

Centre for Advanced Instrumentation

HPLC

NIRMA UNIVERSITY



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Our Motto

तमसो मा ज्योतिर्गमय (Tamso Ma Jyotirgamaya) meaning "From ignorance, lead us to truth"

The motto of the University is taken from Brhadaranyaka Upanishad – I.III.28. The second line of the Pavamana Mantra explains how to lead the life towards knowledge from ignorance that obscures our mind in understanding the reality. As the only remedy from darkness is light, the only remedy from ignorance is knowledge.

Vision

Shaping a better future for mankind by developing effective and socially responsible individuals and organisations.

Mission

Nirma University emphasises the all-round development of its students. It aims at producing not only good professionals, but also good and worthy citizens of a great country, aiding in its overall progress and development. It endeavours to treat every student as an individual, to recognise their potential and to ensure that they receive the best preparation and training for achieving their career ambitions and life goals.



About Nirma University

Established in the year 2003, the Nirma University, Ahmedabad is a research-oriented, student-centric, multidisciplinary, not-for-profit state private university. Within a short period of its existence, it has emerged as a nationally renowned higher education institution. The University and its constituent institutes are highly ranked by different ranking agencies. Nirma University is duly recognised by the University Grants Commission (UGC) under Section 2 (f) of the UGC Act. The University is accredited by the National Assessment and Accreditation Council (NAAC). The University is a member of Association of Indian Universities (AIU) and the Association of Commonwealth Universities (ACU). Dr Karsanbhai K Patel, Chairman, Nirma Group of Industries and Nirma Education and Research Foundation (NERF), is the President of the Nirma University. Under his leadership, the University is expanding every passing year and moving from strength to strength.

Spread across the sprawling lush green 115-acres campus, the University has a host of institutes, departments and centres, including Institute of Technology, Institute of Management, Institute of Pharmacy, Institute of Science, Institute of Law, Institute of Architecture & Planning, Institute of Commerce, Department of Design, Faculty of Doctoral Studies and Research, Centre for Continuing Education, Centre for Entrepreneurship, Centre for Advanced Instrumentation and Centre for Robotics and Automation. These institutions offer numerous undergraduate, postgraduate and doctoral programmes. Apart from these, the University also offers several certificate and diploma programmes.



About Centre for Advanced Instrumentation

Nirma University is one of the leading University in Gujarat and also in India using innovations in Teaching Learning tools as well as establishing itself as important centre for Research. University is giving focused attention to promote research in diversified fields. To enhance and to strengthen research activities, it is necessary to have state of the art infrastructure and sophisticated analytical instrumentation facility which plays important role in interdisciplinary research activities.

This sophisticated instrumentation facility has high-end instruments which provides a platform to graduating students to develop their skills in handling latest instruments and is also helpful to masters' students, doctoral research scholars and faculty members who are involved in pursuing research activities. The facility will help them to carry out cutting edge interdisciplinary research of national and international importance. Centre facilitates procurement of high-end instruments as it requires huge investment, maintenance as well as expertise, which is otherwise difficult for individual researchers. Apart from this, centre offers its services to external researcher working in academia, research centres and industries. In line with the vision of the University to do overall development of its students, to serve society and to achieve new milestones in various fields of research and development, the University has decided to set up Centre for Advanced Instrumentation (CAI). This central facility will enhance the research activities and outcomes of University.

The main objectives of CAI are as follows:

- To promote research in Nirma University in advanced areas of science and technology.
- To provide advanced instrumentation facilities to research scholars and faculty members of Nirma University.
- To offer services to other academic institutes, research laboratories and industries as testing / consultancy services.
- To organise workshop / short term training programs for use / operation / applications of sophisticated instruments.
- To arrange specialised training programs for technicians working on instruments in academic institutions / small industries.
- To provide support to R&D centres and industries to design and develop their products.

List of Instruments

Chromatography

- 1. Gas Chromatography
- 2. High Pressure Liquid Chromatography
- 3. High Performance Thin Layer Chromatography
- 4. Supercritical Fluid Chromatography / Extraction

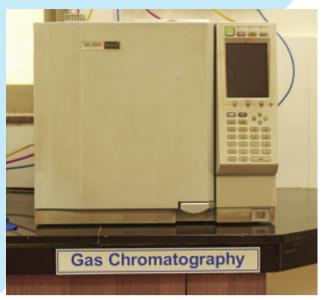
Spectroscopy and Characterization

- 5. Atomic Absorption Spectrophotometer
- 6. Differential Scanning Calorimeter
- 7. FT-IR Spectrometer with Microscope
- 8. High Pressure Homogenizer
- 9. Lyophillizer (Freeze Dryer)
- 10. Particle Size Analyzer
- 11. Total Organic Carbon
- 12. Spectrofluorometer
- 13. UV/VIS/NIR Spectrophotometer

Molecular Biology

- 14. Florescent Microscope
- 15. Flow Cytometer
- 16. Polymerase Chain Reactor (Real Time)





GC: Shimadzu 2010

Gas Chromatography (GC)

Principle

Gas Chromatography (GC) is a technique used in analytical chemistry for separating and analysing gaseous compounds. Applications of GC is not limited to test the purity of a given substance or separation of components in mixture. Inert gas is preferably used as mobile phase, e.g. helium or an unreactive gas nitrogen. The stationary phase is a layer of liquid or polymer on solid support inside a column which must be inert. The desired gaseous compounds interact with coated stationary phase on the walls and thus, each compound elutes at a different retention time. The column is located in an oven with high temperature, thus the gas can be controlled.

Applications

Thermal Conductivity Detector (TCD) – TCD detects changes in the thermal conductivity produced by effluent from column, with and without sample, and report difference. Most compounds have low thermal conductivity as compared to the carrier gases. Thus, when a sample elutes from the column, the reduction in thermal conductivity is observed and a detectable signal is produced. TCD is generally used for detection of gas samples.

Flame Ionisation Detector (FID) – FID is used for detection of inflammable organic compounds. It is widely used detector in gas chromatography as it produces linear response at very high order of magnitude. It is used to detect trace amount of compounds. The organic samples are burned in a flame to ionize the carbon atoms which emit free electrons and these electrons are measured as a current that is in direct proportion to the concentration of the hydrocarbons burned. Sensitivity of FID is at ppb level.

Electron Capture Detector (ECD) - ECD is most widely used for halogenated compounds and for electronabsorbing compounds. The ECD uses a radioactive beta particle (electron) emitter. The electrons coming out from electron emitter strike with molecules of career gas, resulting in many more free electrons. The electrons are accelerated so to reach to positively charged anode which generates current. As the sample travels into the detector by carrier gas, electron-absorbing sample molecules capture electrons and so reduce the current between anode and cathode. The concentration of analyte is considered as proportional with the degree of electron capture. ECD detectors are used to detect halogens, organometallic compounds, nitriles, nitro compounds, etc.

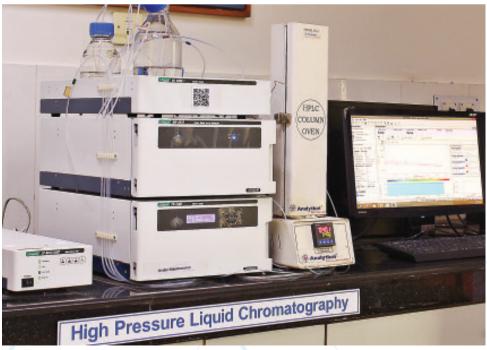
High Pressure Liquid Chromatography (HPLC)

Principle

HPLC (High Pressure Liquid Chromatography) is considered as one of the most important separation technique among all other techniques. It depends on interaction of analytes with two phases, viz. stationary and mobile phase, based upon its affinity and thereby separation. Based upon mechanism for separation, HPLC follows any of the following, but not limited to, mechanism, viz. adsorption, partition, ion exchange, ion pair or size exclusion mechanism.

Applications

HPLC technique is generally used for separation, identification, purification and quantification of a compound from a mixture. The other major applications of HPLC include determination of percentage purity of APIs and separation of impurities, stability study, tablet dissolution study, pharmaceutical quality control, etc. HPLC is majorly useful to Pharmaceutical and Chemical industries. It is also useful in the food industry for quality control and to compare with standards given by government.



HPLC: Jasco PU 2080 PLUS and Agilent G1312C



HPTLC: Camag

High Performance Thin Layer Chromatography (HPTLC)

Principle

High Performance Thin Layer Chromatography (HPTLC) is a superior form of thin-layer chromatography (TLC). Principle involves separation of samples by adsorption. Mobile phase moves by capillary action. Compounds move and get separated as per their affinities towards the stationary phase. Compounds with high affinity towards stationary phase, travel slow; while compounds having high affinity towards mobile phase and low affinity towards stationary phase travel fast, along with mobile phase. A number of improvements can be made in terms of automation in loading of sample, detection of spots and their quantification. These increase resolution and provide more accurate quantitative measurements. 2D chromatography with two different solvents increases spot capacity.

Applications

Major advantages of HPTLC include simplicity, low cost, multiple sample analysis at a time, high sample capacity, quick results, and opportunity for multiple detection. It has numerous advantages over other chromatographic methods including presentation of results as an image. HPTLC is the most extensively applied method in pharmaceutical industry, food and drug analysis, environmental analysis, clinical chemistry, forensic analysis, biochemistry and in cosmetic industries.



Supercritical Fluid Chromatography / Extraction (SFC/E)

Principle

A supercritical fluid has gaseous property (able to penetrate) and liquid property (able to dissolve materials). SFC is a hybrid of GC and LC. As mobile phase is in liquid form below its critical temperature and above its critical pressure, so the technique is liquid chromatography (LC). Mobile phase acts as a gas above its critical temperature and below its critical pressure, so the technique is called gas chromatography (GC). Use of carbon dioxide (CO2) or water in the form of a supercritical fluid provide substitute for other solvents in the food industry and medical supplies. In SFC, the sample passes with a supercritical fluid through a separating column where the mixture is divided into unique bands. This happens based on the intensity of interaction between the analytes and the stationary phase. As these bands leave the column, their identities and quantities are determined by a detector.

Applications

SFC is useful and has wide varieties of applications which include natural products, drugs, food, pesticides, herbicides, surfactants, polymers and polymer additives, fossils fuels, petroleum, explosives and propellants, etc.



SFC: Jasco



AAS: Shimadzu AA-6300

Atomic Absorption Spectrophotometer (AAS)

Principle

Atomic Absorption Spectroscopy (AAS) is a spectro-analytical method for the quantitative determination of chemical elements (metals) at very low concentrations of parts per million (ppm) or parts per billion (ppb) using the absorption of optical radiation (light) by free atoms in the gaseous state. AAS is based on absorption of light/radiation by free metallic ions. At the high temperature, the sample is broken down into atoms and it is the concentration of these atoms that is measured. The total amount of absorption depends upon the number of free ions and degree to which the free ions absorb the radiation. In analytical chemistry, the technique is used for determining the concentration of a particular element in a sample to be analyzed.

Applications

AAS can be used to determine different elements in solution, or directly in solid samples via electro thermal vaporization and is used in agricultural, chemical, environmental, pharmacological, biophysical, and archaeological and toxicological research. AAS is used to analyse soil and plants for necessary minerals required for growth. AAS has many uses in different areas of chemistry such as clinical analysis of metals in biological fluids and tissues such as whole blood, plasma, urine, saliva, brain tissue, liver, hair, muscle tissue, semen, in some pharmaceutical manufacturing processes, minute quantities of a catalyst that remain in the final drug product, and analysing water for its metal content.

Differential Scanning Calorimeter (DSC)

Principle

Differential Scanning Calorimetry (DSC) is the most frequently used thermal analysis technique. It measures the temperatures and heat flows associated with transitions in sample and reference materials as a function of time and temperature in a controlled atmosphere. Both the sample and reference are maintained at nearly the same temperature throughout the experiment. DSC identifies and characterizes materials. A small amount of sample is heated, and if any kind of transition takes place during this process, it will lead to a slight difference between the sample and a reference sample temperature, i.e. differential scanning calorimetry measures the amount of energy (heat) absorbed or released by a sample as it is heated, cooled, or held at a constant temperature. Differential scanning calorimetry is fast, very sensitive and easy to use.

Applications

Major application of DSC is to check the compatibility of drug with excipient. DSC analysis measures endothermic or exothermic reaction by a sample when it is heated, providing quantitative and qualitative data on heat absorption and heat evolution processes. DSC is used widely for examining polymeric materials to determine their thermal transitions. It is also used in the study of liquid crystals. DSC analysis is used to measure melting temperature, heat of fusion, latent heat of melting, glass transition temperature, crystalline phase transition temperature and energy, precipitation energy and temperature, denaturation temperatures, etc. It is also used to measure specific heat capacity, heat flow rate, thermal behaviour and its derivatization such as the heat transformation or any chemical change in the sample.





FTIR: Jasco FTIR 6100

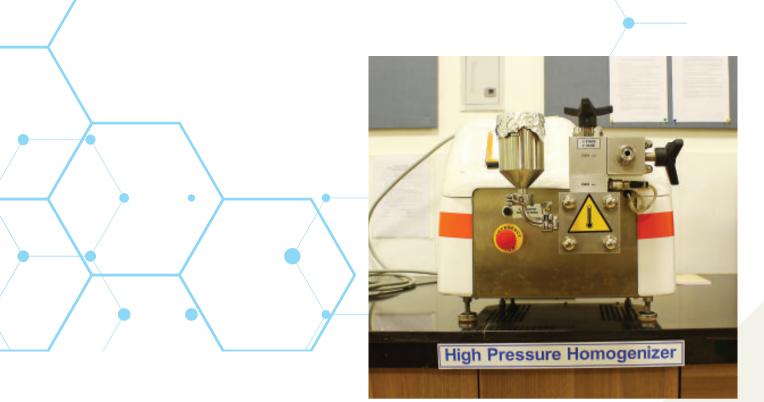
Fourier Transform Infra-Red (FTIR) Spectrometer with Microscope

Principle

Fourier Transform Infra-Red Spectroscopy (FTIR) is a technique used to obtain an infrared spectrum of absorption or emission of a solid or liquid. In FTIR analyses, Infrared light from the light source passes through a Michelson interferometer along the optical path. The light beam splits into two by the beam splitter is reflected from the moving mirror and fixed mirror, before being recombined by the beam splitter. As the moving mirror makes reciprocating movements, the optical path difference to the fixed mirror changes in such a way that the phase difference changes with time. The light beams are recombined in the Michelson interferometer to produce interference light. The intensity of the interference light is recorded in an interferogram, with the optical path difference recorded along the horizontal axis.

Applications

FTIR spectra reveal the composition of solids and liquids. The most common use is in the identification of unknown materials. Apart from applications in pharmaceutical and chemical industries, it has applications in food sciences, forensic sciences, environmental sciences, chemical sciences, polymer and plastic industries, etc. It is used for structural elucidation in basic drug research; used in formulation development and validation; quality control processes for incoming and outgoing materials, etc. FTIR microscope can be combined with a sample heating system, which allow changes in molecules to be measured for their thermo physical properties with image observation. The heating system uniformly heats or cools the measurement area in the IR microscope, assuring high accuracy measurement. Temperature and measurement conditions are software controlled with simultaneous image scanning and IR measurement.



HPH: Panda Plus-2000

High Pressure Homogenizer (HPH)

Principle

High pressure homogenisation is a mechanical process which involves forcing fluid through a narrow homogenising nozzle at high pressure. The liquid is subjected to very high shear stress which causes the formation of fine emulsion droplets. The smaller the nozzle aperture and the higher the pressure, the smaller the droplets that are produced, with the aim being to reduce particles and droplets from micron to nanometer sizes. In the piston-gap homogenizer, the macro-suspension coming from the sample container is forced to pass through a tiny gap; particle diminution is affected by shear force, cavitation, and impaction.

Applications

High pressure homogenizer applications require the most efficient fluid processing equipment for particle and droplet size reduction and cell disruption. Major applications of homogenizer include high pressure pasteurization, particle size reduction, micro/nano emulsions, dispersions and cell disruption. Major areas of applications include pharmaceutical drug development, manufacturing, milk industry, food and beverage processing industry.



Lyophillizer (Freeze Dryer)

Principle

The principle involved in lyophilisation i.e. freeze drying is a phenomenon called sublimation, where water passes directly from solid state (ice) to the vapour state without passing through the liquid state. Sublimation of water can take place at pressure and temperature below triple point. The material to be dried is first frozen and then subjected under a high vacuum to heat, so that frozen liquid sublimes leaving only solid, dried components of the original liquid. The concentration gradient of water vapour between the drying front and condenser, is the driving force for removal of water during lyophilisation.



Lyophillizer: IIshin Bio Base TFD 8503

Applications

The major industries where lyophilisation is useful include, but not limited to, pharmaceutical and biotechnological industries, food industry, technological industry and also useful to conserve special bacterial strains.

Particle Size Analyzer (PSA)

Principle

The instrument uses principle of dynamic light scattering to measure particle size. It is a measurement of fluctuation in scattered light intensity with time. Fluctuation mainly observes due to random brownian movement of nanoparticles. The statistical behaviour of these fluctuations occurs in scattered intensity can be related to the diffusion of the particles. Larger particles diffuse more slowly than small particles, thus one can readily relate particle size to measured fluctuation in light scattering



PSA: Horiba SZ-100Z

intensity. It also measures zeta potential. Nanoparticles or colloidal particles have a surface charge in suspension. When an electrical field is applied, the particles move due to the interaction between the charged particles and the applied field. The direction and velocity of motion is a function of particle charge, suspending medium and electric field strength.

Applications

Particle size analysis is used to characterise the size distribution of particles in a given sample. Particle size analysis can be applied to solid materials, suspensions, emulsions and even aerosols. It is also useful for measurement of zeta potential of nanoparticulate system. Some industries and product types where particle sizing is used includes pharmaceuticals, building materials, paints and coatings, food and beverages and aerosols, etc.

Total Organic Carbon (TOC)

Principle

Total Organic Carbon (TOC) measurement is commonly used to determine the degree of organic contamination in water. TOC is an indirect measure of organic molecules present in water and measured as carbon. TOC is a highly sensitive, non-specific measurement of all organics present in a sample. Organic molecules are introduced into the water from the source water, from purification, and from distribution system materials. TOC is measured for both process control purposes and to satisfy regulatory requirements. The high temperature combustion catalytic oxidation method achieves total combustion of samples by heating them in an oxygen-rich environment. The carbon dioxide generated by oxidation is detected using an infrared gas analyser (NDIR).

Applications

TOC is very important in detecting contaminants in drinking water, cooling water, water used in semiconductor manufacturing, and water for pharmaceutical use. TOC detection is an important measurement because of the effects it may have on the environment, human health, and manufacturing processes. It can be used to regulate the organic chemical discharge to the environment in a manufacturing plant. In addition, low TOC can confirm the absence of potentially harmful organic chemicals in water used to manufacture pharmaceutical products. TOC is also of interest in the field of potable water purification due to by-products of disinfection. Inorganic carbon poses little to no threat. Total Organic Carbon analyser is used for environmental analysis, pharmaceutical and chemical industries, determination of carbon dioxide in food products, etc.



Spectrofluorometer

Principle

The Spectrofluorometer works with the principle of fluorescence spectroscopy. This technique correlates fluorescent properties of some compounds in order to provide information regarding their concentration and chemical environment in a sample. The emission is observed either at a single wavelength or a scan is performed to record the intensity versus wavelength. The system is having high quality optical system and also has high sensitivity.

Applications

The Spectrofluorometer is used to meet demanding needs of laboratories in application areas like biochemistry performing kinetics, stopped flow, titration and anisotropy experiments. This technique has applications in the field of pharmaceutical industry, has environmental significance, geology applications, analytical chemistry as well as biochemistry applications.



Spectrofluorometer: Jasco FP-6500

UV/VIS/NIR Spectrophotometer

Principle

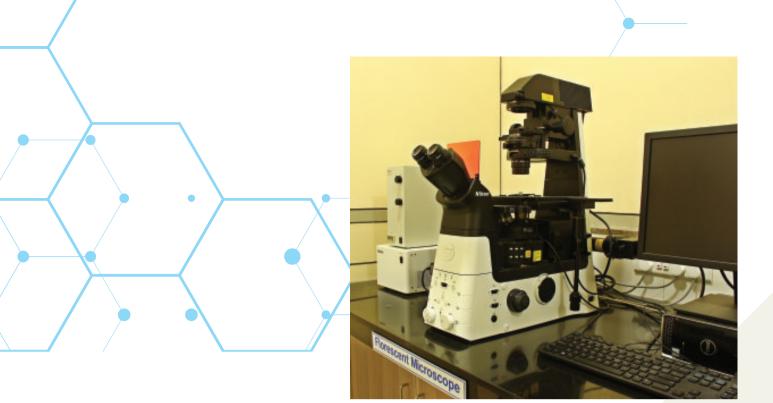
A UV/VIS/NIR Spectrophotometer measures the reflection or absorbance characteristics of a sample. It measures in the range covering UV, Visible and Near IR, from 200 to 3000 nm wavelength. Here, absorption is used to characterise materials. Absorption of radiation may occur in a transmission or reflection mode. A spectrum is generated with a graph of absorption versus wavelength. The spectra is used to determine the spectral response of colour sample in the visible region or the sample's ultraviolet or near infra-red filtering characteristics or the difference in appearance between two pigments when viewed under different light sources.



UV/VIS/NIR: Jasco V-570

Applications

A UV/VIS/NIR Spectrophotometer is useful for the measurement of absorbance of liquid in the range of 200 to 2500 nm. In addition to measuring liquids, it is used to measure the transmittance and reflectance of solid samples. This technique gives basic information about λ_{max} of unknown sample as well as it gives reflection spectra which is useful to get preliminary information about sample.



Florescent Microscope: Nikon Eclipse Ti2

Florescent Microscope

Principle

Many substances absorb light. However some of them, after absorbing light of a particular wavelength and energy, emit light of a longer wavelength and lesser energy. Such substances are called 'florescent substances'. Application of this phenomenon is the base of fluorescence microscope. Microbes are stained with a florescent dye and then illuminated with blue light. The dye absorbs blue light and emits green light. The specimen is previously stained with a fluorescent dye, such as acridine orange, acridine yellow, acriflavine, thioflavine S, thioflavine T or titan yellow G, etc. 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. Thus, the eye perceives the stained portions of the specimen as glowing green object against a jet-black background, whereas the unstained portions of the specimen remain invisible. Fluorescence microscopy requires a very powerful light source such as a xenon or mercury arch lamp.

Applications

Fluorescence microscopy is widely used in biology and medicine, as well as in other fields. Fluorescence techniques can be applied to all kinds of material, but generally, fluorescence microscopy is reserved for applications which require high sensitivity, i.e., to examine substances present in low concentrations. Fluorescence microscopy is also useful to detect particles below the resolution of a light microscope, and in histochemistry to visualize substances which cannot be seen by conventional microscopy. Biological material is commonly stained in same manner with a fluorescent stain and then are observed under fluorescence microscopy.

Flow Cytometer

Principle

Flow cytometry means measuring properties of cells when in motion. The basic principle of flow cytometry is the passage of cells in single line in front of a laser so they can be detected, counted and sorted. Cell components are fluorescently labelled and then excited by the laser to emit light at varying wavelengths. The fluorescence can then be measured to determine the amount and type of cells present in a sample. Up to thousands of particles per second can be analysed as they pass through the liquid stream. A beam of laser light is directed at a hydrodynamically focused stream of fluid that carries the cells.

Applications

The Attune NxT Flow Cytometer has multiple applications in the area of Immunophenotyping. It helps in designing a T-cell backbone panel and validated multicolour T cell panel. It helps in multicolour immunophenotyping analysis of stained human whole blood using a no-lyse, no-wash protocol, with no compensation. It also helps in multiple parameter immunophenotyping of human lysed whole blood which identifies multiple T-cell subsets and myeloid cells, multi-parameter immunophenotyping of human lysed whole blood whole blood identifies B cells, NK cells, multiple T-cell subsets and myeloid cells, multi-parameter analysis of murine regulatory T-cells and dendritic cells and used to detect murine regulatory T-cells. It also helps to detect platelets in whole blood; no-wash, no-lyse detection of leukocytes in human whole blood and no-wash, no-lyse detection of phagocytic cells via pHrodo BioParticles functional assay in human whole blood. It also has application for rapid and accurate analysis of nuclear DNA content in plants and used for detection of fluorescent proteins. It is also used in Flow cytometry analysis of transcription factor expression during differentiation of hPSC-derived cardiomyocytes.



Flow Cytometer: Attune NxT



Real Time PCR (qPCR): QuantStudio 3

Polymerase Chain Reactor (Real time)

Principle

The polymerase chain reaction (PCR) is a laboratory technique for DNA replication by selective amplification of target DNA. PCR uses the smallest sample of DNA to be cloned and amplify it to millions of copies in just few hours. The PCR involves a primer mediated enzymatic amplification of DNA. PCR is based on using ability of DNA polymerase to synthesize new strand of DNA complementary to the offered template strand. Primer is needed because DNA polymerase can add a nucleotide only onto a pre-existing 3'-OH group to add the first nucleotide. DNA polymerase then elongate its 3 end by adding more nucleotides to generate an extended region of double stranded DNA.

Applications

Major applications of PCR in various fields include medical applications like genetic testing, detection of infectious diseases causing genes, alteration to oncogenes, sensitive tissue typing, in gene therapy; forensic applications include genetic fingerprinting in crime scenes, paternity testing, etc. Real-time PCR (or qPCR) has very wide applications in biological science which include agricultural and food industries, gene expression analysis, the diagnosis of infectious disease and human genetic testing.







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