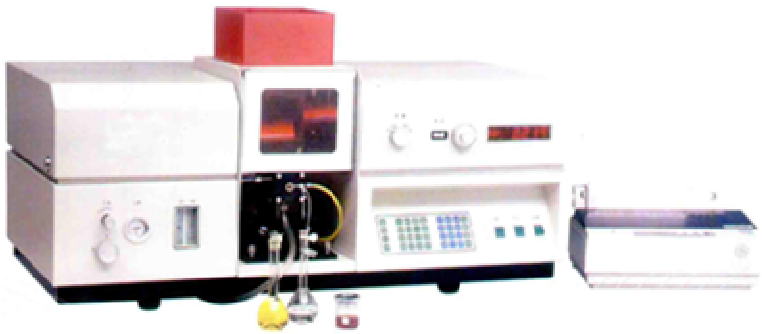
1. Spectroscopy

a. Atomic absorption spectrophotometer

An atomic absorption spectrophotometer analyzes the concentration of elements in a liquid sample based on energy absorbed from certain wavelengths of light (usually 190 to 900 nm). Atomic absorption spectrophotometers typically include a flame burner to atomize the sample (most commonly a hollow cathode lamp), a monochromator, and a photon detector. Depending on the model, some atomic absorption spectrometers are equipped with a turret or fixed lamp socket that can hold multiple lamps (up to eight) to reduce downtime between samples or allow for sequential analysis.

Typical sensitivity for an atomic absorption spectrometer using a flame burner is in the parts per million range. For trace analysis, a graphite furnace can be used in place of a flame burner to increase the sensitivity by several orders of magnitude (in the parts per billion range). Atomic absorption spectrophotometers are used in many industries including environmental testing, metal analysis, semiconductor manufacturing, petroleum and chemical production, and in pharmaceuticals

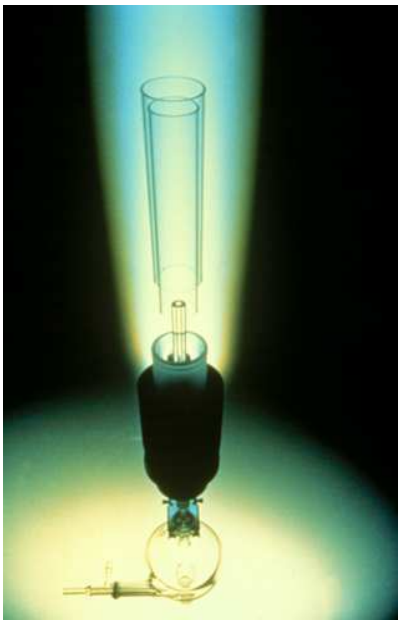


b.UV visible spectrometer

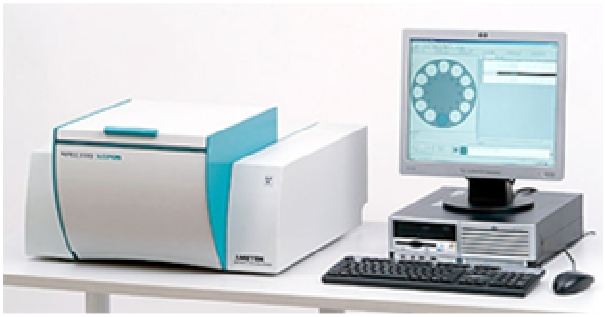
Ultraviolet-visible spectroscopy or **ultraviolet-visible spectrophotometry** (**UV-Vis** or **UV/Vis**) refers to absorption spectroscopy in the ultraviolet-visible spectral region. This means it uses light in the visible and adjacent (near-UV and near-infrared (NIR)) ranges. The absorption in the visible range directly affects the perceived color of the chemicals involved. In this region of the electromagnetic spectrum, molecules undergo electronic transitions. This technique is complementary to fluorescence spectroscopy, in that fluorescence deals with transitions from the excited state to the ground state, while absorption measures transitions from the ground state to the excited state.[1]

c. Inductively coupled atomic emission spectrophotometer

Inductively coupled plasma atomic emission spectroscopy (ICP-AES), also referred to as inductively coupled plasma optical emission spectrometry (ICP-OES), is an analytical technique used for the detection of trace metals. It is a type of emission spectroscopy that uses the inductively coupled plasma to produce excited atoms and ions that emit electromagnetic radiation at wavelengths characteristic of a particular element.[1][2] The intensity of this emission is indicative of the concentration of the element within the sample.



d. X-ray fluorescence spectrophotometer



X-ray fluorescence spectrometers (XRFs) use a spectroscopic technique that is commonly used with solids, in which X-rays are used to excite a sample and generate secondary X-rays. The X-rays broadcast into the sample by X-ray fluorescence spectrometers eject inner-shell electrons. Outer-shell electrons take the place of the ejected electrons and emit photons in the process. The wavelength of the photons depends on the energy difference between the outer-shell and inner-shell electron orbitals. The amount of X-ray fluorescence is very sample dependent and quantitative analysis requires calibration with standards that are similar to the sample matrix.

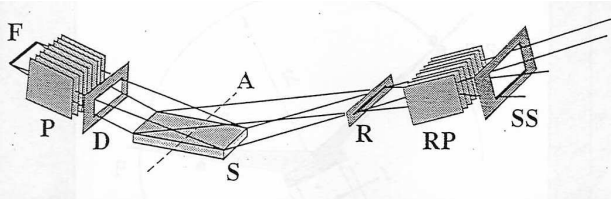
The solid samples used with X-ray fluorescence spectrometers are usually powdered and pressed into a wafer or fused in a borate glass. The sample is then placed in the sample chamber of the XRF spectrometer, and irradiated with a primary X-ray beam. The X-ray fluorescence is measured either in simultaneous or sequential modes, and is recorded with either an X-ray detector after wavelength dispersion or with an energy-dispersive detector.

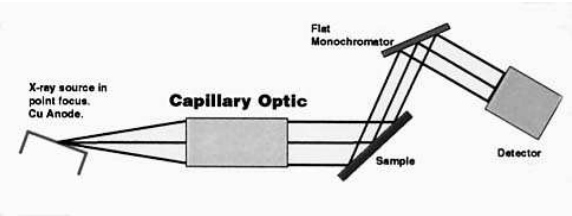
X-ray fluorescence spectrometers measure the emitted X-ray fluorescence in either a simultaneous fashion, or sequential. Simultaneous mode typically measures the entire wavelength range around the emission line of interest simultaneously, while sequential mode typically measures one wavelength at time.

Wavelength dispersive detectors use a nondestructive analysis technique for the identification and quantification of elements in a material. Wavelength dispersive spectroscopy is the measurement x-ray energies emitted from the bombardment of an energy source impinged upon the material, producing characteristic x-rays. Energy dispersive X-ray fluorescence spectrometers also use nondestructive analysis techniques for the identification and quantification of elements in a material. Energy dispersive spectroscopy is the measurement x-ray energies emitted from the bombardment of an energy source impinged upon the material, producing characteristic x-rays.

XRF was originally used to analyze geological samples. The advancement of computers and other technologies allowed the technique to develop even further. XRF found its place in many different types of analytical laboratories and some industrial inspection systems. X-ray fluorescence spectrometers provide a number of distinct advantages including easy sample preparation, nondestructive rapid multi-element analysis, and the ability to screen unknowns in a wide array of sample matrices such as liquids, solids, slurries, powders, pastes, thin films, air filters, and many others. Because of these advantages the technique, it is widely used for research, in industrial settings, and by quality assurance analysts.

e. XRD spectrophotometer





X-ray diffraction (XRD) is a versatile, non-destructive technique that reveals detailed information about the chemical composition and crystallographic structure of natural and manufactured materials.

Crystal lattice

A crystal lattice is a regular three-dimensional distribution (cubic, rhombic, etc.) of atoms in space. These are arranged so that they form a series of parallel planes separated from one another by a distance *d*, which varies according to the nature of the material. For any crystal, planes exist in a number of different orientations - each with its own specific *d*-spacing.

Constructive interference

When a monochromatic X-ray beam with wavelength *lambda* is projected onto a crystalline material at an angle *theta*, diffraction occurs only when the distance traveled by the rays reflected from successive planes differs by a complete number *n* of wavelengths.

Bragg's Law

By varying the angle *theta*, the Bragg's Law conditions are satisfied by different *d*-spacings in polycrystalline materials. Plotting the angular positions and intensities of the resultant diffracted peaks of radiation produces a pattern, which is characteristic of the sample. Where a mixture of different phases is present, the resultant diffractogram is formed by addition of the individual patterns.

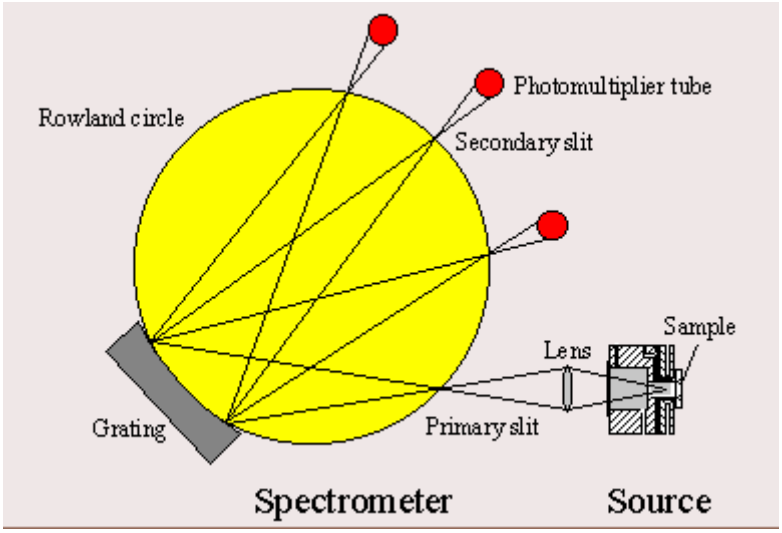
Based on the principle of X-ray diffraction, a wealth of structural, physical and chemical information about the material investigated can be obtained. A host of application techniques for various material classes is available, each revealing its own specific details of the sample studied.

f. Glow Discharge Optical Emission Spectroscopy - GDOES

Glow Discharge Optical Emission Spectrometry (GDOES) is a well established method for the analysis of in-depth concentration profiles

By this method, it is possible to perform fast simultaneous analysis of many elements, in a wide range of concentrations and depths. Especially in the field of galvanised applications GDOES shows a very good performance in respect of the speed of analysis as well as the quality of the chemical analysis.

GDOES combines sputtering and atomic emission to provide an extremely rapid and sensitive technique for element depth profiling. During analysis, a plasma is generated in the analysis chamber by the applied voltage between the anode and the cathode (the sample surface) in the presence of argon under low pressure. Ionised Ar atoms cause sputtering of the sample area. Sputtered atoms excited in the plasma rapidly de-excite by emitting photons with characteristic wavelengths.


Photomultiplier tube
Rowland circle
Secondary slit
Sample
Lens
Grating
Primary slit
Spectrometer
Source

2. Chromatography

Chromatography is a technique that is widely used for the separation of gases, liquids, or dissolved substances. After being separated, the individual constituents can be identified or purified through standard techniques. Its accuracy and ease of use make chromatography a fundamental procedure in analytic chemistry.

a. Gas chromatography

Gas chromatography (GC) is an instrumental technique used forensically in drug analysis, arson, toxicology, and the analyses of other organic compounds. GC exploits the fundamental properties common to all types of chromatography, separation based on selective partitioning of compounds between different phases of materials. Here, one phase is an inert gas helium (He), hydrogen (H2), or nitrogen (N2) that is referred to as the mobile phase (or carrier gas), and the other is a waxy material (called the stationary phase) that is coated on a solid support material found within the chromatographic column. In older GC systems, the stationary phase was coated on tiny beads and packed into glass columns with diameters about the same as a pencil and lengths of 6 to 12 feet, wound into a coil. The heated gas flowed over the beads, allowing contact between sample molecules in the gaseous mobile phase and the stationary phase. Called “packed column chromatographs,” these instruments were widely used for drug, toxicology, and arson analysis. Around the mid-1980s, column chromatography began to give way to capillary column GC, in which the liquid phase is coated onto the inner walls of a thin capillary tube (about the diameter of a thin spaghetti noodle) that can be anywhere from 15 to 100 meters long, also wound into a coil. Capillary column chromatography represented a significant advance in the field and greatly improved the ability of columns to separate the multiple components found in complex drug and arson samples. However, a few applications still require packed columns.

The purpose of the gas chromatograph is to separate mixtures into individual components that can be detected and measured one at a time. A plot of the detector output is called a chromatogram, which charts the detector's response as a function of time, showing the separate components. The separation occurs based on differences in affinities for the two phases. As shown in the figure, the sample is introduced into the GC column by way of a heated injector, which volatilizes all three components and introduces them into the gas flowing over the stationary phase. In this example, the compound represented by the arrowhead has the least affinity for the stationary phase.

As a result, it moves ahead of the other two components and will reach the detector first. The compound symbolized by the diamond has the greatest affinity for the stationary phase and spends the most time associated with it. As a result, this compound will be the last to reach the detector. Separation has been achieved based on the different affinities of the three types of molecules found in the sample. In reality, complex mixtures cannot always be completely separated, with some compounds emerging from the column simultaneously. This is called coelution, which can often be overcome using detectors such as mass spectrometers (MS).

In most forensic applications of GC, a sample is prepared by dissolving it in a solvent, and the solution is injected into the instrument using a syringe. For example, to analyze a white powder suspected of being cocaine, a small portion is weighed out and dissolved in a solvent such as methylene chloride, methanol, or chloroform. A tiny portion of the sample is then drawn up into a syringe and injected into the heated injector port of the instrument. The mobile phase gas (called the carrier gas) also enters the injector port, picking up the volatilized sample and introducing it into the column where the separation process occurs. If the sample contains cocaine, it will emerge from the column at a given time (known as the retention time) that can be compared to the retention time of a known standard sample of cocaine. The retention time in conjunction with information obtained from the detector is used to positively identify the compound as cocaine if indeed it is present. Another method of sample introduction for GC is called pyrolysis, in which a solid sample such as a fiber or paint chip is heated in a special sample holder to extreme temperatures, causing the sample to decompose into gaseous components that can then be introduced into the GC. Pyrolysis is used when the sample is not readily soluble in common GC solvents.

A number of different detectors are available for use in gas chromatography. In forensic applications, the most commonly used are mass spectrometry (often abbreviated as MSD for mass selective detector), flame ionization (FID), and nitrogen-phosphorus (NPD). The MSD is the most common of the three, principally because it can provide definitive identification of compounds (in almost all cases) along with quantitative information. The FID is used in arson cases because of its sensitivity to hydrocarbons, the primary ingredient in most accelerants. The NPD is used in drug analysis and toxicology.

One of the advantages of chromatographic systems such as GC is the ability to provide both qualitative information (identification of individual components) and quantitative information (concentrations of individual components). If the instrument is properly calibrated, it can be used to determine quantities of materials present in samples, and this is commonly done in drug analysis. For example, the purity of a drug sample seized as evidence can provide important information and may be used as part of the prosecution. Similarly, when a sample of plastic or a fiber is analyzed by pyrolysis GC, the relative abundance of the individual components can be useful in creating a chemical signature or fingerprint of that particular sample.

b. High-performance liquid chromatography - HPLC

High-performance liquid chromatography (HPLC) also know as high-pressure liquid chromatography is an instrumental system based on chromatography that is widely used in forensic science. The “HP” portion of the acronym is sometimes assigned to the words high pressure (versus high performance), but it refers to the same analytical system. HPLC is used in drug analysis, toxicology, explosives analysis, ink analysis, fibers, and plastics to name a few forensic applications.

Like all chromatography, HPLC is based on selective partitioning of the molecules of interest between two different phases. Here, the mobile phase is a solvent or solvent mix that flows under high pressure over beads coated with the solid stationary phase. While traveling through the column, molecules in the sample partition selectively between the mobile phase and the stationary phase. Those that interact more with the stationary phase will lag behind those molecules that partition preferentially with the mobile phase. As a result, the sample introduced at the front of the column will emerge in separate bands (called peaks), with the bands emerging first being the components that interacted least with the stationary phase and as a result moved quicker through the column. The components that emerge last will be the ones that interacted most with the stationary phase and thus moved the slowest through the column. A detector is placed at the end of the column to identify the components that elute. Occasionally, the eluting solvent is collected at specific times correlating to specific components. This provides a pure or nearly pure sample of the component of interest. This technique is sometimes referred to as preparative chromatography.

Many different types of detectors are available for HPLC. The simplest and least expensive is the refractive index detector (RI). Although this detector is a universal detector, meaning it will respond to any compound that elutes, it does not respond well to very low concentrations and as a result is not widely used. On the other hand, detectors based on the absorption of light in the ultraviolet and visible ranges (UV/VIS detectors and UV/VIS spectrophotometers) are the most commonly used, responding to a wide variety of compounds of forensic interest with good to excellent sensitivity. The photodiode array detector (PDA) is especially useful since it can produce not only a peak-based output (a chromatogram) but also a UV/VIS scan of every component. In many ways, the ideal detector for HPLC is a mass spectrometer (MS), which provides both quantitative information and in most cases a definitive identification of each component (qualitative information). However, HPLC-MS

systems are relatively complex and expensive and are not readily available in all labs. Other detectors that are sometimes used include fluorescence detectors (which are very sensitive) and electrochemical detectors.

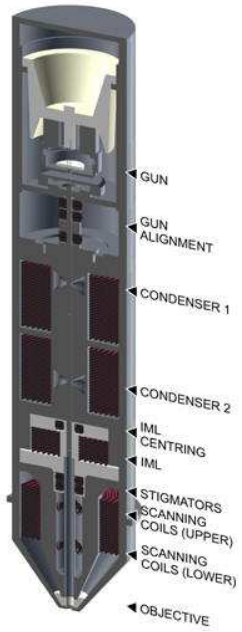
Unlike in gas chromatography (GC) in which the mobile phase is an inert gas, the mobile phase in HPLC can be one of many different solvents or combinations of solvents. This imparts to HPLC a greater flexibility and range of application than has GC. Because the sample does not have to be converted to the gas phase, compounds such as explosives that break down at high temperatures are much more amenable to HPLC than GC. For HPLC, all that is required is that the sample be soluble in the solvents selected for the analysis. In addition, there are several types of HPLC defined by the type of mobile phase and stationary phase that is used. For forensic applications, one of the most commonly used types of HPLC is referred to as “reversed phase.” In this type of HPLC, the mobile phase is a solvent or mix of solvents that are “polar,” meaning that different parts of the individual solvent molecules carry a partial positive or negative charge. Water, methanol (methyl alcohol), ethanol (ethyl alcohol), and acetone are examples of polar solvents. The stationary phase in reverse phase HPLC is a nonpolar material such as a long chain hydrocarbon molecule. In this type of HPLC, components in the sample will partition and separate based on their degree of interaction with the stationary phase relative to the mobile phase. In other words, the separation is based primarily on the relative polarity of the sample molecules. Reverse phase HPLC is used in drug analysis (LSD for example), analysis of cutting agents such as sugars, explosives, and gunshot residue (GSR), and forensic toxicology.

Normal phase HPLC uses a polar stationary phase and a nonpolar mobile phase, but this is not widely used in forensic applications. Size exclusion chromatography (SEC) is more common and separates compounds based on relative sizes. The stationary phase in SEC is composed of a gel with different sizes of microscopic pores through it. The larger the molecule, the longer it takes for it to navigate through the pores and reach the detector. SEC is useful for the analysis of large molecules that come in a range of sizes such as plastic polymers, proteins, and nitrocellulose, a component of GSR. Chiral chromatography, a relatively recent development, is making inroads into forensic science since it is capable of separating enantiomers, molecules that are mirror images of each other. This capability is particularly valuable in forensic toxicology and drug analysis. Finally, ion exchange chromatography is available for detection species such as nitrate (NO_3^-) and other ions.

3. Microscopy

a. Scanning Electron Microscope

The scanning electron microscope (SEM) is a type of electron microscope that images the sample surface by scanning it with a high-energy beam of electrons in a raster scan pattern. The electrons interact with the atoms that make up the sample producing signals that contain information about the sample's surface topography, composition and other properties such as electrical conductivity**.**



b. Transmission electron microscopy - TEM

Transmission electron microscopy (TEM) is a microscopy technique whereby a beam of electrons is transmitted through an ultra thin specimen, interacting with the specimen as it passes through. An image is formed from the interaction of the electrons transmitted through the specimen; the image is magnified and focused onto an imaging device, such as a fluorescent screen, on a layer of photographic film, or to be detected by a sensor such as a CCD camera.

TEMs are capable of imaging at a significantly higher resolution than light microscopes, owing to the small de Broglie wavelength of electrons. This enables the instrument's user to examine fine detail—even as small as a single column of atoms, which is tens of thousands times smaller than the smallest resolvable object in a light microscope. TEM forms a major analysis method in a range of scientific fields, in both physical and biological sciences. TEMs find application in cancer research, virology, materials science as well as pollution and semiconductor research.

c. Focused ion beam - FIB

Focused ion beam, also known as **FIB**, is a technique used particularly in the semiconductor and materials science fields for site-specific analysis, deposition, and ablation of materials. An FIB setup is a scientific instrument that resembles a scanning electron microscope (SEM). However, while the SEM uses a focused beam of electrons to image the sample in the chamber, an FIB setup instead uses a focused beam of ions. FIB can also be incorporated in a system with both electron and ion beam columns, allowing the same feature to be investigated using either of the beams. FIB should not be confused with using a beam of focused ions for direct write lithography (such as in proton beam writing), where the material is modified by different mechanisms.

4. Particle size analyzer

Particle size analyzers measure the sizes of grains or particles in a sample. They use methods such as light scattering, sedimentation, laser diffraction etc to calculate particle sizes. Particle size analyzers can measure the sizes of many particles in a sample very quickly and can provided data on particle size distributions, which is of value to a great many industries.