CENTRAL UNIVERSITY OF GUJARAT


HR-TEM


ESCA

NMR

P-XRD

Central Instrumentation Facility

CENTRAL UNIVERSITY OF GUJARAT
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The Central University of Gujarat (CUG) was established by the Government of India in 2009 through an Act of Parliament (Act No. 25 of 2009). It is emerging as a centre of excellence and is a seat of intensive academic activities with state-of-the-art laboratories and teaching-learning resources. It has earned and identified distinguished reputation for its teaching, research and expansion and outreach achievements. With serval international and national MoUs with university and institutes of repute and having hosted and having been called upon to host and organize academic, intellectual and scientific events of national significance and policy implication the university enjoy an enviable image and stature in academic and scientific circles in India and abroad.

The CUG has established instrumentation facilities for catering research need of Chemical Sciences, Environment Sciences, Life Sciences, Nano Sciences and Material Sciences. The development and establishment of the instrumentation facility and literature will update research. The advanced instruments procured have been centralized under the central instrumentation facility (CIF) at CUG. Instruments are being used for chemical, biological and environmental sample analysis. Advance instruments are also used for characterization of nano materials and their applications. CIF - instruments at present, provides instrumentation facility to the research students and faculty of CUG. CIF services will also be extended to the other institutes and university in the country.

Vision

The CUG has vision to establish itself as a centre of excellence with social commitment by integrating modern, scientific and technological knowledge and skills with the basic human ethos and values. The university shall set forth a model in teaching, research and personality development and create doyen human resource with a sense of responsiveness towards society, the country and the world.

Nodal Officer

Prof. Keshav Lalit Ameta, Dean, SAMS
Nodal Officer
Central Instrumentation Facility (CIF)
Central University of Gujarat

The Central University of Gujarat was established through an Act of Parliament in 2009. The Central University of Gujarat is a premier institution and is emerging as a centre of excellence while imparting teaching and research in higher education in science, humanities and social sciences. The instruments required for the multidisciplinary research in chemical sciences, life sciences, environmental sciences, and nano sciences have been procured.

The instruments are a vital part of the methodology of modern science. The realization of the present day of accomplishing in many disciplines such as chemical, biological, environmental, nano, and material sciences have come out by applying modern scientific instruments. Technology includes developing and using modern scientific instruments and techniques that involve fresh insights, new concepts, and innovative approaches to scientific research. The development and establishment of an instrumentation facility is the cumulative result of synchronizing the technologies involved in physics, chemistry, and biology.

The instrumentation facility available in central instrumentation facility (CIF) includes Powder X-ray Diffraction (P-XRD) and Single-crystal X-ray Diffraction (SC-XRD), Electron Spectroscopy for Chemical Analysis (ESCA), High-Resolution Transmission Electron Microscope (HR-TEM), Nuclear Magnetic Resonance (NMR), Atomic Absorption Spectrophotometer (AAS), Elemental Analyzer (CHNS/O), Gas Chromatography (GC), Liquid Chromatography-Mass Spectrometry (LC/MS-QTOF), Inductively Coupled Plasma Optical Emission Spectrometer (ICP-OES), Scanning of Electron Microscope (SEM), etc. for quality research.

LIST OF CIF INSTRUMENTS

- Powder X-ray Diffraction (P-XRD) and
- Single-crystal X-ray Diffraction (SC-XRD)
- Atomic Absorption Spectrophotometer (AAS)
- Elemental Analyzer (CHNS/O)
- High-Performance Liquid Chromatography (HPLC)
- Gas Chromatography (GC)
- Liquid Chromatography-Mass Spectrometry (LC/MS-QTOF)
- Fourier Transform Infrared Spectroscopy (FTIR)
- Brunauer Emmett Teller (BET) Surface Area Analyzer
- Inductively Coupled Plasma Optical Emission Spectrometer (ICP-OES)
- Spectroscopic Ellipsometer
- Refractometer Index (RI)
- 500 MHz Fourier-Transform Nuclear Magnetic Resonance (FT-NMR)
- Dynamic Light Scattering (DLS)
- Scanning of Electron Microscope (SEM)
- Scanning of Electron Chemical Microscope (SECM)
- Atomic Force Microscope (AFM)
- Electron Spectroscopy for Chemical Analysis (ESCA)
- Ozonolysis
- High Resolution Transmission Electron Microscope (HR-TEM)
- Thermo Gravimetric and Differential Thermal Analysis (TG/DTA)
- Differential Scanning Calorimetry (DSC)
- Gel Permission Chromatography (GPC)
- Potentiostat
- UV-visible Spectrophotometer
- Porosimeter
- Polarimeter
- Total Organic Carbon (TOC)
- Fluorescence-Activated Cell Sorting (FACS)
- Ultra-Centrifuge
- Real Time PCR (RT-PCR)
- High Speed Centrifuge
- Matrix-Assisted Laser Desorption/Ionization (MALDI-TOF)
- Confocal Laser Scanning Microscope (CLSM)
- Fluorescence microscope
- DNA Sequence
- Fast Protein Liquid Chromatography (FPLC)

CIF instruments: Working Principle and Application

High resolution Transmission Electron Microscopy (HR TEM)

Model: JEOL JEM 2100 TEM HR LaB6 Version, Make: JEOL

Principle

In this technique, a beam of high-energy electrons (120-200keV) is collimated by magnetic lenses and allowed to pass through a specimen under high vacuum. The transmitted beam and a number of diffracted beams can form a resultant diffraction pattern, which is imaged on a fluorescent screen kept below the specimen.

Applications

- The HRTEM is used to study of particle size and their morphology of materials
- The energy-dispersive X-ray (EDX) analyzer additional arranged to determine the composition of nano particle (NP).
- The selected-area electron diffraction (SAED) patterns provides crystallographic information (amorphous/crystalline/single crystal) From selected regions of the sample.
- Cross sections and crystallization of metallic alloys, semiconductors, and microstructure of composite materials, etc.

Electron Spectroscopy for Chemical Analysis (ESCA)

Model: B002961 ESCA + Base System, Make: Omicron

Principle

In ESCA technique, when a primary X-ray beam of energy interrupts on sample atoms, inner shell electrons are emitted and the energy of the emitted electrons is measured. The difference in the energy of the interrupted X-ray and the emitted electrons gives the binding energy (E_b) of the electron to the atom. The chemical form/environment of the atom affects the binding energy to a considerable extent and gives rise to some chemical shift, which can be used to identify the valence state of the atom and its exact chemical form.

Applications

- It is mainly used for surface analysis and especially in the qualitative identification of the elements in a sample.
- The chemical environment around the atoms can also be estimated based on the chemical shifts.
- The binding energy of elements can be calculated to identify the elements in sample.
- It is also useful in determining the valence states of the atoms present in various moieties in a sample.
- Quantitative analysis can be made by determining the intensity of the ESCA lines of each element, etc.

High Performance 500 MHz FT-NMR Spectrometer

Model: AV III 500 MHz, Make: Bruker

Principle

In NMR technique, the substances absorb energy in the radio frequency region of the electromagnetic spectrum under influence of a strong magnetic field. It is a well-known fact that the nuclei of the atoms bonded to each other in molecules spin on an axis like a top. Since nuclei are positively charged, this spin will create a small magnetic field. If an external magnetic field is applied to these nuclei this magnetic field will split into two energy levels. The energy difference is very small and corresponds to radiofrequency energy which is unique for every molecule and will give the information regarding the nature of the compounds and the presence of various functional groups and their environment.

Applications

- The NMR instrument used for identification and structural analysis of organic compounds.
- It is a tool for qualitative analysis and determination of structure, dynamics, reaction state, chemical environment of molecules, etc.
- It gives valuable information regarding the position of the functional groups in a molecule and provides distinguished spectra for the isomer.
- The detailed precise information on the structure of the compounds can be obtained using this technique with other magnetic nuclei like Pt^{135}, F^{13}, P^{31}, N^{15}, etc.

Powder and Single Crystal XRD

Model: Bruker AXS D8 Focus P-XRD Bruker and AXS D8 VENTURE SC-XRD, Make: Bruker

Principle

In this technique the primary X-rays are made to fall on the sample substance. Because of its wave nature, like light waves, it gets diffracted to a certain angle. This angle of diffraction, which differs from that of the incident beam, will give the information regarding the crystal nature of the substance. The power of a diffracted beam is dependent on the quantity of the corresponding crystalline substance; it is also possible to carry out quantitative determinations.

Application

- The diffraction of X-rays is a good tool to study the nature of the crystalline substances.
- In crystals the ions or molecules are arranged in well-defined positions in planes in three dimensions.
- This technique provides vital information regarding the arrangement of atoms and spacing in between them and also to find out the chemical compositions and phase of the crystalline substances.
- Analysis of crystal structure, determination of unit cell dimensions, measurement of sample purity, average grain size determination, crystal defects, etc.
- The sample under study can be a thin layer of crystal or in powder form.

Inductively Coupled Plasma (ICP) Emission Spectroscopy

Model: 7300 DV, Make: Perkin Elmer

Principle

The ICP is an atomic emission technique, related to the flame photometry except that the atoms and ions present in the sample are excited in high temperature gas plasma. The plasma provides very high temperature and hence energy. Hence, almost all the atoms present in the sample can be excited with this technique ending up with high efficiency (a hotter source increases both atomization efficiency and excitation efficiency). Thus, the emissions from the atoms would be more intense and even very small concentrations of metals/metal ions can be detected and measured accurately.

Application

- This technique mostly used for detection of trace metals, etc.
- Applications include water testing, soil and other material analysis.
- It is also largest uses in medical and forensic field, specifically study of toxicology, etc.

Liquid Chromatography-Mass Spectrometry (LC/MS-QTOF)

Model: Advanced version LC/MS – QTOF, Make: Agilent

Principle

LC/MS is a hyphenated technique combining the separation power of HPLC, with the detection power of mass spectrometry. It uses an interface that will eliminate the solvent and generate gas phase ions, transferred to the optics of the mass spectrometry. Obtain spectra and molecular mass identification for each peak eluted from the chromatography column.

Application

- Molecular weight determination of liquid sample
- Environmental Application: detection of phenyl urea herbicides, etc.
- Detection of low levels of carbonyl in food, pesticides in foods and beverages, etc.

Scanning Electron Microscopy (SEM)

Model: EVO 18 , Make: Carl Zeiss

Principle

In this technique, an electron beam is focused onto the sample surface kept in a vacuum by electro-magnetic lenses (since electron possesses dual nature with properties of both particle and wave an electron beam can be focused or condensed like an ordinary light) Further, the beam is scanned over the surface of the sample. The backscattered and secondary electrons from the sample are then fed to the detector and then to a cathode ray tube through an amplifier, where the images are formed, which gives the information on the surface of the sample.

Application

- SEM technique applied to the surface studies of metals, ceramics, polymers, composites and biological materials for topography as well as compositional analysis, etc.
- Topography: The surface features of an object and its texture (hardness, reflectivity, etc.)
- Morphology: The shape and size of the particles making up the object (strength and chips, etc.)
- Composition: The elements and compounds that the object is composed of and the relative amounts of them (melting point, reactivity, hardness, etc.)
- Crystallographic Information: How the grains are arranged in the object (conductivity, electrical properties, strength, etc.)

Atomic Force Microscope (AFM)

Model: XE-70, Make: Park System

Principle

This technique operates by measuring the forces between the sample and the tip. The tip is close to the sample surface to detect the repulsive force between the atoms of the tip material and the sample. The probe tip is mounted at the end of a cantilever of a low spring constant and the tip-to-sample spacing is held fixed by maintaining a constant and very low force on the cantilever. If the tip is close to the sample surface, the repulsive force will induce a bending of the cantilever. This bending can be detected by a laser beam, which is reflected off the back of the cantilever. Thus by monitoring the deflection of the cantilever, the surface topography of the sample can be tracked. AFM operates at two modes: Repulsive or contact mode – which detects the repulsive forces between the tip and sample; Attractive or non-contact mode - which detects the van der waals forces that act between the tip and sample.

Application

- AFM find applications widely in material sciences especially for surface studies on a nano scale range.
- AFM finds its applications in measuring the hardness of materials.
- Force spectroscopy is used in biophysics to measure the mechanical properties of living material (such as tissue or cells).

Thermogravimetric/ Differential Thermal Analysis (TG/DTA)

Model: TG/DTA7300, Make: EXSTAR

Principle

In this technique, the change in sample weight is measured while the sample is heated at a constant rate, under air (oxidative) or nitrogen (inert) atmosphere. This technique is effective for quantitative analysis of thermal reactions that are accompanied by mass changes, such as evaporation, decomposition, gas absorption, desorption and dehydration. The micro-balance plays a significant role; during measurement. The change in sample mass affects the equilibrium of the balance. This imbalance is fed back to a force coil, which generates additional electromagnetic force to recover equilibrium. The amount of additional electromagnetic force is proportional to the mass change. During the heating process, the temperature may go as high as 1000 degree celsius inside the furnace. This technique measures the temperature difference between a sample and a reference material as a function of temperature as they are heated or cooled or kept at a constant temperature (isothermal). The sample and reference material are simultaneously heated or cooled at a constant rate. Reaction or transition temperatures are measured as a function of the temperature difference between the sample and reference.

Application

- Determination of thermal stability, mass loss due to decomposition, oxidation or loss of volatiles, etc.
- It provides vital information of the materials regarding their endothermic and exothermic behaviour at high temperatures.
- This technique is applied to most of the polymers in evaluating the curing process of the thermoset materials as well as in determining the heat of melting and melting point of thermoplastic polymers, glass transition temperature (Tg.), endothermic & exothermic behaviour, etc.
- It provides information regarding the molecular weight and structural differences between similar materials, etc.

Gas Chromatography (GC)

Model: GC CERES 800 Plus, Make: Thermo Scientific

Principle

The organic compounds are separated due to differences in their partitioning behavior between the mobile gas phase and the stationary phase in the column. Mobile phases are generally inert gases such as helium, argon, or nitrogen. The injection port consists of a rubber septum through which a syringe needle is inserted to inject the sample. The injection port is maintained at a higher temperature than the boiling point of the least volatile component in the sample mixture. Separating components with a wide range of boiling points is accomplished by starting at a low oven temperature and increasing the temperature over time to elute the high-boiling point components.

Application

- Separation and identification of volatile materials, natural and synthetic polymers, paints and microbiological samples, etc.

- Pollutants like formaldehyde, carbon monoxide, benzene, DDT, etc.

Elemental Analyzer

Model: EURO EA 3000, Make: Euro-vector

Principle

In this technique the substance under study is combusted under oxygen stream in a furnace at high temperatures. The end products of the combustion would be mostly the oxides of the concerned elements in the form of gases. These are then separated and carried to the detector using inert gases like helium or argon. It is one of the few analytical techniques that give a clear quantitative measurement of the carbon, hydrogen, nitrogen and Sulphur. It finds applications in almost every field of chemistry like in the analysis of organics (especially to find out the molecular formula of a newly synthesized compound), polymers, pharmaceuticals, energy (fuels), environmental studies, etc.

Application

- To determine the elemental concentrations of carbon, hydrogen, nitrogen, oxygen, Sulphur, etc. in a given sample.
- It finds applications in the analysis of organics (especially to find out the molecular formula of a newly synthesized compound), polymers, pharmaceuticals, energy (fuels), environmental studies, etc.

Atomic Absorption Spectroscopy (AAS)

Model: ICE3300, Make: Thermo Scientific

Principle

The sample is vaporized by aspiration of solution into a flame or evaporation from electrically heated surface. At this condition where the individual atoms co-exist, a beam of light is passed through them. The atoms will absorb in the visible and ultraviolet region resulting in changes in electronic structure (excited state). So, the resultant light beam coming out of the sample will be missing the light in the corresponding wave length, which is a measure of the characteristics of the sample.

Application

- Applications of AAS determination of metals, only limitation on type of sample is that it must be capable of giving solution.
- Metallurgical and inorganic analysis for determination of alloys as Co, Cr, Mg, Mn, Pb and Zn, etc.
- Analysis of Ores eg. Ag, Co, Cu, Fe, etc.
- Biochemical analysis of various element or determination of Fe level in blood.
- Pollution analysis Agriculture industry, Analysis of Wines, Oils, Petrochemicals, Pharmaceuticals etc.

Spectroscopic Ellipsometer

Model: UVISEL 2, Make: Horiba Scientific

Principle

Ellipsometer measures the change of polarization upon reflection or transmission and compares it to a model. It consists of a laser, a polarizer and a quarter wave plates which provide a state of polarization. The beam is reflected off the layer of interest and then analysed with the analyzer. The operator changes the angle of the polarizer and analyzer until a minimal signal is detected. This minimum signal is detected if the light reflected by the sample is linearly polarized, while the analyzer is set so that only light with a polarization which is perpendicular to the incoming polarization is allowed to pass. In order to obtain linearly polarized light after reflection, the polarizer must provide an optical retardation between the two incoming polarizations which exactly compensates for the optical retardation caused by the polarization dependent reflections at each dielectric interface.

Since the amplitude of both polarizations was set to be equal, the ratio of the amplitudes after reflection equals the tangent of the angle of the analyzer with respect to the normal.

Application

Determination of dielectric properties of thin films and characterize composition, roughness, thickness, crystalline nature, doping concentration, electrical conductivity, etc.

High-Performance Liquid Chromatography (HPLC)

Model: FX-6, Make: Perkin Elmer

Principle

High performance Liquid Chromatography is a technique to separate mixtures of substances into their components on the basis of their molecular structure and molecular composition. This involves a stationary phase (a solid, or a liquid supported on a solid) and a mobile phase (a liquid or a gas). The mobile phase flows through the stationary phase and carries the components of the mixture with it. Sample components that display stronger interactions with the stationary phase will move more slowly through the column than components with weaker interactions. This difference in rates causes the separation of various components.

Application

- The HPLC instrument is used to identification and quantification of a compound.
- Chemistry and biochemistry research analyzing complex mixture.
- Purifying chemical compounds.
- It is also used in quality control to ensure the purity of raw material.
- It is used in analyzing air and water pollutant.
- Monitoring the pesticide levels in environment.
- Detection of phenolic compounds in drinking water.
- Measurement of Quality of soft drinks and water.
- Sugar analysis in fruit juices etc.

Brunauer Emmett Teller (BET) Surface Area Analyzer

Model: Surfer System-Standard, Make: Thermo Scientific

Principle

The nitrogen gas is adsorbed at cryogenic temperatures (liquid nitrogen). Based on the amount of gas adsorbed (adsorbate) at a given pressure, the BET equation is used to calculate the number of adsorbed gas molecules that is required to form a mono layer on the surface. The cross-sectional area of the gas molecule adsorbed and surface area can be calculated. The nitrogen gas is used for adsorption. The thermal conductivity detector (TCD) to measure the change in the concentration of an adsorbate/carrier gas mixture during adsorption or desorption process. It determines the surface area at single point and it can be enhanced for measuring multi point surface area and total pore volume analysis with different gas mixture. The software stores the data and computes the final surface area value. The adsorption and desorption curves are displayed on the screen.

Application

BET instrument is used for analysis of surface area and pore size distribution of sample, etc.

Ultraviolet - Visible Spectroscopy

Model: 2060+, Make: Analytical

Principle

A spectrophotometer is employed to measure the amount of light that a sample absorbs. The instrument operates by passing a beam of light through a sample and measuring the intensity of light reaching a detector. Ultraviolet-Visible spectroscopy is working based on the principle of Beer Lambert's law. It involves, absorbance is directly proportional to intensity of the colour and thickness of the medium.

Application

- Detection of functional groups
- Detection of extent of conjugation
- Identification of an unknown compound
- Determination of the purity of a substance, etc.

Fourier Transform Infrared Spectroscopy (FT-IR)

Model: SP-65, Make: Perkin Elmer

Principle

FTIR relies on the fact that the most molecules absorb light in the infra-red region of the electromagnetic spectrum. This absorption corresponds specifically to the bonds present in the molecule.

It involves the absorption of electromagnetic radiation in the infrared region of the spectrum which results in changes in the vibrational energy of molecule. Since, usually all molecules will be having vibrations in the form of stretching, bending, etc., the absorbed energy will be utilized in changing the energy levels associated with them.

The background emission spectrum of the IR source is first recorded, followed by the emission spectrum of the IR source with the sample in place. The ratio of the sample spectrum to the background spectrum is directly related to the sample's absorption spectrum. The resultant absorption spectrum from the bond natural vibration frequencies indicates the presence of various chemical bonds and functional groups present in the sample.

Application

- Identifying organic compounds which have polar chemical bonds (such as OH, NH, CH, etc.) with good charge separation.
- Use in the identification and structural analysis of organic compounds, natural products, polymers, etc.
- Use in the identification of functional group in a given organic compound, etc.

Polarimeter

Model: MCP 200, Make: Anton Par

Principle

Polarimeter measures the rotation of polarized light as it passes through an optically active fluid. The measured rotation can be used to calculate the value of solution concentrations; especially substances such as sugars, peptides and volatile oils. A Polarimeter consists of a polarized light source, an analyser, a graduated circle to measure the rotation angle, and sample tubes.

The polarized light passes through the sample tube and exhibits angular rotation to the left or right. On the side opposite the polarizer is the analyser. Using optics, visual fields are manually adjusted by the user to measure the optical rotation angle.

Application

- Polarimeter can be used to identify the isomer is present in a sample and also used to measure the ratio of enantiomers in solutions, etc.
- In industry it determines product purity by measuring specific rotation and optical rotation of amino acids, antibiotics, dextrose, steroids, sugars, cocaine, diuretics, etc.
- Polarimeter analyses optical rotation as a means of identifying and characterizing of biopolymers, natural polymers, synthetic polymers, etc.

Gel Permeation Chromatography (GPC)

Model: SPD-20aLC-20AD, Make: Shimadzu

Principle

GPC separates based on the size or hydrodynamic volume of the analytes. Separation occurs via the use of porous beads packed in a column. The smaller analytes can enter the pores more easily and therefore spend more time in these pores, increasing their retention time. Conversely, larger analytes spend little if any time in the pores and are eluted quickly. All columns have a range of molecular weights that can be separated. If an analyte is either too large or too small it will be either not retained or completely retained respectively.

Application

- GPC is used to determine the relative molecular weight of polymer samples, etc.
- To determine the poly dispersity index, characterize molecules such as synthetic polymers, etc.
- What GPC truly measures is the molecular volume and shape function as defined by the intrinsic viscosity

Dynamic Light Scattering (DLS)

Model: NPA152-31A-0000-000-90M, Make: Metrohm

Principle

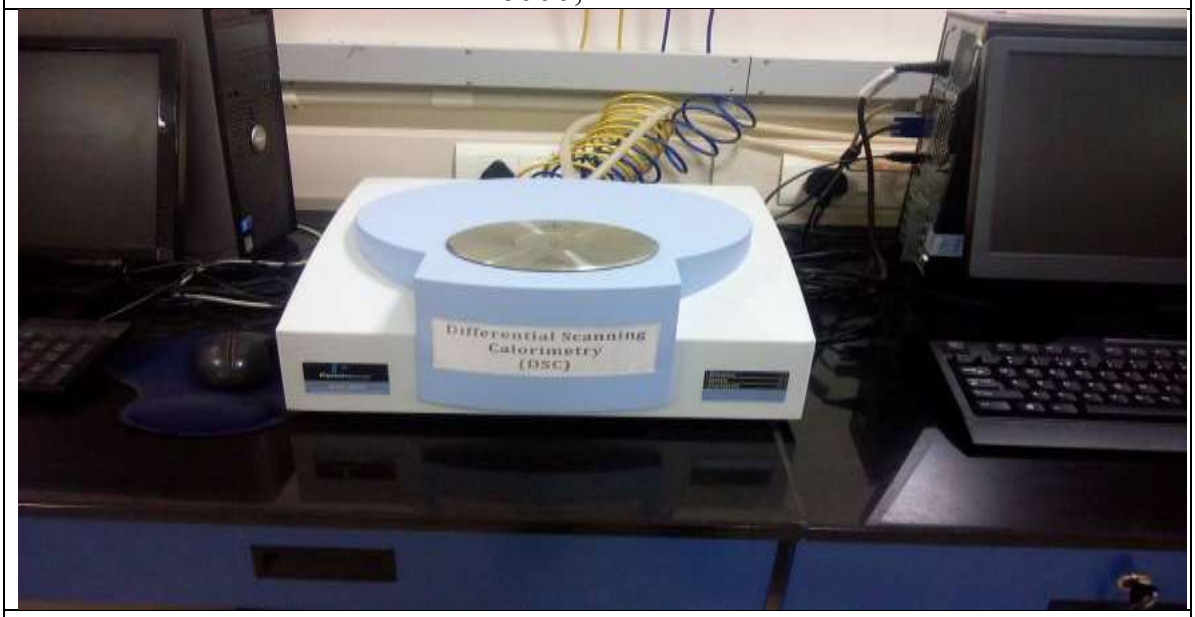
Dynamic light scattering based on principle of intensity fluctuation. DLS measures variation in scattered intensity with time at a fixed scattering angle, while static light scattering measures scattered intensity as a function of angle. When light hits small particles, the light scatters in all directions (Rayleigh scattering) as long as the particles are small compared to the wavelength (below 250 nm). If the light source is a laser, and thus is monochromatic and coherent, the scattering intensity fluctuates over time. This fluctuation is due to the fact that the small molecules in solutions are undergoing Brownian motion, and so the distance between the scatterers in the solution is constantly changing with time. This scattered light then undergoes either constructive or destructive interference by the surrounding particles, and within this intensity fluctuation, information is contained about the time scale of movement of the scatters.

Application

- DLS is used to characterize the particle size and determine zeta potential of various particles including nanoparticles, proteins, polymers, micelles, carbohydrates and, etc.
- The diffusion coefficient of the particles can be determined.
- Stability studies can be done conveniently using DLS.
- It is also suitable for diffusional studies of macromolecules, such as polymers and large biomolecules, etc.

Differential Scanning Calorimetry (DSC)

Model: DSC 6000, Make: Perkin Elmer



Principle

The DSC technique is a thermo analytical technique in which the difference in the amount of heat required to increase the temperature of a sample and reference are measured as a function of temperature. Both the sample and reference are maintained at nearly the same temperature throughout the experiment. Usually, the temperature program for a DSC analysis is designed such that the sample holder temperature increases linearly as a function of time. The reference sample should have a well-defined heat capacity over the range of temperatures to be scanned.

Application

- Differential scanning calorimetry can be used to measure a number of characteristic properties of a sample, etc.
- This technique is possible to observe fusion and crystallization properties as well as glass transition temperatures Tg, polymorphism, etc.
- It can also be used to study oxidation, as well as other chemical reactions, thermal stability and reaction kinetics of a material, etc.
- The melting process results in an endothermic peak in the curve, etc.
- This technique has ability to determine transition temperatures and enthalpies make DSC a valuable tool in producing phase diagrams for various chemical systems, etc.
- Determine the crystallization and melting temperatures and phase transition energies for inorganic compounds, etc.
- It is widely used in the pharmaceutical and polymer industries for studying curing, cross-linking of polymer molecules and provide a drug in the amorphous form, etc.

Scanning of Electron Chemical Microscope (SECM)

Model: CHI920D, Make: CH Instrument

Principle

Electric potential is operated through the ultra-microelectrodes (UME) tip in a bulk solution containing a redox-active couple. When a sufficiently negative potential is applied, (Fe3+) is reduced to at the UME tip, generating a diffusion-limited current. The steady-state current is governed by the flux of oxidized species in solution to the UME disc and is given by: $i_{T,\infty} = 4nFCDa$ (where iT, ∞ is the diffusion-limited current, n is the number of electrons transferred at the electrode tip (O + ne− → R), F is Faraday's constant, C is the concentration of the oxidized species in solution, D is the diffusion coefficient and a is the radius of the UME disc).

In order to probe a surface of interest, the tip is moved closer to the surface and changes in current are measured. There are two predominant modes of operation, which are feedback mode and collection-generation mode.

Application

- The SECM technique has been employed to investigation the topography and surface reactivity of solid-state materials, track the dissolution kinetics of ionic crystals in aqueous environments, screen electro catalytic prospects, elucidate enzymatic activities, and investigate dynamic transport across synthetic/natural membranes and other biophysical systems, etc.
- It is also focused on the solid/liquid interfaces and the characterization of typical solution-based electrochemical systems at higher spatial resolution and sensitivities than bulk electrochemical experiments, etc.
- This technique has been modified to explore the chemical transfer dynamics at liquid/liquid and liquid/gas interfaces, etc.

Matrix-Assisted Laser Desorption/Ionization (MALDI-TOF)

Model: Autoflex speed, Make: Bruker

Principle

The MALDI-TOF is a lenient ionization technique to allow ionization and vaporization of large nonvolatile biomolecules such as intact proteins. It generates mostly single-charged ions (z=1) so that the mass-to-charge ratio (m/z) of the analyte corresponds to its mass value. It is composed of three principal units. The first is the ion source that makes ionization possible and transfers sample molecule ions into a gas phase. The second unit is the mass analyzer that allows ion separation according to m/z. The last unit is a detection device for monitoring separated ions. Samples are prepared by mixing the analyte with a matrix made of small acid molecules that possesses a strong optical absorption in the range of the wavelength used by the laser device as DHB: 2,5-dihydroxybenzoic acid and CHCA: α-cyano-4-hydroxycinnamic acid are optimal matrices for the detection of lower mass ions. After co-crystallization of the sample and matrix, the latter absorbs energy from the laser, leading to the desorption and ionization of the analytes in the gas phase. Ions are then accelerated through an electrostatic field into the high vacuum flight tube until they reach the detector, with smaller ions traveling faster than larger ones.

Application

- MALDI-TOF is a vital tool in mass analysis of biomolecules and organic macromolecules.
- This technique used for the identification of micro-organisms such as bacteria or fungi and yeast.
- It serves as a method for determination of drug resistance of bacteria, especially to β-lactamases (Penicillin family).
- It is used for bacterial identification with high accuracy and applied to clinical samples such as blood, urine, cerebrospinal fluid and pleural fluid, peritoneal liquid and synovial fluid, etc.
- It is a simple and fast analytical technique to rapidly analyze the results of such syntheses as catenanes and rotaxanes, dendrimers and hyperbranched polymers and verify their results.
- In polymer chemistry MALDI can be used to determine the molar mass distribution.

Fluorescence-Activated Cell Sorting (FACS)

Model: BD- FACS Aria, Make: BD Biosciences

Principle

The FACS instrument is a particular form of flow cytometry that enables a mixture of different cells to be sorted one by one into one or more containers. The cells are sorted according to their specific light scattering and fluorescent characteristics. The process begins by placing the cells into a flask and forcing the cells to enter a small nozzle one at a time. The cells travel down the nozzle which is vibrated at an optimal frequency to produce drops at fixed distance from the nozzle. As the cells flow down the stream of liquid, they are scanned by a laser. Some of the laser light is scattered (red cone emanating from the red cell) by the cells and it is used to count the cells. In a complex mixture of biological cells, there may be more than one type of cell that has different antigenic and other markers on their surface. These markers can be tagged using fluorescent labels. As these cells pass a laser, the labels or fluorophores are excited and emit light at longer wavelengths than the laser beam. This can be picked up by detectors, which are then able to sort the cells according to their specific light scattering and fluorescent characteristics.

Application

- The FACS instrument used to measure the size of the cells, etc.
- It is extensively used in research for the detection of DNA damage, caspase cleavage and apoptosis. In marine biology, the auto fluorescent properties of photosynthetic plankton can be exploited by flow cytometry in order to characterize abundance and community structure.
- In protein engineering, flow cytometry is used in conjunction with yeast display and bacterial display to identify cell surface-displayed protein variants with desired properties.
- It has applications in a number of fields including molecular biology, pathology, immunology, plant biology and marine biology.
- It has broad application in medicine (especially in transplantation, hematology, tumor immunology and chemotherapy, prenatal diagnosis and genetics, etc.).

Fast Protein Liquid Chromatography (FPLC)

Model: Akta Purifier, Make: GE Healthcare Biosciences

Principle

The FPLC is a chromatographic separation technique. It contains of a programmable controller for developing and controlling automatic separation procedures, one or more pumps for liquid delivery, a mixer to ensure accurate and reproducible elution gradients, valves for sample injection and flow path control, one or more monitors for measuring chromatographic profile, a recorder for documenting chromatographic profile, a fraction collector and a chromatography rack for mounting the component. It is a micro purification chromatography system that recovers and purifies biologically active material present in a sample at very low concentrations. When the micro-concentration of sample inject to column, due to solvent interaction the sample introduced to the stationary phase of the analytical column and separation take places. After elution of protein as amino acids, they are further use in analytical and biochemical purposes. Biomolecules have various characteristics such as molecular weight, electric charge, and hydrophobicity.

Application

- The FPLC is the fast and simple chromatography technique for the separation of various proteins present in body fluids and other biomolecules.
- It is used for the separation of peptides, polynucleotides and used for the purification of synthetic oligonucleotides, plasmid DNA, rapid purification of RNA, etc.
- Used to measure the levels of tubular proteinuria: during tubular proteinuria, patient's kidney secrets more than 150 mg of urinary proteins daily.
- By using FPLC, the specific proteins can be isolated from the extracted proteins.
- Analysing Pancreatitis: In pancreatic juice, certain type of proteins can be determined by using FPLC, which lead to disease.

DNA Sequencer

Model: AB 3500, Make: Applied Bio-system

Principle

DNA sequence is useful in studying fundamental biological processes. The key principle of sanger method is used was the use of dideoxynucleoside triphosphates (ddNTPs) as DNA chain terminators. The chain termination (T) method requires a single-stranded DNA template, a DNA primer, a DNA polymerase, radioactively or fluorescently labelled nucleotides and modified nucleotides that T-DNA strand elongation. The DNA sample is divided into four separate sequencing reactions, containing all four of the standard deoxy nucleotides and DNA polymerase. The each reaction is added one of the four dideoxynucleoside which are chain terminating nucleotides, lacking a 3-OH group required for the formation of a phosphodiester bond between two nucleotides, thus terminating DNA strand extension and resulting in DNA fragments of varying length. Dye-T sequencing utilizes labelling of the chain terminator (T) ddNTPs, which permits sequencing in a single reaction, rather than four reactions as in the labelled-primer method. In dye-T sequencing, each of the four dideoxynucleotide chain T is labelled with fluorescent dyes, each of which with different wavelengths of fluorescence and emission. Owing to its greater expediency and speed, dye terminator sequencing is now the mainstay in automated sequencing.

Application

- DNA Sequencer helps to understand and comprehend the internal structure of genes in the DNA; it helps to understand which sequence codes for what kind of proteins, etc.
- With quicker, faster, transportable, low-cost sequencing, applications include: Individual sequencing leading to personalized medicine- gene therapy, rapid identification and characterization of pathogens, profiling tumor subtypes for diagnosis and prognosis, hypothesis testing for genotype/phenotype relationships, understanding B and T-cell receptor diversity to allow antibody selection, etc.

Confocal Laser Scanning Microscope (CLSM)

Model: LSM 780, Make: Zeiss

Principle

Confocal laser scanning microscopy (CLSM) is a technique for obtaining high-resolution optical images with depth selectivity. In this technique, a laser is used to provide the excitation light. The laser light reflects off a dichroic mirror. There, the laser hits two mirrors which are mounted on motors; these mirrors scan the laser across the sample. Dye in the sample fluoresces and the emitted light gets descanned by the same mirrors that are used to scan the excitation light from the laser. The emitted light passes through the dichroic and is focused onto the pinhole. The light that passes through the pinhole is measured by a detector, i.e., a photomultiplier tube. So, there never is a complete image of the sample at any given instant, only one point of the sample is observed. The detector is attached to a computer which builds up the image, one pixel at a time.

Application

- CLSM is widely used in numerous biological science disciplines from cell biology and genetics to microbiology and developmental biology. It is also used in quantum optics and nano-crystal imaging, etc.
- It is a powerful tool to get a high contrast image of a thin portion of the sample and useful for imaging in cellular environment, etc.
- To study the rate of diffusion of fluorescent molecules in fluid media (cytoplasm, membranes etc.) and inter-molecular interactions,
- This is used in the evaluation of various eye diseases, and is particularly useful for imaging, qualitative analysis, and quantification of endothelial cells of the cornea.
- It is used for localizing and identifying the presence of filamentary fungal elements in the corneal stroma in cases of keratomycosis, etc.
- In the pharmaceutical industry, used to follow for manufacturing process of thin film pharmaceutical forms, to control the quality and uniformity of the drug distribution, etc.

Real Time PCR (RT-PCR)

Model: AB 7500, Make: Applied Bio-system

Principle

The RT-PCR is carried out in a thermal cycler (TC) with the capacity to illuminate each sample with a beam of light of the one specified wavelength and detect the fluorescence emitted by the excited fluorophore. The TC is also able to rapidly heat and chill samples, thereby taking advantage of the physicochemical properties of the nucleic acids and DNA polymerase. The PCR process generally consists of a series of temperature changes that are repeated 25 to 50 times. These cycles normally consist of three stages: the first at around 95^0C allows the separation of the nucleic acids double chain, the second at a temperature of around 50 to 60^0C allows the binding of the primers with the DNA template; the third at between 68^0 to 72^0C facilitates the polymerization carried out by the DNA polymerase. Due to the small size of the fragments the last step is usually omitted in this type of PCR as the enzyme is able to increase their number during the change between the alignment stage and the denaturing stage. In addition, in four steps PCR the fluorescence is measured during short temperature phase lasting only a few seconds in each cycle with temperature for example 80^0C in order to reduce the signal caused by the presence of primer dimers when a non-specific dye is used.

Application

- The RT-PCR is mainly used to provide quantitative measurements of gene transcription and to determining how the genetic expression of a particular gene changes over time.
- It is used for the detection of specific gene sequences within a complex mixture.
- This is used in the fields of food safety, food spoilage, fermentation and for the microbial risk assessment of water quality (drinking and recreational waters) and in public health protection, etc.
- It is used for the determination of zygosity of transgenic animals used in research and frequently used for genotyping humans and human pathogens.
- An important application of RT-PCR is the measurement of RNA transcript levels to assess gene expression, etc.

Fluorescence Microscope

Model: BX 53, Make: Olympus

Principle

In a fluorescence microscope technique, a high intensity mercury arc lamp is used as the light source. It emits white light, which is passed through an 'exciter filter'. It allows only the blue component of the white light to pass through it and blocks out all other colour components. A dichroic mirror, which reflects blue light, but allows green light is used on the path of the blue light. The mirror is fixed at such an angle that the blue light is reflected downward to the specimen. The specimen is previously stained with a fluorescent dye. Certain portions of the specimen retain the dye, while others do not. The portions, which retain the fluorescent dye, absorb blue light and emit green light. The emitted green light goes upward and passes through the dichroic mirror. It reflects back blue light, if any, and allows only green light to pass through. Then, the light reaches a 'barrier filter'. It allows green light to pass to eye and blocks out any residual blue light from the specimen, which might not have been completely reflected by the dichroic mirror.

Application

- A fluorescence microscope is used to study properties of organic or inorganic substances.
- It is often used for imaging structural components of small specimens such as cells, conducting viability studies on cell populations, etc.
- Imaging the genetic material within a cell (DNA and RNA), etc.
- Viewing the specific cells within a larger population with this technique, etc.

Total Organic Carbon Analysis (TOC)

Model: Multi N/C 3100, Make: Analytikjena

Principle

Total organic carbon (TOC) is the amount of carbon found in an organic compound. It also refers to the amount of organic carbon in soil, or in a geological formation. A small amount of hydrochloric acid is added to sample to be less than PH3 and inorganic carbon is eliminated by bubbling, then it is poured into reaction tube with catalyst and converts to carbonic dioxide by means of sealed combustion oxidation. Carbon dioxide is measured density by infrared light analyzer; value of total organic carbon is calculated in terms of registered carbon dioxide in the data processing part and compares inspection lines of total organic carbon.

Application

- The TOC analyzer is used in various fields, environmental investigations of water (rivers, lakes, dams, sea area) and soils, etc.
- The management of tap water, effluent and ultrapure water, management of pharmaceutical water used in drug manufacturing processes, evaluation of cleaning efficiency, etc.
- Research and observation for organic pollution substances in natural environmental water such

Advanced Potentiostat

Model: 1.848.0010- Titrino plus, Make: Metrohm

Principle

The potentiostat controls the potential of the working electrode (relative to the reference electrode) The potentiostat controls the potential of the working electrode regardless of the characteristics of the cell The counter electrode is required for measuring the current only. The potentiostat controls the potential of the working electrode (relative to the reference electrode). The potentiostat controls the potential of the working electrode regardless of the characteristics of the cell. The counter electrode is required for measuring the current only.

In this technique, a potentiostat control the potential of the Counter Electrode (CE) against the Working Electrode (WE) so that the potential difference between the working electrode (WE) and the Reference Electrode (RE) is well defined, and correspond to the value specified by the user. The value specified by the user (i.e. applied potential or current) is accurately controlled, anytime during the measurement by using a negative feedback mechanism.

Application

The advanced potentiostat for use in a variety of application areas such as physical and electroanalytical electrochemistry, electrochemical corrosion, battery testing, fuel cell testing, and sensor development, battery testing, fuel cell testing, electrochemical noise, and electrochemical frequency modulation, etc.