Review Article

HYPHENATED GAS CHROMATOGRAPHY

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ABSTRACT

Hyphenated gas chromatography is not only coupling of GC to detector but it is coupling of the GC to automated sample preparation techniques. Various detectors include mass spectrometer and infrared spectrometers, whereas automated sample preparation techniques include solid-phase micro extraction (SPME), large-volume injection (LVI), purge and trap (PT), headspace (HS). The other gas chromatographic approach is multidimensional gas chromatography (MDGC) which consists of more than one column with different selectivity. The union of the automated sample preparation techniques with MDGC and detector allows quantitative and qualitative analysis of a wide variety of sample matrices for analytes at parts per billion (ppb) concentrations. Hyphenated gas chromatography is the versatile tool in pharmaceutical sciences with wide range of applications such as determination of volatile oil, separation of enantiomeric volatile components in essential oils using SPME/GC, determination of trace components in water using LVI. It improves precision and provides for more effective use of laboratory personnel, particularly for industrial routine analysis. It also helps to process the high number of samples, necessary to get the many data for method validation to certify an analytical method.

Keywords: Hyphenated gas chromatography, gas chromatography, infrared spectrometers.

INTRODUCTION

Hyphenated gas chromatography refers to not only the coupling of a GC to information-rich detectors but also the coupling of gas chromatograph to automated sample preparation systems. The term "hyphenation" was first coined by Hirschfeld in 1980.Examples of information of rich detectors include mass and infrared spectrometers, whereas automated sample preparation systems include static headspace (HS), dynamic headspace (PT), large volume injection (LVI) and solid -phase microextraction (SPME).Hyphenation of gas chromatographic approaches also include coupling of two gas chromatographs and is commonly referred to as multidimensional gas chromatography (MDGC). Wedding of advanced sample preparation techniques with potent information-rich detectors in presence of capillary GC provides for a sensitive analytical approach for the analysis of volatile and semi-volatile compounds. The coupling of MDGC with IR and MS can provide qualitative as well as quantitative data of target compounds similarly the union of LVI with MDGC and MS can lowers the detection limits from parts per billion to parts per trillion. This does not mean that every hyphenated technique worth using or even considering hence before going for the hyphenation of GC with other techniques we have to check whether this hyphenation helps to increase the sensitivity, separating power, flexibility of GC or not. Hyphenation of gas chromatography can be carried out by two ways i.e. Pre-gas chromatograph Automated On-line sample preparation techniques and another one is Post-gas chromatograph sample analysis techniques.

A) PRE-GAS CHROMATOGRAPH AUTOMATED ON-LINE SAMPLE PREPARATION TECHNIQUES

Automated on-line sample preparation techniques in chromatographic analysis are generally used because the concentration of the analyte of interest is below the detection of the analytical instrument. Sometimes analyte must be removed from sample matrix because the introduction of sample matrix directly into the chromatographic technique is not compatible.

1. HEADSPACE/GAS CHROMATOGRAPHY[1, 2]

Complex samples like biological samples, natural products extract etc. for these samples, headspace sampling is the fastest method for analyzing volatile organic compounds. A headspace is the space which is present above the surface of sample matrix **(Figure 1)**.
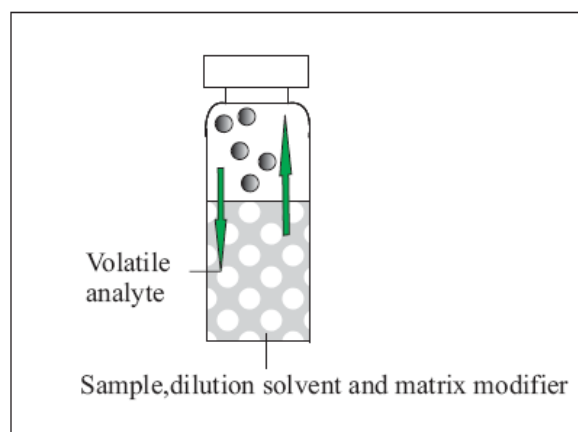

Figure 1: Description of headspace present above the sample matrix.

Volatile components from complex sample mixtures can be extracted from non-volatile sample components and isolated in the headspace or vapor portion of a sample vial. An aliquot of the vapor in the headspace is injected to a GC system for separation of the volatile components. Headspace analyses using GC can be subdivided into two main categories

a) Static headspace/Gas Chromatography

Static headspace analysis is based on the theory that equilibrium between a condensed phase and a gaseous phase can be reproducibly maintained for the analytes of interest and that the gaseous phase containing the analytes can be sampled reproducibly.

When coupling the static HS sample preparation system approach to GC, several parameters must be carefully attended to. The two units i.e. static HS unit and the GC are connected via a thermally controlled inert transfer line that serves as a source of carrier gas for the GC and mode for the transfer of the analyte from the static HS unit to the GC column. Flow control and the purity of the carrier gas are essential. Flow is normally controlled by flow control valves. Static HS experiments carried out in thermally stable sealed environment, with a leak-free thermally stable instrument setup. All commercially available static HS systems provided with a accurate and durable thermocouples.

This technique operates by initially thermostatting the sample in an incubation oven at a given temperature and for a given time until it has reached a state of equilibrium

(**Figure 2, Step 1**). Once the sample has reached equilibrium, pressurization of injection is carried out (**Figure 2, Step 2**), and aliquot is taken from the headspace and it is injected into the GC (**Figure 2, Step 3**).
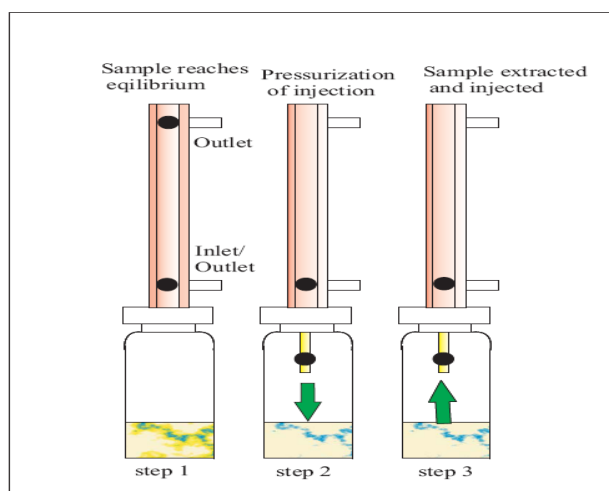

Figure 2: Working of static headspace technique.

The parameters that are manipulated in order to optimize the procedure include times, temperatures and pressures associated with sample heating, sample equilibration, loop fill, loop equilibration and sample transfer.

Limitations

Static headspace analysis approach will not be very successful when the analytes of interest possess very low vapor pressures because very little of the analyte will be found in the gaseous phase but elevated temperatures, changes in pH, and the presence of additional electrolytes can in certain cases be employed to increase the analyte vapor pressures.

b) Dynamic Headspace (Purge and Trap) / Gas Chromatography

Dynamic headspace analysis based on the principle is that the change in mass of a volatile or semivolatile analyte with time can be expressed in terms of volumetric flow rate of stripping inert gas. It means it is possible to take out volatile and semivolatile compounds from solid or liquid matrices by passing an inert gas over or though them and that the amount of material take out can be related to the inert gas passed through the matrices. This physical behavior with the capacity to trap the stripped materials on an inert trapping material affords an excellent opportunity for sample enrichment.

The liquid and solid matrix is essentially nonvolatile and the distribution constant of a analyte is not dependent on the analyte concentration. The issues associated with coupling a PT unit to a GC are very similar to that of static HS unit to a GC. The two units are connected via thermally controlled inert transfer line that can serve as a source of carrier gas for the GC as well as a mode of transfer for the analytes from the PT unit to the GC. A leak-free environment is important with PT as well as thermal stability. In PT techniques the system is not sealed as in the static approach, thus the headspace above the sample is not essential to come to the equilibrium. Instead, the sample is placed into a chamber, at a pre-selected temperature, that is sparged with carrier gas at a specified rate and time (**Figure 3**). The swipping carrier gas removes the analytes from the matrix and transports them under a thermally controlled environment to a trap. The trap is usually allowed to come close to room temperature before to transfer of the analyte. After a sometime the spargin is stopped. The trapping material which we are using is based on the type of the analytes of interest. After the analyte have been trapped a multiple port valve is activated and the trap is heated and backflushed this leads to the desorbing the analytes from the sorbent material and transferring into GC. Sometimes a cryofocusing is use to capture the analyte in a narrow band at the head of the column.
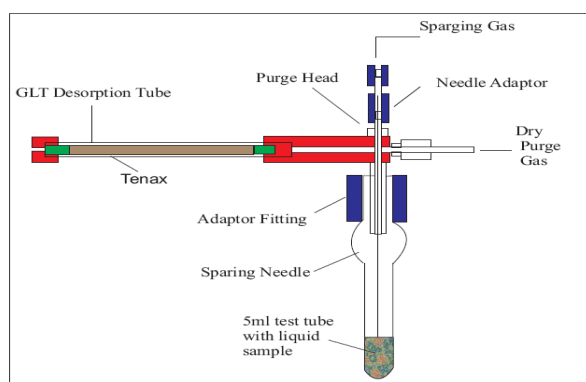
Figure 3: Diagram of purge-trap system.

The parameters that may be manipulated in order to optimize the procedure include sample purge time, carrier gas purge flow rate and sample temperature. Different materials which are used for the trapping of analyte are as follows,

A)   Polymers
-          Tenax
-          Polystyrene
-          Polyurethane foams
B)   Carbon
-          Charcoal
-          Carbon sieves
-          Graphitized carbon black
C)   Silica
D)   Alumina

SOME RECENT DEVELOPMENTS IN HEADSPACE GAS CHROMATOGRAPHY:

A)   Full evaporation headspace technique:

The full evaporation technique was one of the oldest HSGC techniques. It utilizes the headspace sampler as an evaporator rather than an enclosed static vapor liquid equilibrium space. Sample vial contain very small amount of sample to achieve a complete evaporation or transfer of analyte from condensed phase into a vapor phase in the headspace of the vial. Hence vapor liquid equilibrium and sample pretreatment are not required. To achieve good result it is essential to get near-complete conversion of analyte into the vapor phase. Example, Recent application of FE HSGC is to determination of residual monomer in polymer latex. Similarly the measurement of methyl mercaptan (MM) and dimethyl sulphide (DMS) in pulp industry is also possible by the use of FE HSGC.

B)   Phase reaction conversion headspace technique:

The phase reaction conversion (PRC) headspace gas chromatography technique is based on the conversion of a fixed percentage, including complete conversion of an unknown nonvolatile analyte in a liquid or solid state into a gaseous state through chemical reactions. The analyte is then analyzed by the HSGC. This technique is very useful for the analysis of nonvolatile compounds which is either in solid or in liquid state. Example, PRC HSGC technique is used to determine the carboxyl groups in wood fibers by converting carboxyl groups into carbon dioxide using bicarbonate and it is also useful for the determination of hydroxylamine in pharmaceutical preparations by converting hydroxylamine into nitrous oxide by using $FeCl_3$ in a buffer solution of sodium acetate.

C)   Multiple headspace extraction for process kinetics study:

Multiple headspace extraction (MHE) is similar to dynamic headspace gas extraction but it is carried out in steps for the kinetic study.

Applications

The chemical composition of essential oils is related to a variety of factors including age, genotype, and geographical growing locations. In an application of static and dynamic HS techniques quantitative and qualitative differences in the distribution of components in an essential oil can be analysed. Also used for quantification of volatile compounds in goat milk Jack cheese.[3]

2.   SOLID PHASE MICROEXTRACTION[2]

Solid phase microextraction is fundamentally a solvent free sample preparation technique having qualitative and quantitative potential. A relatively thin film extracting phase of very small volume, less than 1µL, is firmly coated and bound to a fused silica fiber **(Figure 4)** which in turn can be exposed to a sample matrix. Room air, aqueous solution or organic solvents acts as an sample matrix. The extracting phase bound to the fiber is very similar to the phases used in capillary GC.
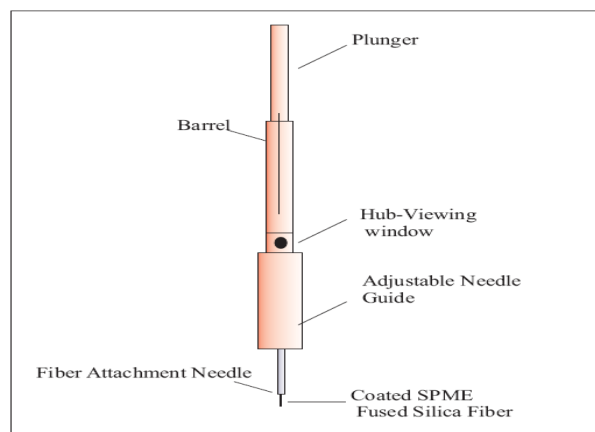

Figure 4: Diagram of SPME fiber.

Available SPME Fibers, by Film Type

A) Absorption Fibers

-Polydimethylsiloxane(PDMS) 7, 30, and 100µm

-Polyacrylate(PA)

-Polyethyleneglycol (PEG)

B) Adsorption fibers (with particles)

-Carboxen-polydimethylsiloxane(CAR-PDMS)

-Polydimethylsiloxane-divinylbenzene(PDMS-DVB)

-Divinylbenzene/Carboxen-Polydimethylsiloxane(DVB-CAR-PDMS)

-Carbowax-divinylbenzene(CW-DVB)

The most convenient configuration of SPME technology is shown in **(Figure 5).** The thin silica fiber is enclosed in tube when it is not in used. Exposure of the fiber to the sample matrix is effected by sliding the fiber outside of the tube into the matrix. This exposure can be performed by manually or automatically. At the time of sampling silica fiber is exposed to the sample matrix of interest due to this extracting phase which is coated on to the silica fiber is comes in contact with the analyte. The extracting phase has an ability to extract the analyte from the sample matrix by adsorption or absorption. The partition coefficient of the analyte of interest between the sample matrix and the fiber coating material governs the amount of analyte extracted by the fiber coating. SPME is useful for the quantitative as well as qualitative analyses by employing a variety of coating differ in polarity.
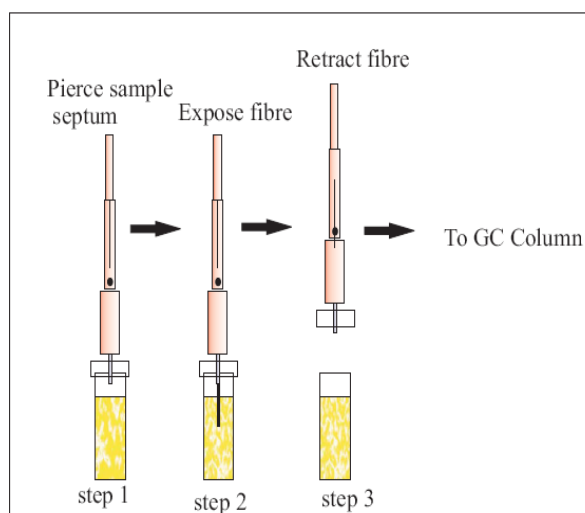

Figure 5: A typical sequence of events in SPME process.

Several extraction parameters have been documented as having meaningful impacts on qualitative and quantitative aspects of the amount of analyte extracted by the fiber.

A) Fiber type:-

Selection of the fiber type can be governed by the age-old adage that ''like dissolves like''. For example, if polar analytes such as flavor compounds are of interest, then a polar material such as CWDVB would be a logical first choice.

B) Temperature:-

Combination of temperature with exposure location can be used to an advantage. For example, in case of headspace sampling above the sample matrix increased in temperature helps to reduce the complexity of the extracted material.

C)   pH:-

Modification of pH alters the nature of the species in solution which leads to changes in extraction performance of fiber.

D)   Ionic strength:-

Addition of salts in the solution such as sodium chloride in solution can bring about the salting-out phenomenon which leads to improvement in the performance of fiber.

E)   Agitation:-

Stirring of liquid cases or fiber vibration improves the performance of the SPME fiber.

F)   Fiber conditioning:-

Proper conditioning of SPME fiber is important. Failure to properly condition an SPME fiber can results in unacceptable accuracy and precision.

The successful interfacing of SPME with GC helps to produce the hyphenated technique SPME/GC. The analyte captured on the nonvolatile thin film can be efficiently thermally desorbed in a reproducible fashion.For the desorption high carrier gas linear velocity is coupled with an increased temperature. Normal desorption times of few seconds at 200^0C should be sufficient for most volatile and nonvolatile analytes.

Applications

The enantiomeric distributions of volatile components in essential oils can be accurately and precisely determined using SPME/GC. Employing a PDMS SPME fiber of 7µm film thickness, a GC fitted with column capable of separating optical isomers using MS an as detector. The accurate results were obtained without establishing equilibrium conditions, because sampling parameters like temperature, fiber exposure time and sample HS volume were carefully controlled.

Solid phase microextraction combined with GC and MS is also use for the characterization of cheese aroma compounds [4]. In addition to well characterized cheese compounds, the fibres successfully also adsorbed many other compounds such as sulfur, pyrazines, furanones compounds. It is also used for analysis of club drug in urine sample[5]. Which are also analyzed by using SPME with GC and MS.

3.   **LARGE   VOLUME   INJECTION/GAS CHROMATOGRAPHY**[6]

The injection systems which are used in gas chromatographic technique are of different types such as split, splitless and cold on-column injection system. The main purpose of injection systems is only to deliver the sample into the column without degrading the maximum separation efficiency of the column. Split injection is normally employed for component analysis where the concentration of an analyte should be greater than 50ppm. The splitless injection technique is widely used for trace component analysis in the range from 0.1ppm to

200ppm. In cold on-column injection system sample is directly deposited on to the front of the column and has concentration ranging between 0.1ppm to 200ppm. The cold on-column technique is better than split and splitless injection system because in this system flash vaporization step is not present and similarly sample is not subjected to the high temperatures.

Main limitation of these three most widely used common injection techniques is that the sample volume is limited to less than 2μL. To overcome this problem programmed temperature sample introduction i.e. PTV is used[7]. PTV allows larger aliquot of the sample analyte to be deposited on to the column while simultaneously eliminating the solvent. Now days this technique is commonly referred to as large volume injection (LVI) **(Figure 6)**.
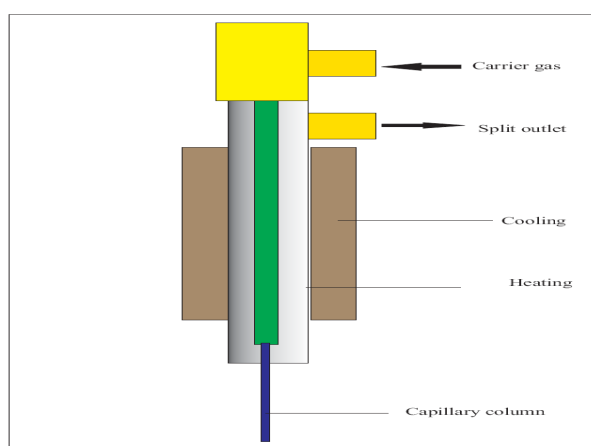

Figure 6: Diagram of a PTV inlet system.

A functional difference between PTV inlet and split or splitless inlet lies in the temperature control. The split and splitless inlet normally operate at elevated temperatures (more than 200^{0}C) where the sample is initially flash vaporized and then enter into the column. Although LVI can operate under these elevated isothermal temperatures but in this rapidly heating or cooling the liner is a carried out. Heating rates are near to 10^{0}C s^{-1} and subambient cooling are common. At the time of working sample is introduced on to the inlet liner usually at a temperature below the boiling point of the solvent followed by rapid heating for the transferring sample from the liner to the front of the column in a very narrow band.

A prerequisite for proper quantitative analysis via LVI/GC requires the vapor pressures of the solutes being analyzed is significantly have higher than that of the solvent. Failure to meet these conditions will leads to poor precision and non reproducible results. The Parameter which affects the performance of LVI/GC are flow rates, injection speed, temperature and vapor pressures. So it is very essential to control these operating parameters for getting accurate and reproducible results.

Applications

The determination of trace components in water usually includes sample preparation steps that enrich the analytes and remove the matrix. The sample enrichment is time consuming and labor intensive operation. Elimination of sample enrichment process would not only decrease analysis cost and also increases the precision of the method. In LVI system water is directly injected without any pre-concentration which helps to increase the precision and reduce time required for the analysis. Other categories such as the analysis of beverages have found significant assay improvements using LVI system.

The most important feature of the PTV is the capability for ultratrace analysis (i.e. sample concentrations below 1ppt) via the introduction of sample aliquots up to 1mL[7]. This is done by operating the PTV in the solvent venting mode and it is referred as SVSF.

4. MULTIDIMENSIONAL GAS CHROMATOGRAPHY[8]

Multidimensional gas chromatography is defined as a GC system of two or more columns of different selectivity and a device that enables the selective transfer of a portion of a chromatographic run from one column to second column (i.e. Heartcutting system). Numerous different hardware implementations of capillary-to-capillary heartcutting MDGC system are commercially available. On the basis of principle of working MDGC mainly divided in to two types are as follows:

A)   Heart-cut MDGC

In this very small portion of the material exiting the first column is introduced to the second column and subjected to both separation dimensions but in case when number of heart-cuts gets high enough and the time for the separation is short this technique is not useful.

B)   Comprehensive MDGC

In this technique material exiting the first dimension is sampled frequently enough, the separation in the first dimension is preserved and all of the compounds in the sample are subjected to both separation dimensions.

The two orthogonal GC columns in the systems are coupled by a special interface (Modulator) that is capable of either sampling or collecting the effluent from the first column and periodically introducing it to the second column at a rate that allows the original first dimension separation to be preserved. The conceptual arrangement of MDGC system shown in **(Figure 7)**.
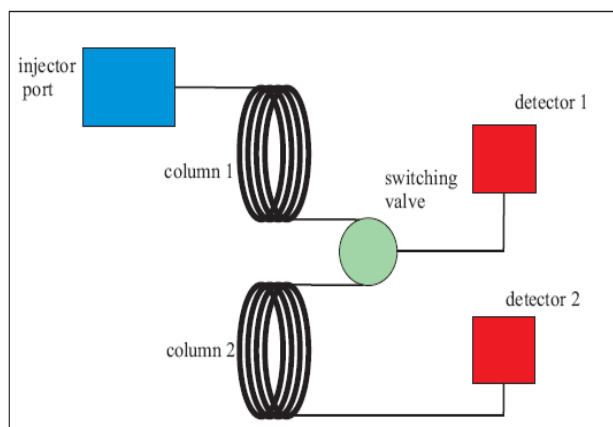
Figure 7: Diagram of multidimensional gas chromatography system.

Basic instrumentation of MDGC is similar to one dimensional gas chromatography. When considering injectors and injection techniques, any technique that is used for conventional GC can in principle also be applied for GC x GC analyses.

Primary columns are generally 15-30m long with internal diameter of 0.25mm and film thickness in the range of 0.25-1.0μm. The first dimension columns typically have a non-polar stationary phase either 100% polydimethylsiloxane or 95/5% methyl/phenyl siloxane. The second dimension separation must be very fast and performed with a stationary phase that is different from that used in the primary column. Typical dimension for secondary columns range from 0.5-1.5m in length with 0.1mm diameter sometimes columns having 0.25mm diameter are also used with the thinner film of stationary phase i.e. 0.1-0.25μm as they of a higher efficiencies. The stationary phase chosen for the second dimension must offer a different separation mechanism than the primary column. Typically wax based phases are used.

Both columns are usually placed in the same GC oven. Sometimes separate oven for the second column is used. The application of a second oven provides more flexibility in the control over the secondary separation. The most important requirement for the detectors used for MDGC is that they should be fast. Detectors like Flame Ionization Detector (FID), Atomic Emission Detector (AED) and Sulphur Chemiluminescence Detector (SCD) are used.

The heart of any MDGC system is the modulator. Based on principle of working modulators divided into two main categories i.e. thermal modulators and valve based modulators.

A) Thermal modulators

1. Heater based modulators

A segment of thick filmed capillary column coated with a layer of gold paint as the interface between the primary and secondary column. As a chromatographic band from the primary column entered the thick-filmed segment of the column, it partitioned into the stationary phase and

slowed down, focusing into a narrow band. The analytes trapped in this way were periodically released by thermal desorption caused by passing electrical current through the metallic coating on the capillary. Trapping was restored again once the capillary cooled down to the oven temperature but due to the high temperature compounds arriving at the modulator which we have not focused them also simply pass through the modulator. This results in broad injection pulses onto second column. To solve this problem, a dual stage trap was proposed. In this both traps operated alternately and because of this broad injection pulses are eliminated.

The original GC interface had many weaknesses; mostly due to the modulators capillaries and the paint coating were not very reproducible. The rotating thermal modulator **(Figure 8)** accomplished trapping, focusing and re-injection through the use of rotating slotted heater that periodically passed along a segment of thick-film GC column acting as the trap. When analytes entered in the interface, they were trapped and after that desorption was carried out by using mechanical heater that was held at an elevated temperature. As the heater passed over the column in the same direction as the carrier gas causes compound focused into a narrow band before entering the second column. This modulator was the first to become commercially available.
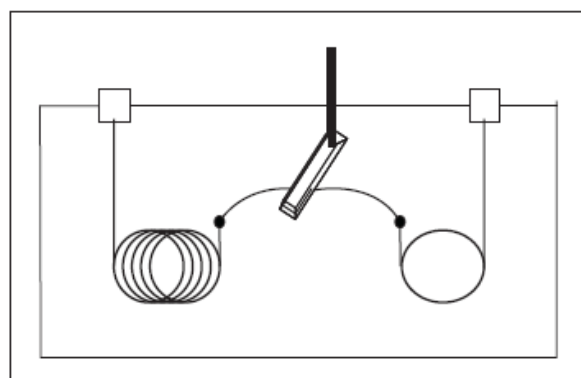

Figure 8: Schematic diagram of the rotating thermal modulator.

2. Cryogenic Modulators[10]

This is one of the types of thermal modulators. Working at high temperature sometimes leads to destruction of sample to eliminate these problems cryogenic modulators are used. Principle behind the working of cryogenic modulator is that the segment of column is cooled with liquid CO_2 to cause analytes to partition into the stationary phase in a small region of the column. In longitudinally modulated cryogenic system (LMCS) to introduce the material to the second dimension column the trap is physically moved to a region of the column upstream of the trapping position. The cooled region is rapidly heated and trap material is focused in second dimension column **(Figure 9)**.
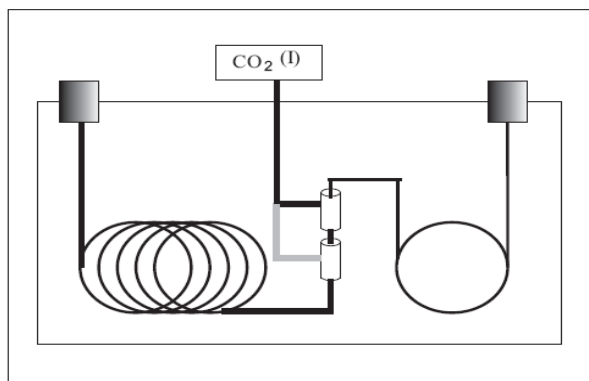
Figure 9: Schematic diagram of the longitudinally modulated cryogenic system.

LMCS system is potentially is weak because of the moving part is used for trapping and for desorption and hence it is not very useful. To overcome this problem another cryogenic system is used in which two liquid CO_2 cold jets for trapping and two hot jets for desorption is used **(Figure 10)**[9]. In this moving parts are not used therefore it is more useful than the longitudinally modulated cryogenic system (LMCS).
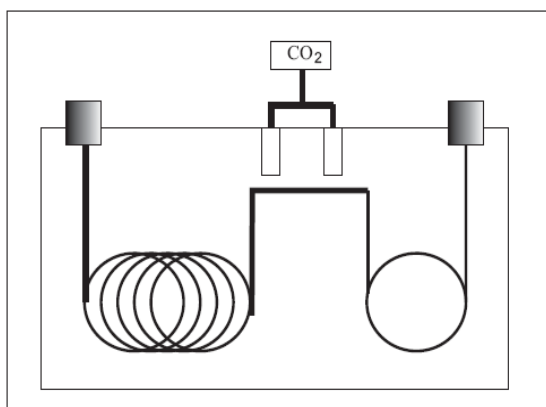

Figure 10: Schematic diagram of the dual cryojet interface.

B)   Valve based modulators

Valve based modulators are one of the type of interfaces used in MDGC. In this most of the effluent coming from primary column was vented to the atmosphere. At the same time carrier gas from an auxiliary supply was delivered to the secondary column. The effluent from the primary column was sampled periodically by actuating the valve for a brief period of time, and the secondary chromatogram was recorded this can be repeated for several time. Very small amount of the effluent from the primary column were taken and separated hence it is less sensitive than the other modulators **(Figure 11)**
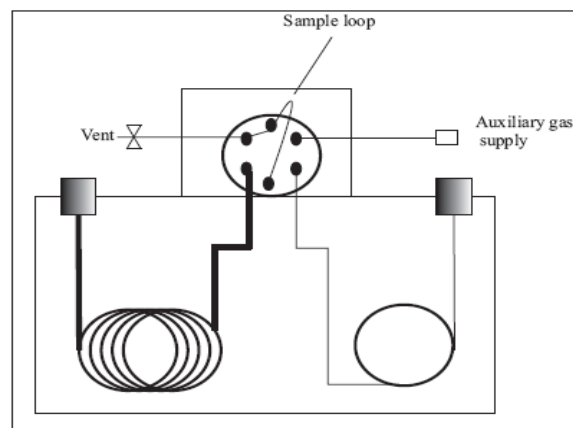

Figure 11: The valve based differential flow modulation GC x GC interface.

Applications[8]

MDGC can provide the user with possibilities as increased separation power, good sensivity and very good and accurate structured chromatograms. Now days MDGC technique is used in various fields such as food and fragrance, forensic and health science and in petroleum industries.

Petroleum samples can contain number of compounds, and in some times many of them are of not interest. MDGC can be applied to the analysis of petroleum samples such as non-aromatic solvent, kerosene, crude oil etc. Similarly MDGC can be utilizing for the analysis of environmental compounds, determination of many small, highly volatile, polar compounds in an urban atmosphere. It is also used to investigate thermodynamic properties of compounds like the characteristic interconversion of E/Z isomer in oximes.

The MDGC technique is also used for the analysis of pyrolytic products generated from primary and secondary pyrolysis of cellulose, lignin and sewage sludge samples.

B)   POST-GAS CHROMATOGRAPH SAMPLE ANALYSIS TECHNIQUES

The principle governing sample detection by GC is governed by well-defined consistent responses of particular sensors to the presence of an analyte within the detector environment. Detectors coupled with GC by appropriate interfacing of the end of the GC column with the sensors. Ideal characteristics of any detectors which are taken in to the consideration are linearity in responses, dynamic range, noise level, detector sensitivity and temperature sensitivity. Different types of detectors are used with GC and some of them are MS and IR.

1.   GAS CHRMATOGRAPHY/ MASS SPECTROSCOPY[11-13]

The mass spectrometer is a universal detector for gas chromatographs, since any compounds that can pass through a GC is converted into ions in the mass spectrometer. At the same time highly specific nature of a mass spectrum makes the mass spectrometer a very

specific GC. GC can couple with mass spectrometer by using suitable interfacing device **(Figure 12)**. Interface systems contain an enrichment device. The high pumping speeds used in mass spectrometers may permit the total effluent from the capillary GC columns to be transported to the ion source of the mass spectrometer. If we are using chemical ionization reagent gas as carrier gas then effluent is directly introduced into the mass spectrometer.
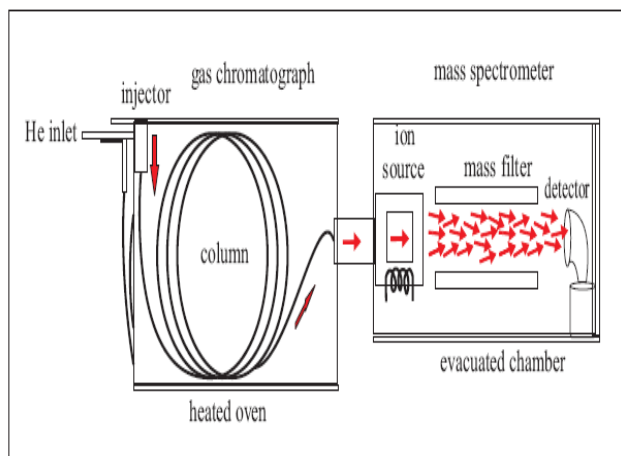

Figure 12: Schematic diagram of GC/MS

Interfacing devices are very important in coupling of GC with mass spectrometer. Different types of interfacing devices are used and these are effusion separator, jet separator and membrane separator.

A) Effusion separator

In this the carrier gas molecules are separated by an effusion chamber which is made of ultrafine-porosity sintered glass with an average pore size of about 10^{-4}cm. The porous barrier is surrounded by vacuum chamber. The lighter carrier gas permeates the effusion barrier in preference to the heavier organic molecules. Enrichment is typically five-to six fold and the yield (sample throughput) is an about 27%.

B) Jet separator

A precisely aligned, supersonic jet system is effective in removing the carrier gas by effusion effluent from the GC is throttled through a fine orifice, where it rapidly expands into a vacuum chamber. During this expansion, the faster diffusion rate of carrier gas results in higher sample concentration in the core of the gas stream which is directed toward a second jet. The short path through the interface to the ion source reduces dead volume, which gives better peak separation.

C) Membrane separator

In this system dimethyl silicon rubber membrane of thickness 0.025-0.040mm is used. Helium has a low permeability whereas the organic molecules pass through the membrane and directly into the high vacuum of MS system. Enrichment values are 10-20 folds.

Several types of mass spectral analyzers are available as GC detectors such as quadrupole, ion trap and time of flight. Many quadrupole mass spectrometers are calibrated using perfluorotributylamine (PFTBA). Data in GC/MS collected in two modes that is scan mode and selective ion monitoring (SIM) chromatogram mode. Scan mode mainly utilizes for the determination of qualitative information of compounds while selective ion monitoring mode is used for the quantitative purpose and sometimes data is collected in scan and SIM mode.

Applications

GC/MS currently finds a very wide array of applications including analyses of biological tissue, petroleum residues, pesticides, essential oils and pharmaceutical. It is used in identification of unknown impurities in pharmaceutical analysis[12]. The main advantages of a mass spectrometer as a detector for gas chromatography are its increased sensitivity and specificity in identifying unknowns or confirming the presence of suspected compounds.

2. GAS CHRMATOGRAPHY / FOURIER TRANSFORM INFRARED SPECTROSCOPY[13-16]

Gas chromatography/ infrared spectrometry is a coupled technique in which the gas chromatograph does the separating and the infrared spectrometer does the identifying. Usually a capillary GC column is a coupled with a Fourier transform infrared spectrometer. The modulated IR beam is focused into a heated "light pipe" cell through which the GC effluent is directed. The light pipe is a glass tube with a gold coating on the inside and IR transmitting windows affixed to each end **(Figure 13)**. The IR beam travels down the light pipe (Internal diameter = 2mm and length = 50cm) by multiple reflections from the gold coating. The transmittance can be 25%.
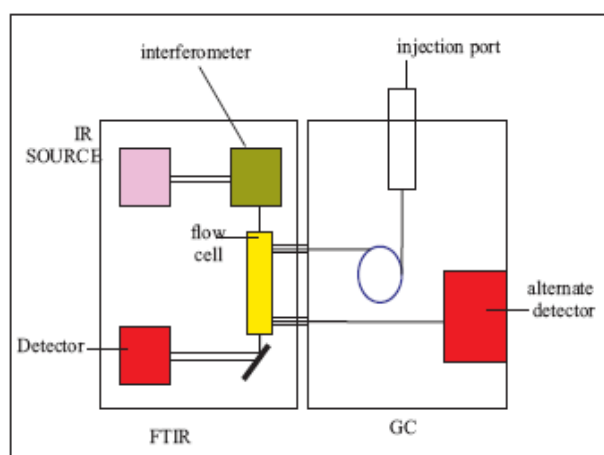

Figure 13: Schematic diagram of GC/FTIR

The detection limit of GC-FTIR systems are usually between 10-100ng, a chromatographic column with 0.3-0.5mm internal diameter and a film thickness of 0.5-1.5µm gives a suitable compromise between adequate

resolution and reasonable sample capacity. Slow column temperature program must be used. A blank spectrum is taken because it is essential for the mathematical calculations involved in the conversion of the interferogram to IR spectrum.

In case of data collection in GC-FTIR Fourier transform mathematics converts the time domain data into the frequency domain. These computations can be accomplished by using computer.

Gas chromatography/Fourier transform vapor phase infrared spectroscopy (GC/FTVPIR) is an approach of GC/FTIR. The main difference in between GC/FTIR and GC/FTVPIR is that the a constant inert and dry purge gas, such as nitrogen, must continuously purge the optical system in Gas chromatography/Fourier transform vapor phase infrared spectroscopy (GC/FTVPIR)[16].

To obtain the necessary sensitivity from the IR detector, the mercury cadmium telluride IR detector must be cooled with liquid nitrogen. Most commercial systems provide IR detectors having Dewars capable of housing enough liquid nitrogen for 4h. Thus, periodic checking of the detector liquid-nitrogen level is essential.

2.3 GAS CHRMATOGRAPHY/ FOURIER TRANSFORM MATRIX ISOLATED INFRARED SPECTROSCOPY[2]

The successful operation of a Gas chromatography/ Fourier Transform Matrix Isolation Infrared Spectroscopy (GC/FTMIIR) set-up depends on complete isolation of the effluent of a GC column at temperatures approaching 10K in an inert noble gas matrix. Complete isolation of the molecule in the matrix virtually eliminates all intermolecular interactions. In addition, molecular rotation is reduced at the very low temperature. Once captured at 10K, usually in argon, maintenance of the temperature is fundamental to the success of the approach because the FTIR spectra are gathered post-decomposition. The availability of argon in which the analytes are captured is made possible by employing helium GC carrier gas containing approximately 5% argon. At 10K helium does not freeze and hence it is pumped away via a diffusion pump-roughing pump configuration.

Instrumentation parts of GC/FTMIIR consist of GC, GC transfer line, collection disk, vacuum chamber and infrared spectrometer **(Figure 14)**. The collection disk is constructed of oxygen free high conductivity copper plated with hardened gold. The curved surface of the collection disk is highly polished to yield a uniform mirror finish. The surface of collection disk serves as platform for the collection of the effluent from the GC column. The effluent is the carrier gas containing about 5% argon as well as any analytes that may be present. When the effluent strikes the surface of collection disk at 10K the analytes and argon freeze on the disk and the carrier gas is pumped away. Simultaneously, while the effluent is exiting from the transfer line, the collection disk is rotated at a predetermined rate. Thus a collection disk rotates

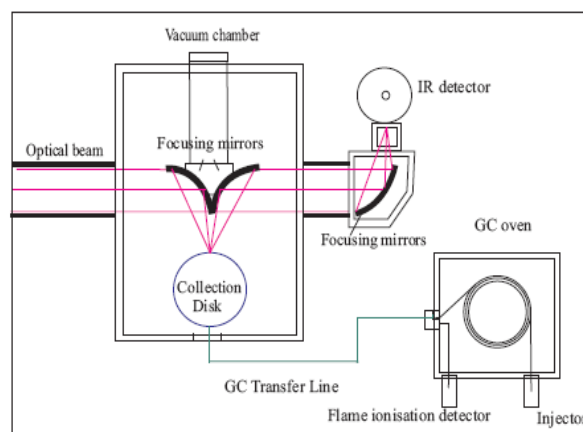thin layer of argon formed on the surface of collection disk.

Figure 14: Schematic diagram of GC/FTMIIR

The position and characteristics of the GC transfer line relative to the surface of the collection disk are very important for the optimum deposition of the argon. The capillary GC transfer line is made of deactivated fused silica of approximately 150µm in diameter. The end of GC transfer is positioned within approximately 150µm of the surface of the collection disk. Incorrect positioning of the tip of the GC transfer line will result in an uneven or broken appearing thread of argon on the surface of the collection disk.

In case of data collection the initial portion of the thread is scanned to establish a background then the disk is rotated at a predetermined rate while FTIR interferograms are rapidly taken and stored on the computer. After the entire run has passed under the IR beam, the data are converted into a plot of IR adsorption versus time in FTMIIR experiments the data are collected on post run.

Applications

This technique is very useful and advantageous because in this technique absence of intermolecular interaction and absence of molecular rotation leads to IR absorption spectra from the MI experiment having significant differences from IR spectra taken in other matrices. It is useful for the study reactive molecules such as free radicals and reaction intermediates. The virtual absence of molecular interaction and rotation in MI species has the capability of detecting difference in the IR spectra of cis and trans isomers in the selected compounds. Example, cis and trans isomers of pinane, meso and racemic-2,4-pentanediol were readily distinguishable one from the other by using FTMIIR spectra.

CONCLUSION

Development efforts in modern GC base instrumentation depending on the integration of GC, computers, and powerful information rich detectors like MS and FTIR have produced a wide array of very potent systems. Combination of online sample introduction SPME with GC

and MS has let to hyphenated systems with applications in wide variety of areas.

Future analytical systems will most probably encompass the use of high speed GC with company high speed mass spectrometers, infrared detectors. Advancement in online sample introduction systems such as SPME, LVI, MDGC and HS techniques will assist in lowering further both the qualitative and quantitative limits of detection. This advancement will helps in data collection and rapid conversion of data into the information and this information into knowledge.

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