



Naif Arab University for Security Sciences
Arab Journal of Forensic Sciences & Forensic Medicine
المجلة العربية لعلوم الأدلة الجنائية والطب الشرعي
<https://journals.nauss.edu.sa/index.php/AJFSFM>



Nanotechnology: A Powerful Tool in Forensic Science for Solving Criminal Cases

تقنية النانو: وسيلة فعالة في مجال علوم الأدلة الجنائية لحل القضايا الجرمية



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Received 06 April 2021; Accepted 08 September 2021; Available Online 30 Dec. 2021

Abstract

Nanotechnology has emerged as a phoenix in the field of forensic science and proved to be of great importance in solving criminal cases where other techniques failed to provide conclusive results. This field of science possess humongous potential in the field of forensic science and assist in crime detection. It holds huge amount of value in making a positive contribution in assisting forensic experts and scientists in nabbing the criminals and most importantly prevent any wrongful conviction.

In the past decade, many researchers have reported the satisfactory application of Nano technique in Forensic Science for the analysis of latent fingerprints, drugs in alleged drug-facilitated crimes, warfare agent detection, DNA analysis, counter terrorism, GSR detection, post-blast residue analysis, security measures, etc. It has been proved to be a robust approach for the detection of crime with greater selectivity, sensitivity, reliability and results are produced in a timely appropriate manner.

The constant development of nanotechnology and its application in the field of Forensic Science over the past decade has been highlighted in this review article.

Keywords: Forensic Science, Nanotechnology, Nano Tracker, Fingerprint, Warfare Agents, Explosives, Drugs.



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المستخلص

ظهرت تقنية النانو في مجال علوم الأدلة الجنائية كوسيلة متطورة ومستحدثة، وأثبتت أنها ذات أهمية كبيرة في حل القضايا الجنائية، في حين فشلت تقنيات أخرى في ذلك أو أسفرت عن نتائج غير مجدية؛ حيث يوفر هذا المجال من العلوم إمكانات هائلة لعلوم الأدلة الجنائية من حيث تقديم المساعدة في الكشف عن الجرائم. ولذلك فهو يحظى بقيمة هائلة لدى خبراء الأدلة الجنائية والعلماء، والأهم من ذلك أنه يساعد على منع وقوع حالات إدانة خاطئة. خلال العقد الماضي، وأفاد العديد من الباحثين بنجاح تطبيق تقنيات النانو في مجال علوم الأدلة الجنائية فيما يتعلق بتحليل بصمات الأصابع الخفية، وتحليل المخدرات في الجرائم اليسر ارتكابها بالمخدرات، والكشف عن المواد السامة المستخدمة في الحرب، وتحليل الحمض النووي، ومكافحة الإرهاب، وكشف بقايا الطلقات النارية، وتحليل بقايا ما بعد الانفجار، بالإضافة لاستخدامها في التدابير الأمنية، إلخ. لقد ثبت أن تقنية النانو تمثل طريقة فعالة للكشف عن الجريمة حيث توفر مزيداً من الانتقائية والحساسية والموثوقية. كما تقوم هذه التقنية بالتوصل إلى النتائج في الوقت المناسب وبطريقة ملائمة.

تسلط هذه الورقة الضوء على تطور تقنية النانو وتطبيقاتها في مجال علوم الأدلة الجنائية خلال العقد الماضي، كما تبين أهمية تقنيات النانو في مواجهة التحديات التي تواجه التحقيقات الجنائية.

الكلمات المفتاحية: علوم الأدلة الجنائية، تقنية النانو، المتعقب النانوي، بصمات الأصابع، مواد سامة تستخدم في الحرب، المتفجرات، المخدرات.

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doi: [10.26735/YDDJ4516](https://doi.org/10.26735/YDDJ4516)

1. Introduction

Forensic science is a broad field which is a combination of different tools and techniques adapted from all the other scientific domains. It is an application of scientific tools and techniques derived from natural sciences which assist in solving criminal cases, collect evidences for the same, analyze them and produce the reports in the court of law. This field of science assists law enforcement agencies in nabbing the criminal and preventing the court of law from convicting any innocent person. Forensic science deals with collection, packaging, preservation, examination and analysis of the evidences collected from the crime scene which itself presents a big window for error, be it human or instrumental. Nanotechnology solves this problem as it gives precise, accurate, reliable, selective and highly sensitive result in real time as it employs use of Nano sprayers, Nano sensors, Nano-chips, and various other Nano technical methods which can analyze the sample on spot in real time leaving no window for error arising due to collection, preservation, transportation etc. It basically utilizes its increased surface to volume ratio proportion which can certainly be used for integration of all the time taking wet lab tedious protocols to be integrated into a small chip containing nanoparticles on the basis of nature of analyte and can be employed in hand held detector for real time investigations. "Lab-on-a-chip" and "Nano sprayers" are recent advancements in the field of nanotechnology assenting forensic science in crime scene investigation. These Nano sprayers have tremendous ability to collect the Nano sized evidences from the crime scene as in most of the cases, the amount of sample or evidence present on the scene of crime is in minute quantities. With the emerging capabilities of offenders operating at a molecular level, devices such as Nano sprayers, Nano sensors, etc. needs to be employed into the law enforcement agencies

for investigation. Nano sprayers collect trace evidences present in minute quantities which can be detected by nanoparticles embedded inside the lab on a chip devices. Lab-on-a-chip devices are small chips with selective nanoparticles embedded inside them which react chemically or biologically with the species or specimen they are selective to in particular. These devices can perform all the analytical (chemical or biological) procedures on a small amount of trace evidence collected from the crime scene and give accurate result in real time that is on spot which ultimately reduces the time and justice is done via court of law in a timely manner unlike the pending cases piling up due to traditional methods which require proper collection, packing, preservation and transportation to the forensic science laboratories.

The introduction of nanotechnology into forensic science will not only save labor and time but the speed with which the cases will be solved will generate a positive approach of the society towards the proceedings of law enforcement agencies because justice delayed is justice denied. Also, the advancements in nanotechnology like Nano devices, Nano sensors, Nano-chips, lab-on-a-chip, Nano tags etc. can be used in preventing crimes as well. Nanotechnology can be utilized to safeguard people from various criminal activities like violent crimes, rape cases, extortion, theft, fraud, forgeries and terrorism. It can also be utilized to protect children, women, teenagers and elder people from kidnapping, cybercrime, and abduction. Nanotechnology can help to reduce the piling up of cases and also to prevent criminal activities as there will prevail a fear of being caught.

2. Concern of the Study

In the past decades, there has been a huge spike in criminal cases, which continue to pile up



due to inefficient traditional tools and techniques, comparatively lesser professional staff in relation to the cases, contaminated samples, minute sample quantities and due to spillage or adulteration of sample (evidence) while being transported to laboratories. Nanotechnology debuted here in solving all these problems single handed. Nanotechnology utilizes the Nano sized particles in the analysis of criminal cases associated with drug analysis, fingerprint detection, DNA analysis, warfare detection, post-blast residue analysis, GSR (gunshot residue analysis), fraud, forgeries, time since death and Nano tagging for security measures. Nanotechnology produces accurate, precise, selective, sensitive, reliable and reproducible results in real time i.e. on spot reducing the time taken via traditional techniques for collection, packaging, preservation and transportation of the sample to their respective laboratories. Improper sampling and storage of samples can deteriorate the specimen or analyte as in cases of drugs. We can overcome this by using integrated laboratory protocols on a device called lab-on-a-chip containing Nano sized particles specific to their ligand for the analysis of such drug related crimes. Use of time-consuming and traditional methods delay the process of justice in a court of law which ultimately lower down the trust of society in the law enforcement agencies and offenders get their loop hole to escape in such scenarios encouraging them to commit more such crimes as they have no fear of law and justice.

Therefore, we need nanotechnology to aid forensic science in solving and preventing criminal cases in a timely, accurate and authentic manner. This will also help in a timely jurisdiction and execution of criminals and in safeguarding the society from such destructive forces.

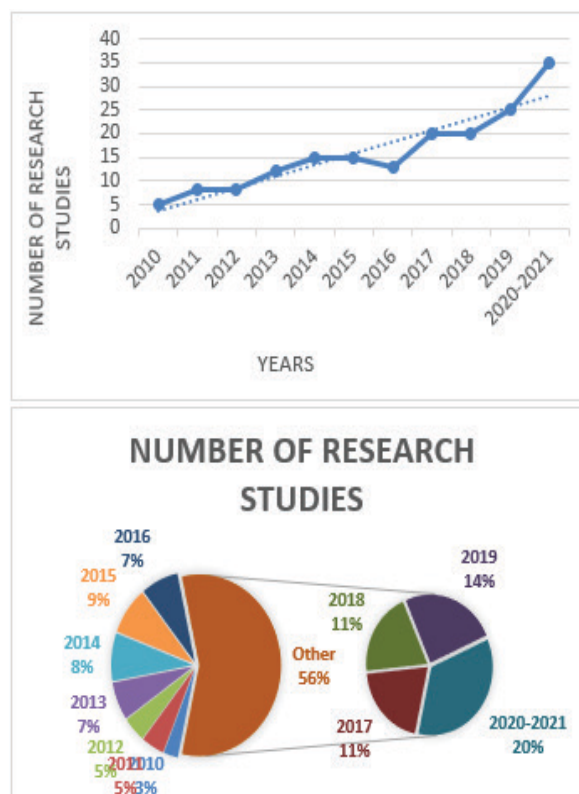


Figure 1- Graphical representation of number of researches reported for use of nanotechnology in forensic domains.

3. Applications of Nanotechnology in Forensic Science

Nanotechnology possesses a wide range of applications in the field of forensic science. A graphical representation of studies reported in the past decade has been shown in Figure-1.

The applicability of nanotechnology finds its ways in solving criminal cases as follows:

3.1 Nanotechnology in Analysis of Warfare Agents

Warfare agents are lethal chemical or biological agents which can cause mass destruction in a short span of time. These warfare agents have been in use since world war period for destroying the armies of opponents. Warfare agents have direct toxic effects on humans, plants and animals. These



components are highly toxic, sensitive, have rapid action potential and are highly persistent. Warfare agents exist in liquid, gas and aerosol form which gets dispersed into the environment rapidly causing mass destruction on exposure. On the basis of physiological effects on humans, warfare agents are classified as follows-

- i. Nerve agents
- ii. Blistering agents
- iii. Blood agents (vesicants)
- iv. Choking agents
- v. Riot controlling agents
- vi. Toxin
- vii. Biological agents (bacteria, viruses, etc.)

These warfare agents when dispersed into the environment causes mass destruction by acting on skin, CNS (Central nervous system), blood, respiratory tract, and eyes etc. The identification of these chemical and biological weapons becomes important to determine the root cause of the harmful side effects and the source from where these weapons have been released into the atmosphere. The physiological effects on humans can only be treated systematically only if the agents causing the disease is known. All these factors underpins the need for a method through which analysis of these warfare agents can be made in real time so that necessary action could be taken to minimize the casualties. Nanotechnology plays a vital role in the analysis of these warfare agents on spot by use of nanoparticles embedded in Nano sensors or biosensors that are highly selective towards their ligand molecule. The nanoparticles are chosen on the basis of the warfare agent which is being targeted i.e., chemical, biological or physical [1-3]. Chemical nerve agents like Sarin (GB), Sulphur mustard are highly volatile; therefore do not persist in environment for long. But toxins like Ricin persists in environment for longer

duration which needs immediate action for decontamination and prevention of further harmful effects. Nanoparticles here prove to be of great value in making a huge contribution by recognizing these agents through nanostructured sites where they bind, react and show a change in color. Nanoparticles have high potential applications in preparation of sensors for warfare agents, their destruction, tagging, detection and tracking [1]. Strem Chemicals, in collaboration with Max-Planck-Institute fuer Kohlenforschung, have developed sensors for chemical, biological and radiological warfare agents, special reactors and metal oxides for the destruction of the warfare agents as well as their analysis on the spot [1]. Microchips having nanoparticles specific to a chemical or biological agent are embedded in a gel covered by a layer of polymer and a layer of substrate is placed on top of it. These microchips are very small and their thickness is in nanometer range. These nanomaterial microchips are embedded on to the clothes, buildings, arms, and transports etc. These microchips changes the color showing presence of warfare agent when it comes in contact with the specific agent. Nanotechnology not only helps in the analysis and detection of warfare agents on spot but also decontaminates the area where these agents are dispersed. Nanoparticles of non-toxic metal oxides, when come in contact with warfare agents, forms a strong bond and convert them into non-reactive by-products n [1, 3].

Gold NPs are extensively used for the detection of various types of trace evidences. Sensors made up of gold nanoparticles are used for the calorimetric and florescent detection of warfare agents (Figure-2).

1) Calorimetric detection

Gold nanoparticles are red in color in the free form when dispersed in a solution at a range i.e., 3



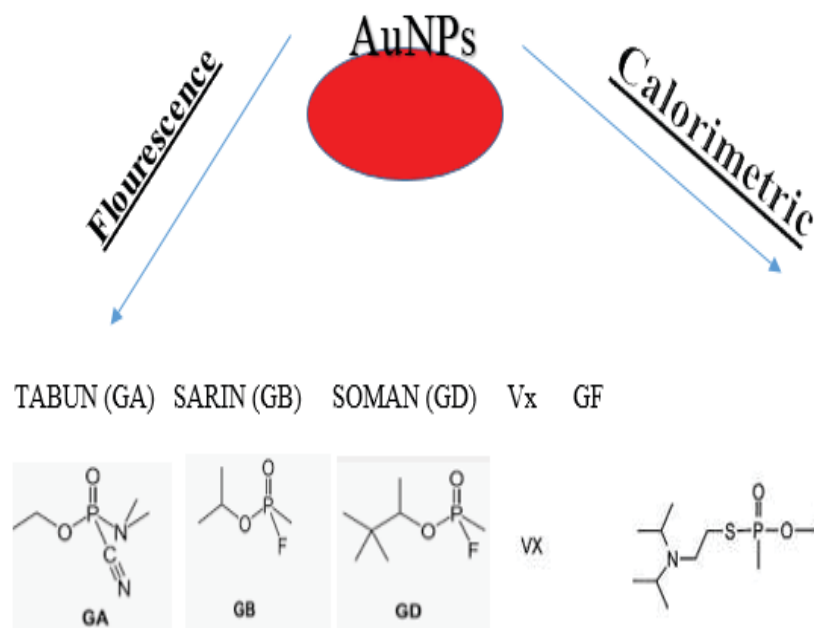


Figure 2- Warfare agents that can be detected using AuNPs.

to 10 nm due to strong SPR (Surface Plasmon Resonance) around 520 nm. SPR is a keystone property of these gold nanoparticles that arises from the free electrons present at the metal surface. These free electrons on exposure to light shows absorption in visible and near infrared region of electromagnetic spectrum. Au nanoparticles on agglomeration results in Surface Plasmon coupling and absorption shifts from visible to near infrared region and color changes from red to blue. This change in color from red to blue can again change to red on re-dispersion of the agglomerated AuNPs (gold nanoparticles) can occur due to environmental factors, pH changes, reaction with specific ligands, ionic strength etc. as shown in Figure-3 [4, 5].

Gold nanoparticles probed with a specific agent can lead to functionalization of these AuNPs with the nerve agents which shows change in color from red to blue. The concentration of the agent could be further analyzed based on the intensity of color change via UV-Vis spectroscopy. Lanthanum

(La³⁺) probed gold nanoparticles are used for the detection of Methyl Parathion. Lanthanum combines with Methyl parathion causing agglomeration of AuNP-La particles through coordination and bonding interactions. A change in color from red to blue is observed based on the concentration of the analyte. Dichlorvos can be detected in water, wheat etc., via AuNPs capped with ascorbic acid. Dichlorvos, on reaction with AuNPs attached with Ascorbic acid, forms strong hydrogen bonding. This agglomeration shifts the color of solution from red to blue suggesting presence of Dichlorvos in the sample. AuNPs modified with cysteine can be used for the detection of phosgene. Ricin detection is possible by aptasensor qualitative analytical procedure where gold nanoparticles' reactivity increased for TMB (3,3',5,5'-tetramethylbenzidine) in presence of the Ricin Binding Aptamer resulting in enhanced absorption of TMB from the light to dark blue color [4].

Another important fact about AuNPs agglomerate is that in the presence of the metabolite formed after



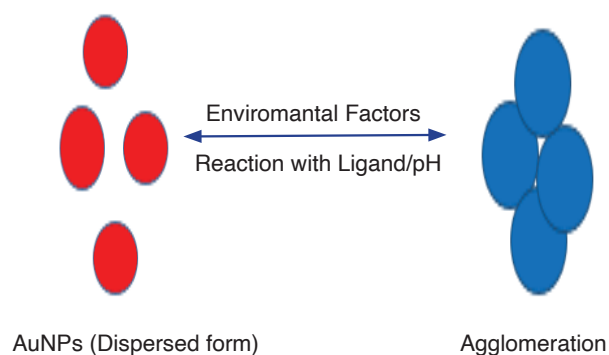


Figure 3- Mechanism of transition in color of AuNPs.

the breakdown of acetylcholine i.e., Thiocoline (Tch). Tch gets attached to gold nanoparticles through strong Au-S and causes agglomeration. While in the presence of warfare agents like Sarin (GB), paraoxones, Vx (venomous agent X), Soman (GD) etc. Acetylcholine is not metabolized and Thiocoline is not produced because these agents inhibit ACHE i.e., acetylcholine esterase enzyme responsible for the breakdown of acetylcholine. AuNPs in the absence of thiocholine shows no agglomeration and no transition in color of solution from red to blue is observed [4, 5].

1) Fluorescence based detection

AuNPs based fluorescence works on the principle of surface modified fluorescence (SMF) and fluorescence resonance energy transfer (FRET) mechanism. FRET occurs as a result of dipole-dipole interaction causing resonant energy transfer between the donor fluorophore and acceptor fluorophore as depicted in Figure-4. Fluorescence is affected by the metal atoms present in its surroundings [4, 6]. SMF could be applied to depict the fluorescence change due to the presence of excited fluorophore. For instance, if distance between the metal surface and fluorophore is less than 10 nm, then it absorbs energy and change in the fluorescence signal is observed. FRET be-

ing the popular mechanism uses an excited donor fluorophore that excites the ground state acceptor molecule which absorbs energy at a particular wavelength and re-emits energy in the form of fluorescence. It is based on various factors but the most important one is the distance between the donor and acceptor. FRET works best in the range between 1 to 10 nm. AuNPs have high absorption coefficient and broad energy wavelength which makes them excellent quenchers for FRET based procedures. OPH (organophosphate hydrolase) conjugated with AuNPs were prepared by researchers on incubation with fluorescent enzyme inhibitor decoy for paraoxon. In the absence of substrate decoy attached to the surface of OPH-AuNPs show enhanced surface fluorescence while in the presence of substrate, decoy detaches from the surface of NPs where substrate binds causing a change in fluorescence signal. For example, Eu^{3+} ions when attached to AuNPs undergo strong enhancement, but detaches from the surface in presence of organophosphorus compounds giving rise to the changed fluorescence signal. Fluorescence sensors can also be prepared in the form of films like indole self-assembled multilayer (SAM) films for detection of organophosphorus compounds prepared by l-cysteine modified quartz/AuNPs surface by electrostatic interaction between OPH and l-cysteine. These produce linear fluorescence signals for monocrotophos and methyl parathion [4-6].

3.2 Nanotechnology in Developing Latent Fingerprints

Latent fingerprints are basically those invisible print evidence at scene of crime left by the offender



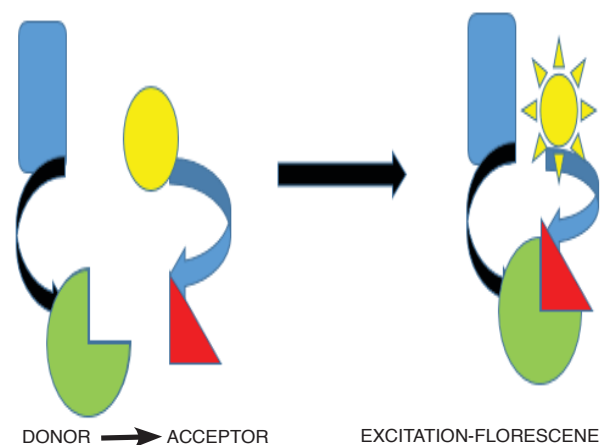


Figure 4- FRET (Fluorescence resonance energy transfer).

which on analysis and detection can generate the whole profile of that criminal. These prints are formed due to deposition of fatty acids, amino acids and proteins released in the form of sweat via pores present in the ridges of fingers and palm. Commonly, very few prints are found at the crime scene which needs proper attention and care. The study of these fingerprints becomes very important in linking the suspect to the crime and in exonerating the innocent. These fingerprints can be developed with the help of various types of powders like carbon black, white, colored, and magnetic powders etc. (Figure-5). These powders, due to big granular size, not only attaches to the fatty acid part of the print but also gets deposited in the furrows which stains the background as well. Background staining makes analysis and comparison of the fingerprint with suspect a tedious task. Whereas, nanoparticles are microscopic in size which is thousand times smaller than the size of traditional developing powders. These particles have increased surface area to volume ratio due to decrease in size. The reaction between these particles and the fatty acids takes place on surface which is higher in nanoparticles due to the enhanced surface to a volume ratio. More the reactivity more is

the enhancement of latent fingerprints [1].

Nanotechnology in latent fingerprint development was first used by late Dr. Menzel et al. (2000) where he developed a photo luminescent CdS (cadmium sulfide) semiconductor Nano crystal conjugated with dioctyl sulfosuccinate to enhance latent fingerprint detection. The concept he applied was to add Nano crystal fluorescent dye on objects/articles that have been pre-fumed with cyanoacrylate ester [7].

Gold nanoparticles suspended in petroleum ether having long hydrocarbon chain find huge applicability here as these stick to the latent fingerprints by forming a strong hydrophobic interactions which, on immersion into silver ions solution, forming a dark silver metallic outline corresponding to the imprint that is very clear for analysis. AuNPs attach to the surface containing latent prints like glass, plastic, etc. where it forms electrostatic as well as hydrophobic interactions. The Au attached sites works as nucleation sites for silver ions present in physical developer which in the presence of a reducing agent, reduces to dark metal [1, 8]. These developed fingerprints are very stable unlike traditional ones. For non-porous surfaces, cadmium selenide nanoparticles or sulphide nanoparticles attached to long chains of amines suspended in petrol are used. The fingerprints developed using this solution showed fluorescence under UV-light on illumination. Professor Fred Rowell found silica nanoparticles to be useful in detecting the age of the fingerprint donor at the University of Sunderland, where the principle was detection of chemicals present in the fingerprint. Nanoparticles used along with SALDI-TOF2-MS (Surface-assisted laser desorption-Time of flight-mass spectrophotometry) were useful in indicating whether the person handled any sort of illegal drug, explosive, etc. Zinc oxide (ZnO)



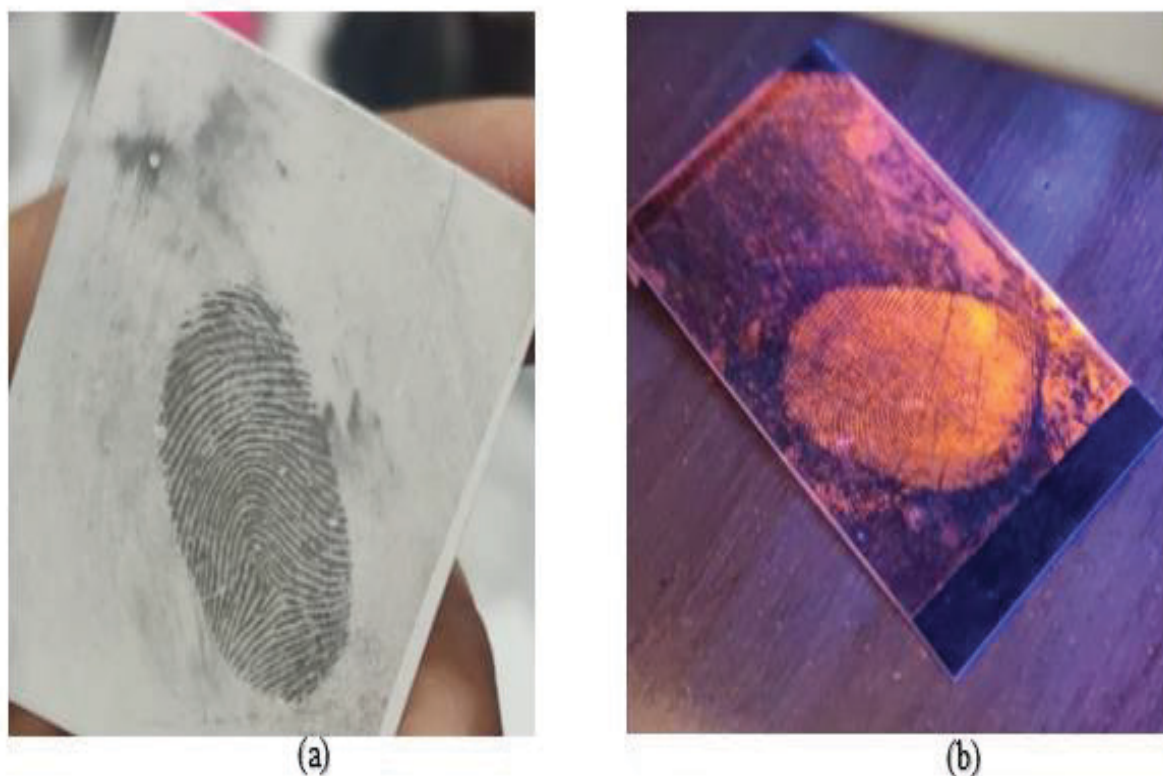


Figure 5- Fingerprint developed using a) Black powder and b) Fluorescent powder. [Central fingerprint bureau, NCRB, DELHI].

NPs (nanoparticles) works beautifully in wet conditions [1]. X-Ray, along with nanoscale developer, makes fingerprint present on gun casings visible even after wash. TiO (titanium oxide) NPs are quite efficient for both porous and non-porous surfaces develops prints and are stable too but do not fluoresce until and unless doped with a suitable dye [1, 9, 10]. CdSe (cadmium selenide) nanocrystal make latent fingerprints present on adhesives visible in presence of stabilizers like Mercaptoethanol and mercaptoacetic acid [9]. Molybdenum disulphide (MoS₂) particles form grey deposits on interaction with fatty acid which could be photographed and preserved for further investigative studies.

Gold NPs mixed with antibodies specific to cotinine, a metabolite of nicotine, are useful in detecting fingerprints and also helps in identifying if the suspect was a smoker or not. Similarly, drugs and

explosives if handled by the person could be detected by adding specific ligand according to the molecule of interest [1].

Another method for developing these fingerprints is Micro X-Ray fluorescence (MXRF). This techniques relies on the amount of inorganic matter present in the prints left on the surfaces.

Unlike conventional methods where reagents react with fatty acids or amino acids to generate the fingerprints, MXRF produces these prints based on the inorganic elemental composition in prints. Most commonly observed inorganic residues are potassium and chloride ions along with silicon, calcium, and aluminum. This technique is very advantageous over others as it is non-destructive and the inorganic matter present in prints is quite stable too. After analysis the prints remains intact which further can be used for other additional tests like Gunshot residue



analysis. The elements foreign to hands like TiO₂ or ZnO nanoparticles are present in lotions, sunscreen which can also be detected via MXRF [7].

3.3 Nanotechnology in the Analysis of Drugs

Drug facilitated crimes referred to as date rape crimes are those scenarios where the illicit drugs are used for committing the crimes like rape, sexual assaults along with murder, extortion, robbery etc. The detection and identification of such drugs thus becomes a crucial task for forensic investigators in assisting law enforcement agencies. The conventional methods are associated with setbacks like high-cost, time consumption, sensitivity, and instrumentation constraints etc. Here nanotechnology blends in to assist forensic science as it is well known for its cost effectiveness, sensitivity, selectivity and rapidity. Devices used for “on-site” detection of drugs at trace levels are Nano chips, Nano sensors and Nano probes etc. [11]

Different type of chemo sensors, biosensors, nano sensors and aptasensors, depending upon the nature of analyte, are used in biological and chemical analysis. These nano sensors have various electrical, thermal and optical properties which can be used as recognition site or as transducer. Major drugs of abuse which need immediate action in the field of forensic investigation are listed as follows:

- | | |
|-----------------|------------------|
| i. Amphetamines | vii. Diazepam |
| ii. Alcohol | viii. Clonazepam |
| iii. Opioids | ix. Flunitazepam |
| iv. GHB | x. MDMA/Ecstasy |
| v. Ketamine | xi. Midazolam |
| vi. Alprazolam | xii. Zolpidem |

A smart and rapid system for the detection of these drugs encompasses the use of citrate-based gold nanoparticles as shown in Figure-6. AuNPs, stabilized with citrate, work as a probe in the detection of codeine sulphate using smartphone that

makes it a simple and portable technique. It gives results on the spot with rapid and ultrasensitive Nano aggregation. Codeine sulphate have multiple binding sites with oxygen and sulphate group forms strong coordinated complexes with AuNPs by exchanging weak surface bound citrate ions leading to cross linking of AuNPs. These particle on aggregation form big complexes and change color from red to blue based on the intensity of cross linking. The aggregation is caused due to the high affinity of codeine sulphate towards the Nano sensors containing AuNPs because of the three oxygen and one sulphate group. Camera of the smartphone is used to detect even the minor calorimetric change in the presence of codeine sulphate. Similarly Clonazepam detection could be made with the help of gold nanoparticles in the presence of melamine [11, 12].

Quantum dots (QDs) are Nano crystals which work on the principle of fluorescence resonance energy transfer (FRET). When UV light falls on these nanoparticles, they emit light of different colors. These QDs have found immense importance in the detection of drugs in biological samples. The components of these optical biosensor QDs are-

- i. Light source
- ii. Optical transmission medium
- iii. Immobilized biological recognition element (enzymes, antibodies, receptors, or microbes)
- iv. Optical detection system

Cocaine detection was made possible using these QD aptasensors. The probe developed for cocaine detection was anti cocaine aptamer bonded with MNS 4.1. Cocaine binds with the aptamer through a hydrophobic pocket. After incubation period of 12 hours, decolourization of blue colored precipitate occurs in samples containing cocaine. Here Thioflavin T can also be used as a fluorescence indicator as when cocaine will bind to the anti-cocaine



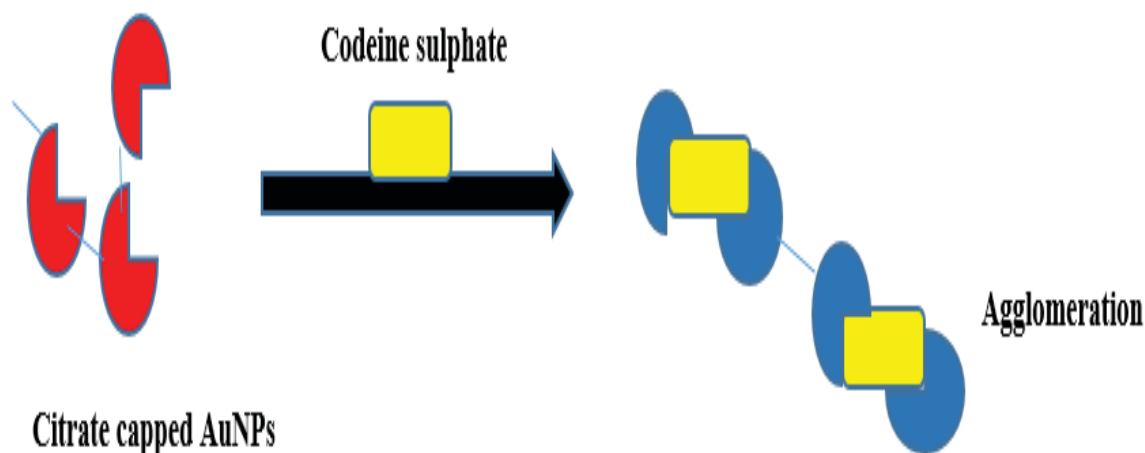


Figure 6- Demonstration of interaction between codeine sulphate and citrate capped gold NPs.

aptamer–MNS 4.1, a drastic change in fluorescence is observed [11, 13].

Fingerprints containing cocaine is deciphered using nanoparticles of silver and gold ranging from 10 to 20 nm. These fingerprints on development with Au and Ag NPs if analysed via FTIR gives enhanced IR peaks of Cocaine at 2948 cm^{-1} , 1714 cm^{-1} , and 1324 cm^{-1} [14].

3.4 Nanotechnology in the Analysis of Explosives

Terrorism has emerged as one of the greatest crime against mankind. There is a vital need for techniques through which the explosives could be detected if found in unexploded form, hidden in luggage, toothpaste, vehicles, garages, aircraft or even for the detection of precursors used for manufacturing explosives. Nanotechnology has proven its value in the detection of explosives and post blast residue via Nano sensors like nanotubes, electronic noses, nano mechanical devices and Nano chips [15].

Tracing explosives is a costly and tedious job as it covers a wide range of geographical area and

conventional techniques are quite intricate and costly to work with. The use of nano-sensors has been shown cost-effective to manufacture and give on site reports [16]. Recent studies have revealed the use of curcumin based nanoparticles in the detection of trinitrotoluene (TNT). The curcumin powder is extracted from turmeric. These curcumin NPs are highly fluorescent and sensitive probes which can detect up to 1nM TNT in an aqueous solution. Another method for detecting TNT is via amine terminated nanoparticles [12]. An antibody-based protocol for PETN (pentaerythritol tetranitrate) was developed recently where the antibody against PETN reacts with the ligand where calorimetric analysis could be done [16]. The AuNPs have been exploited for their Plasmons where they are used for SERS (Surface Enhanced Raman scattering spectroscopy). Raman spectrum generated is based on a molecular vibrational energy which is particular and specific to each and every molecule. Therefore, detection of explosives using SERS via AuNPs is possible. Silver and gold nanoparticles are chosen ones as their SPR range is 400 -800 nm, these are



stable, inert and strong oxidizing as well as reducing agents. Nanoparticles, attached with cysteine, form Mesienheimer complexes-explosives with nitro aromatic compound and detection of RDX (royal demolition explosive) up to 0.15 mg/L in water samples is also possible. QDs as previously mentioned can also be used for the detection of DNT (dinitrotoulene), TNT, RDX, PETN and tetryl [17].

Nano sensor system for detection like nanotubes, nanowires have high surface area to volume ratio which are high absorption capabilities. The system consists of array of sensors generating electrical signal for vapor being absorbed by the sensing chamber. The vapor produced by explosives when come in contact with these sensors gets absorbed into them and produce a chemical signature or a print unique to a chemical compound which can be matched to the library of known standard compounds. [18].

3.5 Nanotechnology in Gunshot residue analysis (GSR)

Nanotechnology also finds its way in the analysis of Gunshot residues. These residues are minute particles that primarily consists of Barium, Antimony and Lead which are the components of priming mixture of ammunition. So whenever somebody shoots, these particles of gunshot residue come out followed by discharge of firearm and are often found on the hands, face and clothes of the shooter. High resolution Scanning electron microscope (SEM) can image these particles at nano-scale and X-ray spectroscopy can be used to determine the elemental composition of these evidences. Another very revolutionary technique for analyzing, identifying and quantifying evidences like GSR, ink, soil, and fingerprint is "ion beam analysis". This instrument works by drawing out elemental composition of evidence found at crime scene by comparing it with reference material or standard. The ion beam

analysis is very selective, specific and sensitive technology which employ ion beam of milli electron volts (meV) to identify the evidences or samples in question in the sub monolayer and its elemental depth ranging from few to 10 nm. The surface of the sample is struck by a beam of ions where charged particles interact with electrons and nuclei of atoms of the material being analyzed. Particle emission occurs which reveal the qualitative and quantitative elemental analysis of the sample. Further, ion beam analysis could also be coupled with particle-induced X-ray emission, particle-induced Gamma-ray emission for the analysis of the samples [12, 15].

3.6 NanoTrackers

Nano trackers are secret patterns introduced into variable product to prevent thieves from stealing them for example in Mobile phones, watches, pen drives, etc. Nanotrackers are being injected into the body of criminals so that if they even try and succeed in escaping from the prisons, could again be caught via using this tracking system. Criminals after being released from the jail or correctional centers might not always follow the societal norms and show deviant behaviour which needs to be monitored. This is possible by injecting Nanotrackers into their body which will enable the monitoring process of such released convicts a less tedious process. Serial offenders can be traced down if they get involved in crimes again [12].

The objects can also be tagged with peptide based taggants which physically mark them. These can be applied on the surface of items and when recovered, swabs of such taggants on analysis show color or fluorescence change. Also, as per the Locard's exchange principle, these taggants can also be recovered from the person who is involved in the crime associated with the tagged article [19].



Table 1- Forensic applications of nanotechnology.

S.No.	NANOTECHNOLOGIES	APPLICATIONS	REFERENCES
1.	Metal Nano clusters of sensors, microchips embedded in polymer layers containing specific ligands. AuNPs in alkanethiols layer	Warfare agent detection- Calorimetric change in color in presence of warfare agents	[1, 59,65]
2.	Biosensors (antigen antibody interaction mechanism) Magnetic nanoparticles (NPs), AuNPs, Ln doped NPs, Quantum dots, Ag Nano rods, nanowires, carbon nanotubes	Biological warfare agent detection E.g.; Variola virus, bacterial species, Brucella spp. , BoNT, etc.	[2]
3.	Carbon NT, graphenes, Nano diamonds, fluorescence sensors, Nano platforms	Chemical warfare agent detection	[3, 60]
4.	Lab on a chip, AuNPs, triarylcarbinol functionalized AuNPs	Calorimetric detection of warfare and nerve agents	[4, 5, 60, 61 93]
5.	Ag-NPs and tetraphenylene probe and silica NPs	H ₂ O ₂ detection	[88]
6.	Chromophore- chromosensors AuNPs/QDs Nano probes	Ricin detection	[69]
7.	Turmeric extracted curcumin NPs –fluorescent probe Amine terminated NPs	TNT detection up to 1 Nm in aqueous solution	[12,62]
8.	AuNPs and QD	DNT, TNT, RDX, PETN, tetryl detection	[17, 36]
9.	Cellulose Nano crystal	Nitrophenolic explosives detection	[89]
10.	Electronic noses, sensors, nanowires, Nano chips, nanotubes, Nano mechanical devices	Explosives detection	[18, 36, 85, 86]
11.	Tetraphenylethene probe based fluorescent silica NPs	Nitramine explosive detection	[35]
12.	Luminescent sensors, Carbon dots	Nitoaromatic explosive detection	[37, 38]
13.	p-ATP (Para-aminothiophenol) attached AuNPs	TNT detection	[63]
14.	Cysteine modified AuNP- SERS probe (free surface enhanced Raman spectroscopy)	TNT detection	[64]
15.	Amine functionalized CdSe- SiO ₂ NPs fluorescence Nano sensor	Picric acid detection	[87]
16.	CdS semiconductor nanocrystal capped dioctylsulfo-succinate, ZnO-SiO ₂ nanopowder SPR method	Development of latent fingerprint	[12, 18, 25]
17.	ZnO (20nm), CdSe/ZnS (10nm) Nano powder in conjugation with SALDI-TOF-MS	Generate UV fluorescent fingerprints on development.	[1, 8, 9, 16. 24, 27, 36, 50]



Continue Table 1

S.No.	NANOTECHNOLOGIES	APPLICATIONS	REFERENCES
18.	Gold NPs capped with antibody against cotinine (metabolite of nicotine)	Along with fingerprint development, determines if the person is a smoke or not.	[1]
	Silica stable NPs	Age and ethnicity of fingerprint donor.	
19.	TiO ₂ -NPs (show fluorescence when conjugated with dyes)	Develop fingerprints on porous and non-porous surfaces.	[9, 53]
20.	Molybdenum disulfide NPs	Fingerprint analysis- forms grey deposits on reaction with fatty acids	[1, 36]
21.	Gold NPs capped with citrate ions / AuNPs with Ag-PDA	Silver metallic prints obtained on analysis of latent fingerprints	[1, 27, 35]
22.	Silver NPs (1-200 nm size), Aluminum oxide NPs (30- 60 nm size)	Latent FP analysis Al-NPs give good contrast in wet conditions	[8,29, 36, 49]
23.	Eu ³⁺ doped Al ₂ O ₃ Nano crystalline powder(36.8 nm average) Fluorescent starch based carbon NPs (10-210 nm)	Latent Fingerprint development Enhanced contrast on non-porous surface.	[8, 9, 22, 29, 36, 49]
24.	Poly (styrene-alt maleic anhydride)-b-polystyrene (SMA-b-PS) functionalized gold NPs	Latent Fingerprint development	[16]
	Quantum dots		[35, 36, 50]
25.	Rare earth fluorescent nanomaterial- YVO ₄ ,(red fluorescence) Eu Nano crystal, LaPO ₄ , Ce, Tb, Nano belts	Latent Fingerprint development	[51,54]
26.	Red and green color emitting Calcium – molybdate Nano phosphors	Enhanced development of latent fingerprint	[55]
27.	Citrate stabilized AuNPs with smart-phone camera	Codeine sulfate detection	[12, 25]
28.	AuNPs in presence of melamine	Clonazepam detection	[12, 25, 36]
29.	Gold and silver NPs	Cocaine detection in fingerprints	[14]
30.	Nano probe based detection	Detection of morphine ,codeine, methamphetamine, clonazepam	[11, 73, 90]
	Nano chips based detection	Detection of gamma hydroxybutyrate , benzoyllecgonine, cocaine	
31.	Mix and detect- Thioflavin with anti-cocaine aptamer MNS 4.1	Cocaine detection (fluorescence)	[13]
32.	CdSe/ZnS QDs Peptide taggants	Morphine biosensor Fluorescence for drugs	[19]
33.	Carbon dots, metal NPs, QDs and magnetic iron oxide NPs, AuNPs, citrate-AuNPs, Thymine-AuNPs	Halide detection	[44]



Continue Table 1

S.No.	NANOTECHNOLOGIES	APPLICATIONS	REFERENCES
34.	Functionalized magnetic NPs like tryptophan modified Si-NPs, L-cys-Au/Fe ₂ O ₃ , etc.	Separation of enantiomers	[45]
35.	Portable/ wearable electrochemical sensors	Detect metabolites of drugs on site in saliva, sweat and tears.	[46-48]
36.	Label free gold NPs – SERS probe	Cyanide detection	[66]
37.	Nanoparticles of gold, Fe ₂ O ₃ , metals, Carbon nanotubes	Analysis of narcotics	[91]
38.	AuNPs	Calorimetric detection of organophosphate pesticides	[92]
39.	Egg-white encapsulated gold Nano clusters	Detection of organophosphorus compounds	[56]
40.	Aptamer based AuNPs attached with cationic peptide	Detection of malathion in environmental samples	[57]
41.	Electronic sensor- polyaniline nanofibers and single wall carbon nanotube	Malathion detection	[58]
42.	Nanoparticles of Cu ₂ O	Destroy methyl parathion	[67]
43.	AuNP modified with citrate and HAuCl ₄ .3H ₂ O calorimetric light scattering assay	Pb(II) detection in paints, plastic and water samples Pb detection	[70] [71, 72]
	AuNPs capped with glutathione and pentapeptide	Mercury(II) , Lead (II) detection	[75, 76]
	AuNPs	Mercury detection	[77,78,79]
	Anti- Hg ₂ ⁺ aptamer with AuNP	Mercury (II) detection	[80]
	Thymine modified AuNPs		
44.	Aligent 2100 Bioanalyzer Biochips Microfluidic devices	DNA Quantification and analysis , Real time PCR quantification	[1, 16, 81]
45.	Light it up	Detection of biological fluid- calorimetric analysis	[1, 16]
46.	Magnetic and silica based NPs	Extraction of DNA	[1, 21, 29]
47.	Magnetic NPs with dimercaptosuccinic acid, Sensors , AuNPs with Halloysite nanotubes, Si-NPs	Extraction of DNA	[22, 82, 84]
48.	Nano trackers Barcodes and disappearing inks	For tracking criminals, stolen items Establishing authenticity and checking if the item is counterfeited and duplicated	[20,36]



Barcodes, in addition to Nano trackers, also help in preventive forensics. These are specific patterns drawn on articles which can determine the authenticity of a product which helps in preventing the forgery or duplication of such items. The counterfeiting, forgery or duplication of any item raises a question on the reliability of a particular product being manufactured by a company leading to socio-economic losses and damage to the reputation of such companies. The barcodes along with watermarks are reliable factors that can be checked for the authenticity of a product which cannot be reproduced on the counterfeited items.

Nano technology has introduced other unique techniques such as Nano fibres and Nano dots with different color which is used for the detection and authentication of products. The Nano fibres and Nano dots can be analysed using different forensic light sources such as UV (ultraviolet), IR (infrared), etc. that will detect and decipher these Nano fibres and Nano dots [20].

3.7 Nanotechnology in DNA Analysis

DNA is a unique identifying feature of a person as is a fingerprint of an individual. Analysis of DNA gives the whole genetic profile of a person and is one of the best identity marker to detect a person involved in a crime. It involves the study of repetitive tandem repeats, RFLP (restricted fragment length polymorphism), etc. DNA acts as one of the most crucial corroborative evidence in linking the suspect and victim to the crime. Magnetic nanoparticles are used in DNA analysis in a more effective and faster way than the conventional techniques like capillary electrophoresis. The magnetic NPs like Fe_3O_4 extract the DNA from the hair, saliva, semen at a very faster rates and also quite effectively when used as

a solid support medium. The samples could be obtained from suspect, victim or even from the scene of crime [21]. The first step involves the extraction of DNA from these cells which is very tough task if the sample collected from the scene of crime for comparison is in nanograms. Nano technological techniques like lab-on-chips, bionanochips, and microfluidic systems has showed its value in DNA analysis with less volume in less time up to even one molecule of DNA [12].

Microfluidic system is used for the quantification of samples after PCR (polymerase chain reaction) amplification. Bioanalyzer Agilent 2100 is available in market that analyses DNA molecules (mitochondrial DNA) after PCR amplification at a very fast speed and is being extensively used in most forensic science laboratories for analysis of a single sample in less than 2 minutes. Biochips are probes manufactured by lithography process where DNA captures the probe molecules and show fluorescence. These probes have been successfully used in real time PCR amplifications where the continuous analysis along with amplification can be achieved. Also, DNA molecules immobilized over gold pads and carbon nanotubes have been probed with Atomic Force Microscope (AFM) for analysis [1]. Forensic Science & Drug Monitoring department of King's College (London), developed a solution called 'Light it up' used for the detection of different types of biological materials/evidences like saliva, sweat, seminal fluid, blood, etc. This detection helps in a way that if biological fluid is detected, then the extraction of DNA from a particular matrix becomes easy for further investigation. Here, a solution of mixture of fluorescent tagged NPs when bind to molecule under investigation emits fluorescence in different colors based on the biological material [1, 21].



Another magnetic DNA extraction method is using bifunctional supramagnetic nanoparticles manufactured by modified pyrolysis method and surface modification with meso-2,3-dimercaptosuccinic acid (DMS) containing carboxyl groups that absorb DNA molecule. The DMS modified NPs used successfully for DNA isolation from human blood that isolated up to 86.16 % DNA [22].

3.8 Nanotechnology in Questioned Document Analysis

Not only we deal with documents in our daily life but we also rely on documents for various purposes, be it any order, recruiting employees, establishing credibility, etc. These documents are used to profile an individual, thus possess high vulnerability and needs very much attention at every step before authentication. The process of distinguishing a fraudulent, built up or forged document from an original comes under forensic document examination. Surprisingly, nanotechnology plays an important role in studying the questioned documents at nano scale. The analysis of ink used for execution and the sequence of strokes tell a lot if there has been any addition, substitution or obliteration. Atomic force microscopy (AFM) works at nanoscale and analyze the crossing inks by scanning the documents surface via which the strokes and 3 Dimensional surface morphological characters can be established. It also provides information about the line alignment formed by ball point pen ink and ribbon dye [8, 29, 31].

3.9 Nanotechnology in Bloodstain age estimation

Bloodstains at a crime scene are quite useful in the estimation of how the perpetrator committed the crime, entered or took exit from the scene of crime

and also the direction of the movement. However, the age estimation of the bloodstain remain to be a big question during investigations. However, the age of the bloodstain could be determined, then the exact time of crime could be established. Here, AFM rises up like a phoenix and resolve this issue in daily routine analytical practices. With time, the morphological characters and cellular components of blood undergo changes based on the environmental factors which can be analyzed by AFM at nano level. It analyzes morphological and surface elasticity of Red blood cells which decreases with time due to degradation and coagulation process by force-distance curves recording. This bloodstain age estimation by recording the elasticity curve with respect to time further help investigators in drawing out successful conclusions [31].

3.10 Nanotechnology in Determining Time since Death (TSD)

Determination of time since death (TSD) is a crucial task in forensic medicine and legal medicine to estimate the time when crime was committed or when the victim died. There are various physical and chemical postmortem changes that occur in a body. Blood, aqueous and vitreous humor, spinal fluid, pericardial fluid, synovial fluid undergo changes which could be analyzed for the estimation of TSD. Vitreous humor out of all these is one reliable factor that remains unchanged for a long period of time after death and undergo a slow linear change for upto 96 hours, i.e., change in levels of amino acid is observed. Recently, a very cost effective, sensitive and specific "lab-on-a-chip" method was devised to analyze the levels of amino acid cysteine which could tell the TSD precisely for upto 96 hours as cysteine increases significantly in vitreous humor and depicts a linear correlation with time. TSD could also be estimated via flow cytometry method which



quantify the DNA degradation rate of brain and spleen for upto 96 hours. Also, fluorescent nanoparticles can help in quantifying the concentration of vitreous humor along with flow cytometry in near future [12, 26, 31].

Some of the applications of nanotechnology in different domains of forensics have been summarized in Table-I.

4. Conclusion

With the emerging trends in technologies and resources, criminals have also updated themselves and now operate at molecular level leaving no or nanoscale evidences which needs immediate attention for analysis. This review paper summarizes how nanotechnology can help forensic experts overcome this problem. The nanoscaled trace materials can be detected using nanotechnology like lab on a chip, Nano chips, Nano sensors, biosensors, microfluidic systems, quantum dots and various types of nanoparticles like gold, silver, zinc oxide, titanium oxides, magnetic nanoparticles, etc. These technologies will help forensic scientists in assisting law enforcing agencies in solving criminal cases and nabbing the perpetrator. Nanotechnology plays a very important role in the analysis of drugs, explosives, post-blast residue, chemical warfare agents, development of latent fingerprints, tracking items and criminals, DNA detection, security measures, etc. The integration of time taking procedures carried out in lab on a microchip assists in real time investigation reducing time taken in collection, packing and transportation of evidences. The Nano trackers and barcodes help in preventing and combating crimes where the tracking of any item or criminal is possible along with its authenticity detection. These new techniques might sooner or later replace the time taking lengthy pro-

cesses for analysis of evidences which will prevent piling up of cases in forensic laboratories.

5. Future aspects

The wide horizon of nanotechnology enables forensic scientists in detecting trace evidence which otherwise is difficult via conventional techniques. This technology holds immense potential in various fields of forensic and other applied sciences, where more and more advancement can lead to the development of new techniques and formulation of related equipment. The combination of traditional technique along with new techniques will enhance the investigation system manifolds. The selectivity, sensitivity and cost-effectiveness of these technologies might replace all the costly, time consuming wet experiments that are performed in forensic laboratories. Lab-on-a-chip is one such device that is said to be an integrated form of lab procedures which gives calorimetric tests for trace materials on-site. Cost-effectiveness of these procedures is another basic necessity which is superior to the traditional methods. The rapid on-site analysis will fasten the speed of court proceedings as well leading to faster convictions and vice-versa.

Instrumental technique analysis can only be done by a well-trained expert or a professional which is not the case in new emerging nanotechnologies. A person with comparatively less expertise or training can handle and report the results generated.

With the everyday increasing crime rate, the need for modern and faster tools is justified. The addition of nanotechnology in the field of forensic science will improve the quality of justice.

Conflict of Interest

The authors declare that they have no conflict of interest.



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