the retention of evidence, allowing for re-examination if necessary, making it a reliable and invaluable tool for forensic firearm investigations. The requirement for highly trained personnel has also been overcome with the introduction of automated SEMs" (nanoscience.com). Many automated SEM are compact, have a great speed, thus saving floor space and time. The automated SEM comes with an automated particle analysis software built onto the platform, offering a dedicated GSR analysis solution that is compliant with instrument. The workflow of a typical GSR run involves defining a scan area for

each sample stub, scanning the scan area frame by frame, using the backscattered electron detector to detect particles, and determining the elemental composition of each particle. The automated SEM also comes with various user-friendly features like customize and save workflows, Auto Video function to optimize brightness and contrast at any point throughout the GSR run and Dual Thresholding feature (azom.com).

"The revelation of ballistic gunshot residue (GSR) is commonly performed with the aim of determining whether or not a suspect handled a firearm and or to estimate a firing distance" (Gallusseretal, 2002; Kersh etal, 2014). "Most frequently, gunshot residues are collected from the suspect's hands, where the concentration of these traces is maximal immediately after shooting. In case more time has elapsed since the event traces should be collected rather from the hair, face and clothes where they usually remain for longer than on hands. It has been established from empirical studies that only three-component particles containing lead, antimony and barium are unique primer residues" (Basu, 1982; Woltenetal, 1980). "But according to Gunshot Residue Subcommittee of Chemistry Scientific Area Committee in the organization of Scientific Area Committees (OSAC) for Forensic Science classifies Lead, barium, calcium, silicon combination as well as the combinations of Gadolinium, titanium, zinc; Gallium, copper, tin; Titanium, zinc and Strontium as Gunshot Residue. Particles containing iron, chromium, nickel, copper, zinc and other elements are typical for the case, projectile and its jacket as well as the barrel"(OSAC guidelines)."However, they can also originate from subjects of everyday use or pollution and thus are considered as false positives and cannot be considered as an evidence of firearm shooting" (Mosher etal, 1998; Romolo & Margot, 2001, Chohra, 2015).

"Physicochemical examinations of gunshot residues, called also chemical ballistics, are helpful, e.g. in identification of damages and injuries as the effect of the use of firearms (with indicating the entrance and exit of projectile), estimation of the shooting distance and also establishing, whether a person has used a firearm. Only few particles, of total mass no greater than 100 pg, can be accepted as the evidence relating an individual with a shooting incident. Fulfilling these tasks is very helpful for the reconstruction of an investigated crime. However, new challenges for gunshot residues examiners arise. With a growing frequency the administration of justice asks about the type of ammunition, and so the firearm used in cases when the only accessible for examinations evidence are gunshot residues. Thus, one more task can be formulated, i.e., the identification of an ammunition from the gunshot residues detected.Johnson et al reported the procedures of using morphological and elemental indicators for differentiation between ammunition as well as firearms used to discharge a round, case and bullet".(Lebiedzik& Johnson, 2000).

In an investigation byBrożek-Mucha Z. (2014), it was reported that "the distribution of particles in the surroundings of the shooting gun is not uniform. The number of particles, their chemical composition, and their morphological features depend on the distance from the muzzle of the shooting gun and the type of the substrate the particles sediment on. Results of this study gave

rise to working out the method of shooting distance estimation from the physical and chemical examinations of GSR pattern around the gunshot wounds and damages—extending the possibilities of shooting distance estimation with range of about 50–100 cm, in addition to the three categories commonly used until now: (i) contact or a nearest vicinity shot (about 0-1 cm), (ii) close distance shot (about 1–50 cm), and (iii) distant shot".

Collins et.al, (2003)performed for the analysis of gunshot residue along with glass fragments from the hands of shooters using SEM/EDX and according to him, SEM-EDX could easily discriminate the glass fragments from the inorganic GSR particles (fusion of Pb and Ba).Hellmiss et.al,in 1987 equipped an SEM instrument with Auger electron spectroscopy for the analysis of gunshot residue instead of using EDX.

B) Car Paint Residue:

Vehicular accidents and crimes involving automobiles are increased nowadays. Paint smears or chips are found on the crime scene as trace evidences leading to establishing the identity of vehicle involved in the crime by analysing with control sample of suspected vehicle. Investigation of such cases are influenced by many factors, such as information from CCTV footages, eye witness andother possible physical evidences.

According to the Forensic Paint Analysis and Comparison Guidelines by Scientific Working Group on Materials Analysis (SWGMAT) Forensic paint analyses and comparisons are typically distinguished by sample size that precludes the application of many standard industrial paint analysis procedures or protocols. The guidelines instruct that when paint evidence is recognized, every effort should be made to manually remove it before using tape lifts to collect other types of evidence. If paint is collected with tape lifts, the collector should be aware of the possible difficulty encountered when attempting to manipulate paint samples bearing adhesive residues." In addition, components of the adhesive could contaminate the paint sample and change its apparent chemistry. When contact between two coated surfaces is indicated, the possibility of cross transfers must be considered. Therefore, if available, samples from both surfaces should be collected. Paint flakes can be removed from the parent surface by a number of methods. These include but are not limited to the following: lifting or prying loosely attached flakes, cutting samples of the entire paint layer structure using a clean knife or blade, or dislodging by gently impacting the opposite side of the painted surface. When cutting, it is important that the blade mustbe inserted down to the parent surface. It should be noted that only one method of sampling should be relied upon exclusively" (SWGMAT, 2000).

"For the forensic analysis of multi-layered paint chips of hit-and-run cars, detailed compositional analysis, including minor/trace chemical components in the multi-layered paint chips, is crucial for the potential credentials of the run-away car. The number of layers, painting process, and used paints are quite specific to the types of cars, colour of cars, and their surface protection

depending on the car manufacturer and the year of manufacture, and yet overall characteristics of some paints used by car manufacturers might be quite similar" (Malek etal, 2019). "The composition includes one or more under coats, topcoat and clear coat on the surface and each layer have organic pigments, additive and binder. Automotive paints generally consist of three or four layers and may vary from manufacturer company. The paint coat of a car body consists of a number of successively overlaid paint layers. These layers differ from each other in terms of their ingredients, i.e., resin, pigments and fillers. The number of layers making up a car covering depends on its type. In brand new cars and in those that have not been repainted there are only three to four layers. The components present in the various layers can be identical and thus, individualization of vehicle by its automotive paint is the prime information for investigators for discriminating. Characterizing and individualizing of automotive paints using instrumental analysis enabled breakthrough in scientific investigation" (Lavine etal, 2015).

Paint coverings of renovated cars consist of a larger number of layers (sometimes even more than a dozen), including not only enamels, but also putties, painters' putties and ground undercoats. In identification and comparative studies of paint chips, scientists define their macroscopic properties—colour, shade and texture—and their microscopic properties relating to their morphology (the number and sequence of layers, their thickness and colour). The next stage is a detailed analysis of the chemical content of each layer, including identification of the binder, pigments and fillers. SEM–EDX can be very useful in the case, when the compared paint samples are similar in the microscopic properties relating to their morphology (the number and sequence of layers, their thickness, colour) and the results of previous infra-red spectroscopy analysis (like Fourier Transform Infrared Spectroscopy), if any. Thus, the identification of a particular paint layer can be carried out comparing the contents of the elements, since they are characteristic for a given layer and the combination of elements does not get repeated for other layers.

In a study done by Kaur etal (2022), examination has been done on the layers of automotive paint chips collected from Maruti Suzuki from Kottayam region of Kerala using SEM-EDX. The result shows the presence of Thallium in white and grey paint samples and Aluminium in the red paint samples of automobile paint. Thus, profiling chemicals and elements in the sample may lead to individualization of the automobiles and can be include in the paint database.

In a study done by Malek etal, SEM/EDX were applied in combination with ATR-FTIR imaging and RMS for a detailed characterization of three samples of car paint chips. The molded and polished cross-sections of the car paint chips maintaining their layered structures were specially prepared for the multi-modal analysis. Unambiguous molecular speciation of the chemical components within the layers as well as a determination of the physical layered structures were possible using this multi-modal approach. Although elemental composition information from SEM/EDX analysis is insufficient for molecular speciation, the detection of chemical elements in the layers is consistent with and supportive of the ATR-FTIR and Raman data for polymer resins, inorganics, and pigments. Five types of polymer resins, such as alkyd, alkyd-melamine, acrylic,

epoxy, and polybutadiene resins, were clearly distinguished along with TiO_2, SnO_2, FeS_x, Fe_3O_4, $CuCl_2$, ZnO, and Al_2O_3 as pigments, and kaolinite, talc, pyrophyllite, $BaSO_4$, $Al_2(SO_4)_3$, $Zn_3(PO_4)_2$, and Al flakes as fillers. This study provides detailed information on the chemical identities of every layer of three car paint chips. This is useful for tracing the origin of the car manufacturer when compared to the coating history database of the respective company, which clearly provides the potential credentials of cars involved in hit-and-run accidents. (Maleketal, 2019). Infrared microspectrometry and Raman microscopy were applied in characterisation of paint coatings, i.e.in identification of pigments and in differentiation between paint samples of similar colour and shade, in a complementary way. Some of inorganic pigments and fillers like titanium dioxide or chromates are visible on infrared spectra of the paint samples while organic pigments and dyes can be identified only on the base of Raman spectra. Raman spectrometry in many cases, enables differentiation between paints of similar polymer binder, colour and shade. Because no sample preparation is required, the method provides an excellent means of rapidly screening reference panels for the presence of certain pigments. Raman imaging techniques enable us to determine, how chemical composition varies at the surface of, and within, samples (Zieba-palus et al, 2011). Micro-Raman spectroscopy, like micro-FTIR spectroscopy, can be used for characterization as far as they provide information about characteristic vibrational levels. Nevertheless, not all transitions between vibrational molecular levels are allowed. Some transitions can appear only in the infrared spectrum, some only in the Raman one, and some in both of them at coincidental frequencies; others cannot be observed in either of the spectra. The IR- or Raman-allowed or forbidden transitions are determined by the selection rules. Thus, characterization by micro-FTIR and micro-Raman presents difficulties with some pigments as they are not able to give complete chracterization. In these cases, analysis by EDX solves most of these doubts. The combined use of both spectroscopic techniques, together with SEM–EDX microanalysis, provides one of the most useful methods in the characterization process (Franquelo et al, 2009).

C) Glass Residue:

Glass is an amorphous translucent or transparent material that is made up of a mixture of silicates. It is made by fusion of molten silicate and then solidification without crystallization. Most commercially produced glasses utilize sand (SiO_2), and normally other oxides like CaO, Na_2O, and K_2O are added, which help reduce the viscosity and melting point of SiO_2 (SWGMAT, 2004)

Other materials are also added depending upon the required properties of the glass; for example, PbO increases refractivity, B_2O_3reduces thermal expansion, Al_2O_3increases durability etc. Therefore, there are various elemental components in glass that help determine properties.Elementary analysis of glass using SEM determines the ratio of concentrations of Na/Mg, Ca/K, Mg/Al, Ca/Na, and Na/Al. Through these ratios, the glass is categorized, and properties are determined (Khan, 2022)

Glass is most commonly associated as evidence in property offenses (for example, break-in into properties by thieves and robbers), it may be encountered in almost every type of criminal investigation. In fact, glass fragments have been reported as one of the most frequently encountered physical evidence at crime scenes (Parker and Peterson, 1972).

The large frequency of occurrence of glass as forensic evidence is attributable to many factors. Glass is ubiquitous, produced in large quantities and for a wide variety of applications, and due to the way in which it fractures, small particles of glass are common to the environment. These tiny glass fragments, either generated during the commission of a crime or pre-existing at the scene, can be carried away from the crime scene unwillingly by the perpetrator by adhering to clothing or shoes. Further, glass is chemically stable and resistant to environmental factors and therefore can be analyzed by the forensic laboratory with meaningful results even after a considerable amount of time has lapsed between the occurrence of the crime and the collection of questioned glass samples associated with the suspect(Buscaglia, 1994).

Glass as the evidence material often occurs in very small quantities. Thus, investigations of glass samples require sensitive analytical methods providing satisfactory results from small amounts of the examined material such us the quantitative elemental analysis using SEM–EDX method. The elemental composition of glass strongly depends on the properties of glass products.

Almiralletal (2012) performed elemental analysis of Glass residues with a number of methods like Micro X-Ray Fluorescence (μXRF), Laser Induced Breakdown Spectroscopy (LIBS) and Laser Ablation Inductively Coupled Plasma Mass Spectrometry (LA-ICP-MS)and they observed that SEM-EDX should only be used either for classification between glass types or for the exclusion of an association when the glass samples have an obvious compositional difference. This is due to the extremely limited utility of SEM-EDX in differentiating between different glass samples as a result of the poor sensitivity (Limit of Detection LOD of ~ 1000 ppm) and the fact that SEM-EDXsuffers from differences in analytical results depending on sample morphology (flat vs irregular surface).Thus, the researchers have used this technique as a mere comparison tool in glass analysis.

In most published studies, the element intensity ratios obtained from SEM-EDX measurements are used for classification of glass types (Ryland 1986; Terry et al. 1982). An analytical scheme that combines measurement of Ca/Mg intensity ratios obtained using SEM-EDX with Ca/Fe ratios obtained using X-ray fluorescence spectrometry has been used with good success by several forensic laboratories to classify glass fragments into sheet and container categories (Keeley and Christofides 1979; Ryland 1986).

For discrimination among glass sources, a SEM-EDXprotocol was reported for determining the ratios of the intensities of Na/Mg, Na/Al, Mg/Al, Ca/Na, and Ca/K in glass fragments (Andrasko and Machly, 1978). Measurement of these ratios by SEM-EDX was incorporated into a scheme

with refractive index, density, and emission spectrograph. Thirty-eight out of 40 window glasses analysed by this scheme were found to be distinguishable. The variation in the measured element intensity ratios bySEM-EDX was found to be consistent across a new sheet, an old sheet, and within a single fragment of glass.

Jack Mershon, an electrical engineer has reported a simulated trace-evidence scenario subjecting an iPhone 4S to a gunshot, generating glass particles from the iPhone's front and back covers. The report says that as SEM, EDS, and micro-CT are complementary tools for forensic science, so the combination of these tools have helped to establish a match between a glass particle and its source on the surface of an iPhone. The use of variable pressure imaging conditions, as well as wide field, depth, and resolution scan modes were critical for image acquisition. The creation of panorama images enabled the correlation of SEM, micro-CT, EDS and optical data. SEM 3D reconstruction and micro-CT imaging were used to correlate the surface and topography, as well as to give insight into the damage within the phone. Finally, EDS microanalysis data in the form of spectra, line scan, mapping, and large area mapping were acquired from samples to characterize the compositional variations within the glass, which provides corroborating evidence of the origin of the glass. (Mershon, 2021).

4. Conclusion

The concept of trace evidence in forensic science originates from Locard's exchange principle, and trace evidence is typically in the form of particles of skin, hair, fibers, clothing, soil, paint, and glass, among other materials. The effectiveness of SEM-EDX as a non-destructive technique,over the other destructive techniques, makes it the most suitable technique for the analysis of forensic evidences from a crime scene.

In the category of destructive techniques, atomic absorbance spectroscopy (AAS) is used to detect Pb, Ba, and Sb in trace samples. Inductively coupled plasma (ICP) combined with mass spectrometry (MS) can rapidly detect various elements, but is also a destructive technique. These techniques are slow, lack spatial specificity, and require highly trained personnel to run the instruments and to interpret the results (Hellmissetal, 1987).

The SEM-EDX technique, belongs to non-destructive method, has been used by many scientists to carry out the investigations and analysis of commonly retrieved forensic evidence from the crime scene. In this article, it is discussed in details that SEM-EDX can serve as a powerful tool for forensic scientists to classify and discriminate evidence material because they can simultaneously examine the morphology and the elemental composition of objects. The advantage of SEM, over any other light microscope, is their resolution< 1 nm. Thus, resolution of SEM is at least 100 times better than a light microscope. Another important region where SEM excels over light microscope is magnification. The magnification of SEM is at least 300 times better than a light microscope. One of the most important advantages of SEM is its ability to

combine imaging with the elemental analysis; when the SEM (imaging) is combined with energy dispersive X-ray spectrometry (elemental analysis), it is known as SEM–EDX.

The non-destructive nature of the SEM-EDX permits the retention of evidence, allowing for re-examination if necessary, making it a reliable and invaluable tool for forensic firearm investigations. The requirement for highly trained personnel has also been overcome with the introduction of automated SEMs (nanoscience.com). Many automated SEM are compact, has a great speed, thus saving floor space and time. The automated SEM comes with an automated particle analysis software built onto the platform, offering a dedicated GSR analysis solution that is compliant with instrument. The workflow of a typical GSR run involves defining a scan area for each sample stub, scanning the scan area frame by frame, using the backscattered electron detector to detect particles, and determining the elemental composition of each particle. The automated SEM also comes with various user-friendly features like customize and save workflows, Auto Video function to optimize brightness and contrast at any point throughout the GSR run and Dual Thresholding feature. (azom.com)

The analysis of evidence collected from a crime scene, such as GSR, car paint and glass residues, are discussed and explained with illustrations pertaining to the working principle and guidelines of the techniques employed. The obtained results are expected to be useful for the reconstruction of a crime scene under investigation, which can also be used as a tool in the subsequent judicial proceedings.

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