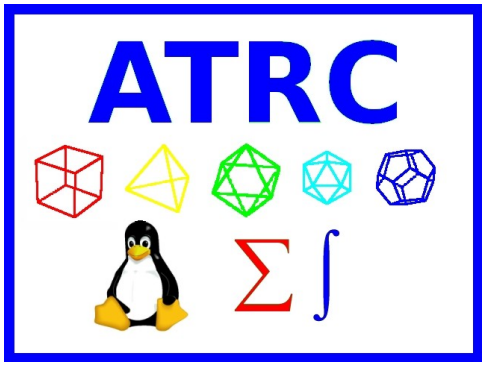
Chromatigration ™

by Applied Technology Research Center ™.

Date : 15 April 2021



Chromatigration ™ is a system designed by ATRC to facilitate the integration of many types of lab instruments from various vendors.

We also integrate the factory from the PLC and VFD to the ERP and DMS for traceability compliance with the HVAC, BMS, HPLC, CMDB and DMS in between.

Our combination allows the users to be able to have more capabilities than they usually have by the use of a set of products from a single vendor.

The services is provided based on the requirements of the customer and more products from other vendors can be added for combining. Proprietary components related to hardware, software and procedures are created by ATRC so more integration between systems is possible.

As vendor neutral and independent system developer and integrator, our sales and marketing do not have to sell any hardware to be able to sell a solution. We offer our customers the best combination of hardware, software and proprietary integration components so our customers can more easily choose the right instrumentation for their laboratory, regardless of age or manufacturer. We support more varieties of old and new laboratory instrumentation than common vendors. Our products and services allow our customers to maximize ROI on instrument purchases and embrace new technologies and features of the latest instrumentation.

File Formats supported

- ABSciex DAD (*.wiff)
- ABSciex Reports (*.txt)
- ASCII text (.*)
- ASCII GC signal (.*)
- Agilent (HP) Chemstation GC/MS, MS (.d)
- Agilent (HP) Chemstation GC Type 138 signal (.d)
- Agilent (HP) Chemstation LC Type 233 signal (.ch)
- Agilent (HP) Chemstation GC Type 331 signal (.ch)
- Agilent ChemStation FID (*.D/ *.ch)
- Agilent (HP) Chemstation GC/MS, MS, GC or LC (.d/.ms)
- Agilent MSD (*.D/DATA.MS)
- Agilent MSD (*.D/data01.icp)
- Agilent VWD (*.D/vwd1A.ch)
- Beckman Gold CE (.chr)
- Bruker Flex MSD (fid)
- Bruker/Varian MSD (*.SMS)
- Bruker/Varian MSD (*.XMS)
- ChemStation Reports (*.ch)
- ChromTech MSD (*.dat)
- Chromperfect GC or LC (.??r)
- Chromperfect GC signal (.??r)
- CAMAG VWD (*.DFM)
- DataApex FID (*.PRM)
- EasyChrom GC or LC (.*)
- EZChrom Reports (*.dat)
- EasyChrom GC signal (.*)
- Excel spreadhseet GC signal (.xls)
- Finnigan ITDS GC/MS or MS (.dat)
- Finnigan GCQ, Saturn GC/MS or MS (.ms)
- Finnigan ICIS GC/MS or MS (.dat)

- Finnigan GCQ MS, Saturn MS (.ms)
- Finnigan Incos MS (.ms)
- Finnigan ITDS MS (.dat)
- Galactic Grams GC/MS or MS (.spc) or GC (.gcm)
- Galactic Grams GC/MS, MS (.spc), GC or LC (.gcm)
- Inficon GC/MS or MS (.acq)
- JCAMP-DX GC/MS, MS, or GC (.jdx)
- Kratos Mach3 GC/MS or MS (.run)
- Magnum MS
- MassFinder FID/MSD (*.mfg)
- MassFinder Library (*.mfl)
- MassHunter MSD (*.D/AcqData)
- MassHunter (*.D/CPDET1A.CH)
- netCDF GC/MS, MS, GC or LC (.cdf)
- netCDF GC/MS, MS, or GC (.cdf)
- NIST Database MSD (*.db)
- PerkinElmer FID (*.raw)
- PE TurboMass GC/MS or MS (.raw)
- Shrader/GCMate GC/MS or MS (.lrp)
- Shimadzu MS (.d??)Thermo/Finnigan FID (*.dat)
- Shimadzu MSD (*.qgd)
- Shimadzu QP-5000 MSD (*.R##)
- Shimadzu Database MSD (*.spc)
- Shimadzu GC10 FID (*.C0#)
- Shimadzu FID (*.gcd)
- Thermo/Finnigan FID (*.raw)
- Thermo/Finnigan MSD (*.cgm)
- Thermo/Finnigan Element II (*.dat)
- Thermo/Finnigan ICIS (*.dat)
- Thermo/Finnigan ITDS (*.DAT)
- Thermo/Finnigan ITS40 (*.MS)
- Thermo/Finnigan MAT (*.dat)

- Thermo/Finnigan RAW MSD (*.raw)
- Varian FID (*.run)
- Varian Saturn 2000 GC/MS or MS (.sms)
- Varian Saturn XMS GC/MS or MS (.xms)
- Varian Saturn 2000 GC/MS or MS (.sms)
- Varian Saturn XMS GC/MS or MS (.xms)
- Varian Star GC (.run)
- VG MSD (*.DAT_001)
- Waters MSD (*.RAW/_FUNC001.DAT)


Task Quick Access
Project Expl...
Data Explorer
Sequence Ex...
Drives
Home
User Location
Chromatogram1.D
Chromatogram10.ULF
Chromatogram11.ULF
Chromatogram12.ULF
Chromatogram13.ULF
Chromatogram14.D
Chromatogram15.ocb
Chromatogram2
record.dat
Chromatogram3.PRM
Chromatogram4.ocb
Chromatograms5.cdf
Chromatogram6.cdf
Open Selected Measurements
*chromatogram4 [MSD]
*record [MSD]
DEFAULT
Intensity
Relative Intensity [%]
Minutes
Edit History
Targets
Combined Scan
Scan range: 1614–2112 | RT range: 20.872–25.132
Intensity
Intensity [%]
m/z
Chart
Table
Console
Scan Chart
AUTO_DETECT
Scan: 1692 | RT: 21.534 | RI: 0 | Detector: MS2 | Type: Centroid | Signal: 45220120
175 > 112.0 @15
302 > 175.0 @15
302 > 111.0 @18
Feedback
Scan Table
Molecule View
m/z
abundance
parent m/z
parent resolutio
daughter m/z
59 > 31.0 78814.0 59.0 1.2 31.0 1.2
59 > 59.0 343252.0 59.0 1.2 59.0 1.2
175 > 111.0 2598436.0 175.0 1.2 111.0 1.2
302 > 111.0 145540.0 302.0 1.2 111.0 1.2
175 > 112.0 25270624.0 175.0 1.2 112.0 1.2
132 > 117.0 108885.0 132.0 1.2 117.0 1.2
258 > 175.0 5608312.0 258.0 1.2 175.0 1.2
302 > 175.0 5401272.0 302.0 1.2 175.0 1.2
302 > 175.0 5344152.0 302.0 1.2 175.0 1.2

Complete Machine Backup without any downtime.

ATRC has developed a proprietary backup solution which does a real time backup of the computers which are running the HPLC devices and analyzing the data.

This allows CDS systems to have all of their data intact while meeting CFR 21 part 11 data security compliance requirements.

Our backup system backups the complete machine along with any OS, Data, files, softwares which are running within it. This allows our system to be compatible with all types of operating systems and applications running in it.

We also have proprietary software connectors for tracking the usage of active CDS applications. This allows heavier optimization tasks to be performed only when the CDS or Acquisition applications are inactive.

Under Development

We are developing an integrated configuration management system and also a document management system so that all lab data can be consolidated into one area and have the facility of digital signatures.

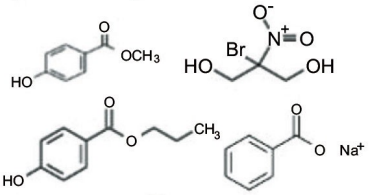
This shall make all of the data components of the complete lab be compliant for FDA CFR 21 Part 11 requirements.

We are also developing the integration of the following types of machines for facilitating tracibility in Pharma and food processing industries.

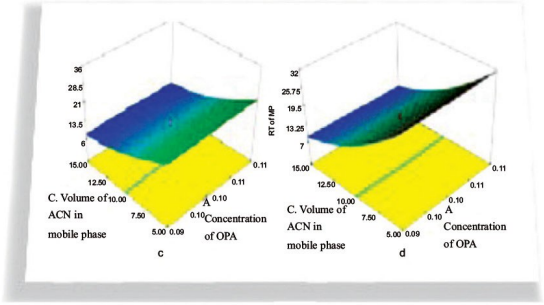
Special databases for peaks and masses to facilitate quality assurance of herbal medicines which are composed usually of multiple compounds.

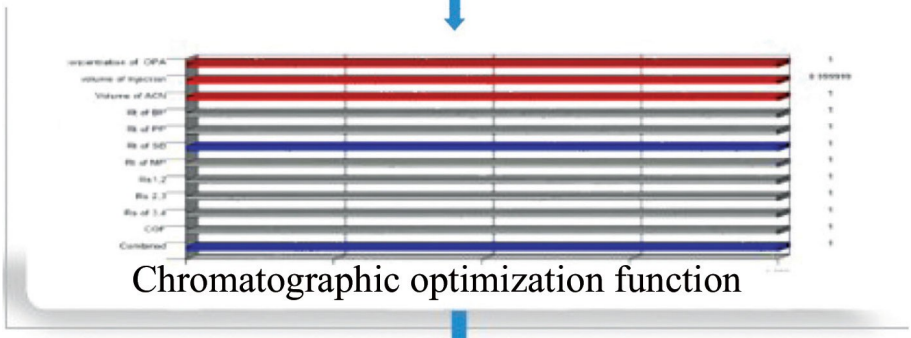
Herbal Formulation

Quaternary Preservatives

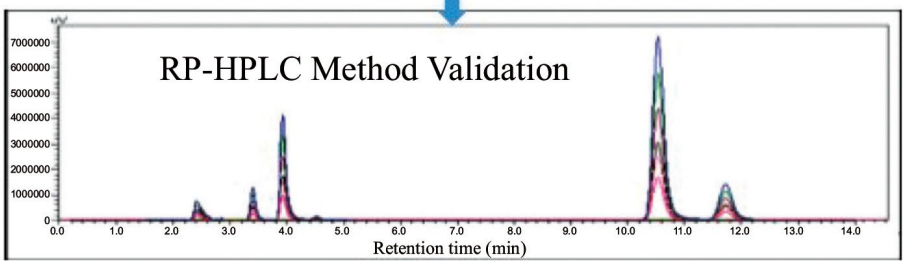


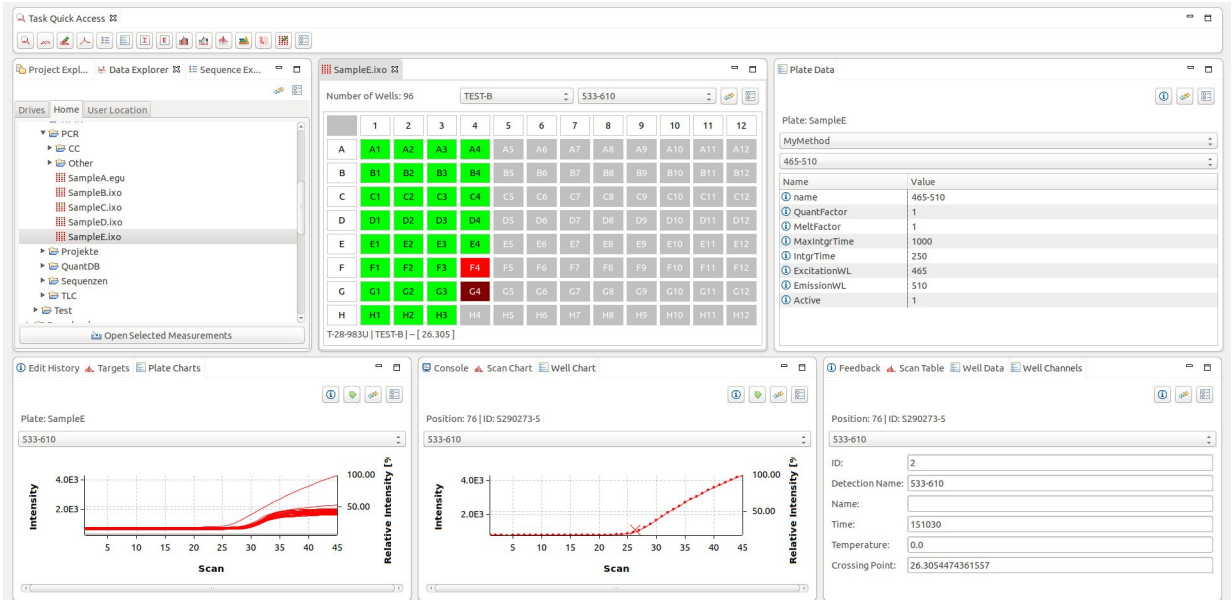
Central Composite Design





Chromatographic optimization function

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Project Expl... Data Explorer Sequence Ex...
Drives Home User Location
PCR
CC
Other
SampleA.egu
SampleB.ixo
SampleC.ixo
SampleD.ixo
SampleE.ixo
Projekte
QuantDB
Sequenzen
TLC
Test
Open Selected Measurements
SampleE.ixo
Number of Wells: 96
TEST-B
533-610
T-28-983U | TEST-B | – [26.305]
Plate Data
Plate: SampleE
MyMethod
465-510
Name
Value
name 465-510
QuantFactor 1
MeltFactor 1
MaxIntgrTime 1000
IntgrTime 250
ExcitationWL 465
EmissionWL 510
Active 1
Edit History Targets Plate Charts
Plate: SampleE
533-610
Intensity
Relative Intensity [%
Scan
Console Scan Chart Well Chart
Position: 76 | ID: S290273-5
533-610
Intensity
Relative Intensity [%
Scan
Feedback Scan Table Well Data Well Channels
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533-610
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Name:
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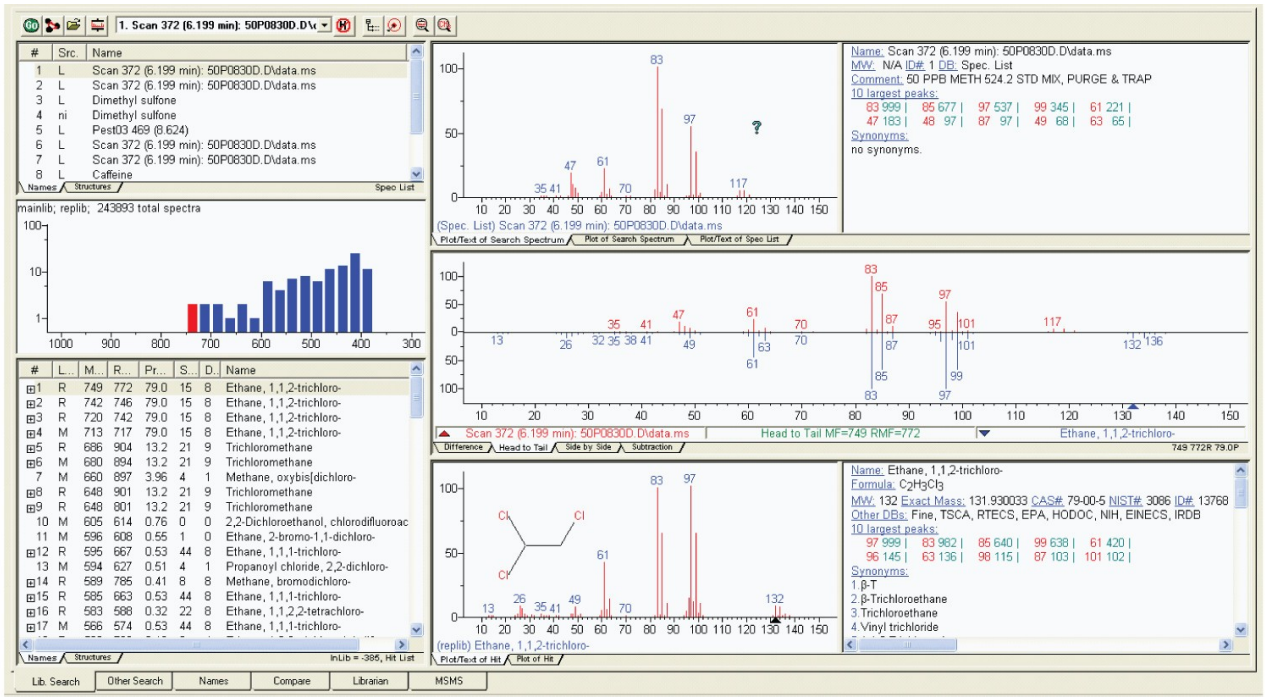
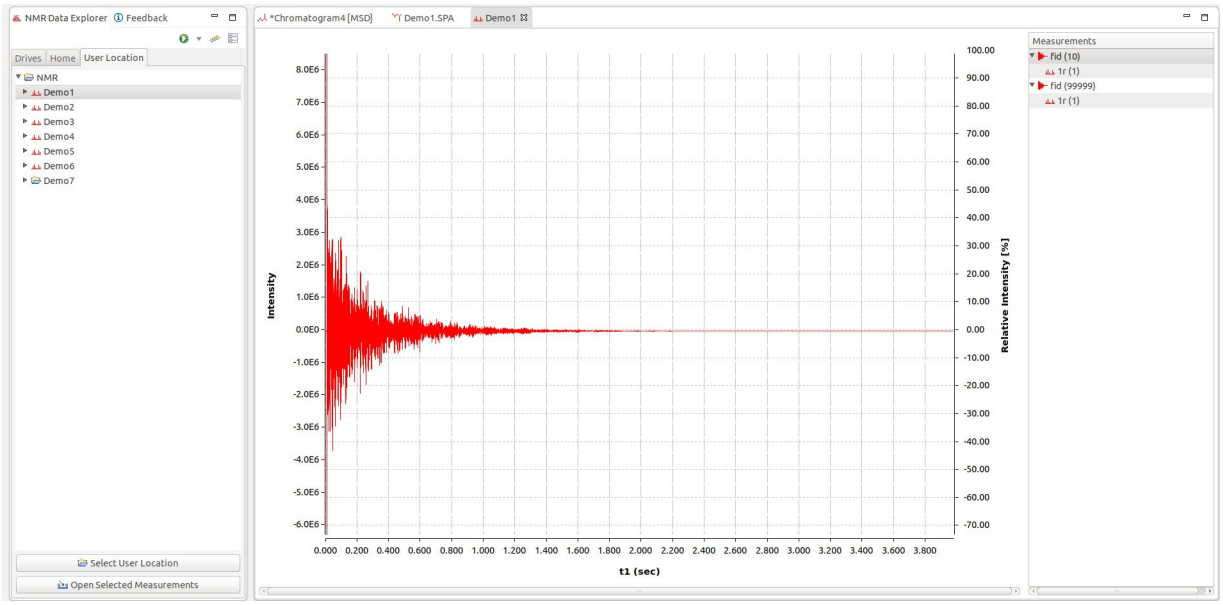

1. Scan 372 (6.199 min): 50P0830D.D\
Src. Name
1 L Scan 372 (6.199 min): 50P0830D.D\data.ms
2 L Scan 372 (6.199 min): 50P0830D.D\data.ms
3 L Dimethyl sulfone
4 ni Dimethyl sulfone
5 L Pest03 469 (8.624)
6 L Scan 372 (6.199 min): 50P0830D.D\data.ms
7 L Scan 372 (6.199 min): 50P0830D.D\data.ms
8 L Caffeine
Name: Scan 372 (6.199 min): 50P0830D.D\data.ms
MW: N/A ID# 1 DB: Spec. List
Comment: 50 PPB METH 524.2 STD MIX, PURGE & TRAP
10 largest peaks:
83 999 | 85 677 | 97 537 | 99 345 | 61 221 |
47 183 | 48 97 | 87 97 | 49 68 | 63 65 |
Synonyms:
no synonyms.
(Spec. List) Scan 372 (6.199 min): 50P0830D.D\data.ms
mainlib; replib; 243893 total spectra
L... M... R... Pr... S... D... Name
1 R 749 772 79.0 15 8 Ethane, 1,1,2-trichloro-
2 R 742 746 79.0 15 8 Ethane, 1,1,2-trichloro-
3 R 720 742 79.0 15 8 Ethane, 1,1,2-trichloro-
4 M 713 717 79.0 15 8 Ethane, 1,1,2-trichloro-
5 R 686 904 13.2 21 9 Trichloromethane
6 M 680 894 13.2 21 9 Trichloromethane
7 M 660 897 3.96 4 1 Methane, oxybis[dichloro-
8 R 648 901 13.2 21 9 Trichloromethane
9 R 648 801 13.2 21 9 Trichloromethane
10 M 605 614 0.76 0 0 2,2-Dichloroethanol, chlorodifluoroac
11 M 596 608 0.55 1 0 Ethane, 2-bromo-1,1-dichloro-
12 R 595 667 0.53 44 8 Ethane, 1,1,1-trichloro-
13 M 594 627 0.51 4 1 Propanoyl chloride, 2,2-dichloro-
14 R 589 785 0.41 8 8 Methane, bromodichloro-
15 R 585 663 0.53 44 8 Ethane, 1,1,1-trichloro-
16 R 583 588 0.32 22 8 Ethane, 1,1,2,2-tetrachloro-
17 M 566 574 0.53 44 8 Ethane, 1,1,1-trichloro-
Scan 372 (6.199 min): 50P0830D.D\data.ms
Head to Tail MF=749 RMF=772
Ethane, 1,1,2-trichloro-
Name: Ethane, 1,1,2-trichloro-
Formula: C2H3Cl3
MW: 132 Exact Mass: 131.930033 CAS#: 79-00-5 NIST#: 3086 ID#: 13768
Other DBs: Fine, TSCA, RTECS, EPA, HODOC, NIH, EINECS, IRDB
10 largest peaks:
97 999 | 83 982 | 85 640 | 99 638 | 61 420 |
96 145 | 63 136 | 98 115 | 87 103 | 101 102 |
Synonyms:
1.β-T
2.β-Trichloroethane
3.Trichloroethane
4.Vinyl trichloride
(replib) Ethane, 1,1,2-trichloro-
Lib. Search
Other Search
Names
Compare
Librarian
MSMS


NMR Data Explorer
Feedback
Drives
Home
User Location
NMR
Demo1
Demo2
Demo3
Demo4
Demo5
Demo6
Demo7
Select User Location
Open Selected Measurements
*Chromatogram4 [MSD]
Demo1.SPA
Demo1
Intensity
Relative Intensity [%]
t1 (sec)
Measurements
fid (10)
1r (1)
fid (99999)
1r (1)


Task Quick Access
Project Expl...
Data Explorer
Sequence Ex...
Drives
Home
User Location
Chromatogram1.D
Chromatogram10.ULF
Chromatogram11.ULF
Chromatogram12.ULF
Chromatogram13.ULF
Chromatogram14.D
Chromatogram15.ocb
Chromatogram2
record.dat
Chromatogram3.PRM
Chromatogram4.ocb
Chromatogram5.cdf
Chromatogram6.cdf
Open Selected Measurements
*Chromatogram4 [MSD]
*record [MSD]
DEFAULT
Intensity
Relative Intensity [%]
Minutes
Edit History
Targets
Combined Scan
Scan range: 1614–2112 | RT range: 20.872–25.132
m/z
Chart
Table
Console
Scan Chart
AUTO_DETECT
Scan: 1692 | RT: 21.534 | RI: 0 | Detector: MS2 | Type: Centroid | Signal: 45220120
175 > 112.0 @15
302 > 175.0 @15
302 > 111.0 @18
Feedback
Scan Table
Molecule View
m/z
abundance
parent m/z
parent resolutio
daughter m/z
59 > 31.0 78814.0 59.0 1.2 31.0
59 > 59.0 343252.0 59.0 1.2 59.0
175 > 111.0 2598436.0 175.0 1.2 111.0
302 > 111.0 145540.0 302.0 1.2 111.0
175 > 112.0 25270624.0 175.0 1.2 112.0
132 > 117.0 108885.0 132.0 1.2 117.0
258 > 175.0 5608312.0 258.0 1.2 175.0
302 > 175.0 5401272.0 302.0 1.2 175.0
302 > 175.0 5344152.0 302.0 1.2 175.0

The systems being integrated for the factory include the following :

Manufacturing ERPs.

Configuration management databases based in ITIL and ITSM.

Kanban based management systems.

Popular ERPs like Postbooks, Odoo, OFBiz, OpenBravo, Tryton, ERPNext, Adempiere, Dynamics, iDempere, Metafresh, Oracle, Web ERP, BluSeer, MixERP, EasyERP, WP ERP, Axelor, ERP5, Netsuite, Deltek, Sage, Infor, Syspro, SAP, IFS Applications, Macola, Intacct and many more.

PLC and CPU based factory machines to be integrated to complete the tracking of actions for tracibility.


Schneider
Electric


IDEC
Think Automation and beyond...


HITACHI


OMRON


B&R


rexroth
A Bosch Company


Koyo


DELTA
DELTA ELECTRONICS, INC.


WAGO


pilz
more than automation
safe automation


PHOENIX CONTACT


YOKOGAWA
Co-innovating tomorrow™


EATON
Powering Business Worldwide


Panasonic


LS ELECTRIC

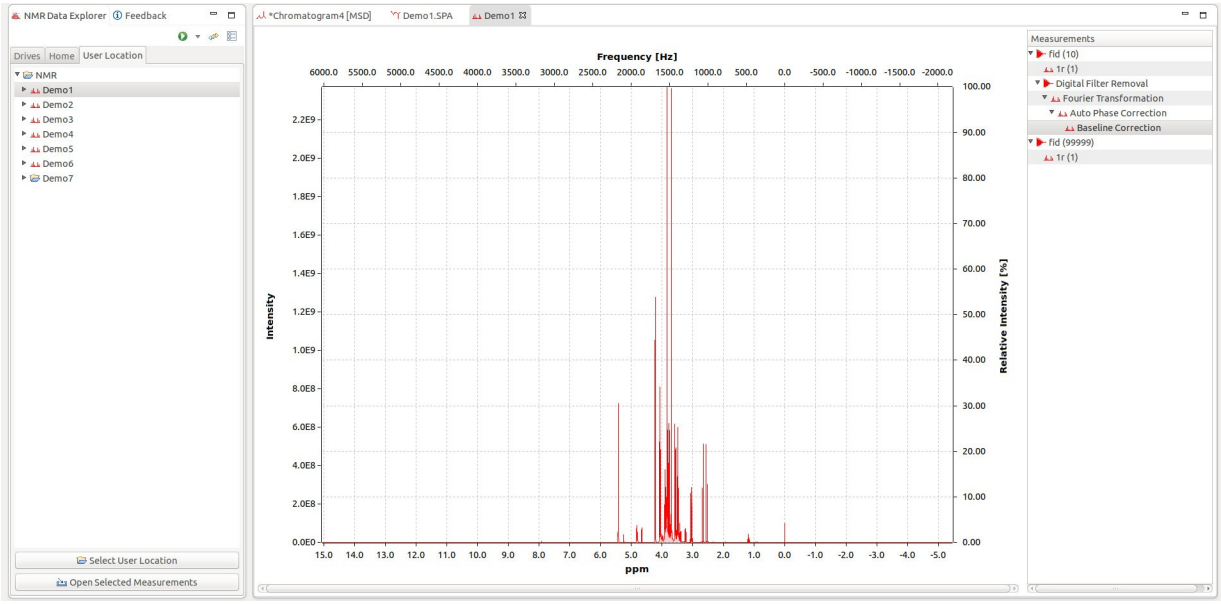

KEYENCE

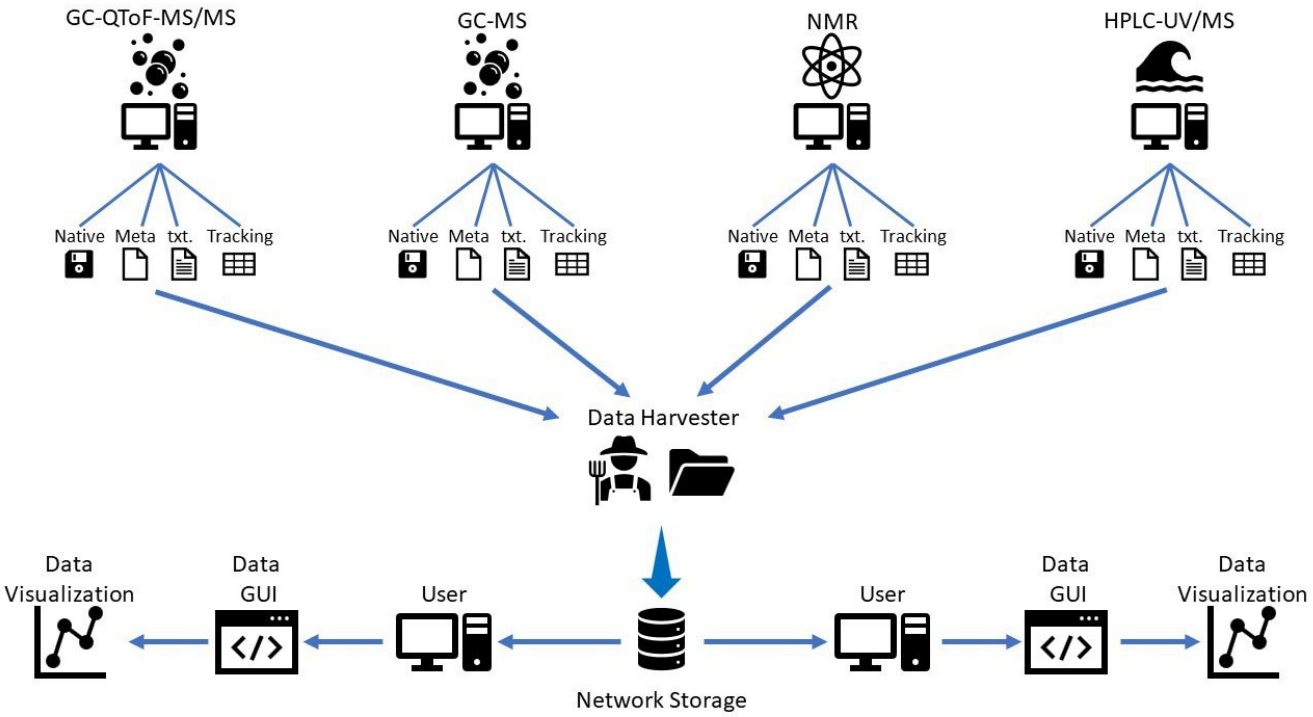

TOSHIBA
Leading Innovation

1D and 2D Barcode scanners and printers.

The Global Language of Business

RFID tagging and readers.


NMR Data Explorer
Feedback
Drives
Home
User Location
NMR
Demo1
Demo2
Demo3
Demo4
Demo5
Demo6
Demo7
Select User Location
Open Selected Measurements
*Chromatogram4 [MSD]
Demo1.SPA
Demo1
Frequency [Hz]
Intensity
Relative Intensity [%]
ppm
Measurements
fid (10)
1r (1)
Digital Filter Removal
Fourier Transformation
Auto Phase Correction
Baseline Correction
fid (99999)
1r (1)


GC-QToF-MS/MS
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Tracking
Data Harvester
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Data GUI
User
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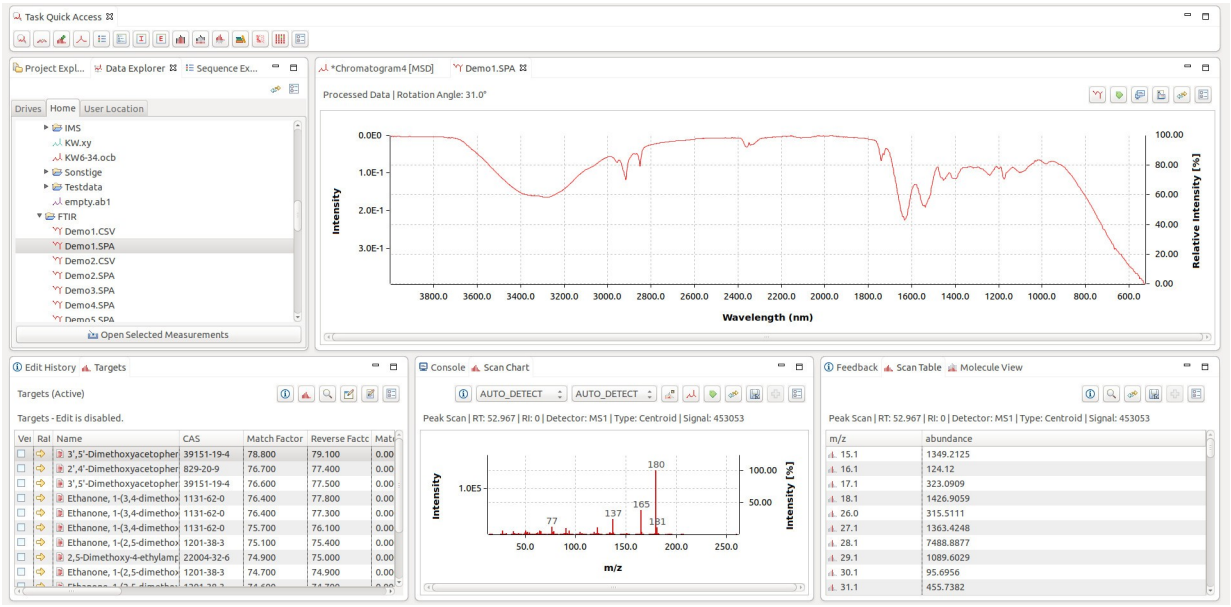
Data Visualization


Task Quick Access
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Data Explorer
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Drives
Home
User Location
IMS
KW.xy
KW6-34.ocb
Sonstige
Testdata
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FTIR
Demo1.CSV
Demo1.SPA
Demo2.CSV
Demo2.SPA
Demo3.SPA
Demo4.SPA
Open Selected Measurements
*Chromatogram4 [MSD]
Demo1.SPA
Processed Data | Rotation Angle: 31.0°
Intensity
Relative Intensity [%]
Wavelength (nm)
Edit History
Targets
Targets (Active)
Targets - Edit is disabled.
Ve
Ral
Name
CAS
Match Factor
Reverse Factc
3',5'-Dimethoxyacetopher
39151-19-4
78.800
79.100
2',4'-Dimethoxyacetopher
829-20-9
76.700
77.400
3',5'-Dimethoxyacetopher
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76.600
77.500
Ethanone, 1-(3,4-dimethox
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77.800
76.400
77.300
75.700
76.100
Ethanone, 1-(2,5-dimethox
1201-38-3
75.100
75.400
2,5-Dimethoxy-4-ethylamp
22004-32-6
74.900
75.000
74.700
74.900
Console
Scan Chart
AUTO_DETECT
Peak Scan | RT: 52.967 | RI: 0 | Detector: MS1 | Type: Centroid | Signal: 453053
m/z
Feedback
Scan Table
Molecule View
abundance
15.1
1349.2125
16.1
124.12
17.1
323.0909
18.1
1426.9059
26.0
315.5111
27.1
1363.4248
28.1
7488.8877
29.1
1089.6029
30.1
95.6956
31.1
455.7382


Task Quick Access
Project Expl...
Data Explorer
Sequence Ex...
Drives
Home
User Location
Chromatogram1.D
Chromatogram10.ULF
Chromatogram11.ULF
Chromatogram12.ULF
Chromatogram13.ULF
Chromatogram14.D
Chromatogram15.ocb
Chromatogram2
Chromatogram3.PRM
Chromatogram4.ocb
Chromatogram5.cdf
Chromatogram6.cdf
Chromatogram7.d
Open Selected Measurements
*Chromatogram4 [MSD]
DEFAULT
Phenol, 2-methoxy-4-meth
Phenol, 2-methoxy-4-pr
4-[(3-Aminobutyl)-2-metho
Benzoic acid, 4-(acetylox
Apocynin
2,6-Dimethoxyamphetami
Guanosine
3',5'-Dime
2-Propanone, 1-(4-hyc
Phenol, 2,6-dimethoxy-4
N-acetyl-3,4-dimethoxyan
1,3,2-Dioxaphosphorinan-2
3-methoxy-2-(1-methyleth
Intensity
Relative Intensity [%]
Minutes
Edit History
Targets
Targets (Active)
Targets - Edit is disabled.
Console
Scan Chart
AUTO_DETECT
Peak Scan | RT: 52.967 | RI: 0 | Detector: MS1 | Type: Centroid | Signal: 453053
m/z
Feedback
Scan Table
Molecule View
2',4'-Dimethoxyacetophenone | COC1=C(C=CC(=C1)OC)C(C)=O

Types of separations which are possible

Gas Chromatography (GC)

Gas chromatography (GC) is an analytical technique used to separate the chemical components of a sample mixture and then detect them to determine their presence or absence and/or how much is present. These chemical components are usually organic molecules or gases. For GC to be successful in their analysis, these components need to be volatile, usually with a molecular weight below 1250 Da, and thermally stable so they don't degrade in the GC system. GC is a widely used technique across most industries: for quality control in the manufacture of many products from cars to chemicals to pharmaceuticals; for research purposes from the analysis of meteorites to natural products; and for safety from environmental to food to forensics. Gas chromatographs are frequently hyphenated to mass spectrometers (GC-MS) to enable the identification of the chemical components.

GC-MS

GC-MS can be used to study liquid, gaseous or solid samples. Analysis begins with the gas chromatograph, where the sample is effectively vaporized into the gas phase and separated into its various components using a capillary column coated with a stationary (liquid or solid) phase. The compounds are propelled by an inert carrier gas such as helium, hydrogen or nitrogen. As components of the mixture are separated, each compound elutes from the column at a different time based on its boiling point and polarity. The time of elution is referred to as a compound's retention time. GC has the capacity to resolve complex mixtures or sample extracts containing hundreds of compounds.

Once the components leave the GC column, they are ionized and fragmented by the mass spectrometer using electron or chemical ionization sources. Ionized molecules and fragments are then accelerated through the instrument's mass analyzer, which quite often is a quadrupole or ion trap. It is here that ions are separated based on their different mass-to-charge (m/z) ratios. GC-MS data

acquisition can be performed in either full scan mode, to cover either a wide range of m/z ratios, or selected ion monitoring (SIM) mode, to gather data for specific masses of interest.

The final steps of the process involve ion detection and analysis, with fragmented ions appearing as a function of their m/z ratios. Peak areas, meanwhile, are proportional to the quantity of the corresponding compound. When a complex sample is separated by GC-MS, it will produce many different peaks in the gas chromatogram and each peak generates a unique mass spectrum used for compound identification. Using extensive commercially available libraries of mass spectra, unknown compounds and target analytes can be identified and quantified.

GC-TOFMS

Comprehensive two-dimensional gas chromatography/time-of-flight mass spectrometry (GC×GC TOFMS) and gas chromatography/high-resolution time-of-flight mass spectrometry (GC-HRT) were used to detect and identify halogenated natural products (HNPs) in tissue homogenate, in this case brominated analytes present in a marine snail. Two classes of brominated anthropogenic compounds, polybrominated diphenyl ethers (PBDEs) and brominated dibenzofurans, were analyzed for comparison. Following conventional preparation, the sample was analyzed using GC×GC-TOF-MS. Isotope ratio scripts were used to compile a list of putatively brominated analytes from amongst the thousands of features resolved in the two-dimensional chromatogram. The structured nature of the chromatogram was exploited to propose identifications for several classes of brominated compounds, and include additional candidates that fell marginally outside the script tolerances. The sample was subsequently analyzed by GC-HRT. The high-resolution mass spectral data confirmed many formula assignments, facilitated confident assignment of an alternate formula when an original proposal did not hold, and enabled unknown identification. Identified HNPs include hydroxylated and methoxylated PBDE analogs, polybrominated dibenzo-*p*-dioxins (PBDDs) and hydroxyl-PBDDs, permitting the environmental occurrence and fate of such compounds to be studied.

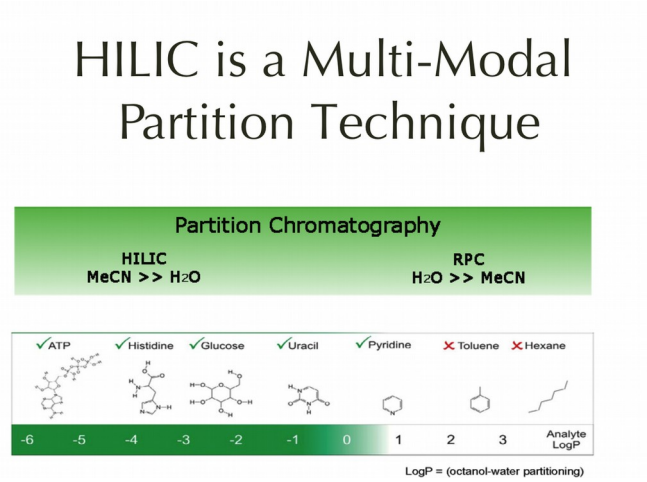
GC×GC-ToF-MS

Conventional gas chromatography with electroantennographic detection (GC-EAD) and two-dimensional (GC × GC) gas chromatography using a time-of-flight mass spectrometric detector (TOFMS), were combined to analyse the female sex pheromone gland extract of the persimmon bark borer, *Euzophera batangensis*. GC-EAD analysis produced two EAD responses in GC areas where no compounds were detected by FID detection. GC × GC/TOFMS analysis of this area indicated the presence of several chemicals, including (*Z*9,*E*12)-tetradeca-9,12-dien-1-ol and (*Z*9)-tetradec-9-en-1-ol, pheromone components of closely related *Euzophera* species. Spectral characteristics, retention behaviour and the ability to elicit GC-EAD responses imply that both identified unsaturated alcohols are candidates for *E. batangensis* sex pheromone components. GC × GC/TOFMS facilitated the analysis of complex matrices on a subnanogram level and was shown to have great potential as a powerful tool in the analysis of insect pheromones.

HPLC

High Performance Liquid Chromatography (HPLC) is a form of column chromatography that pumps a sample mixture or analyte in a solvent (known as the mobile phase) at high pressure through a column with chromatographic packing material (stationary phase). The sample is carried by a moving carrier gas stream of helium or nitrogen. HPLC has the ability to separate, and identify compounds that are present in any sample that can be dissolved in a liquid in trace concentrations as low as parts per trillion. Because of this versatility, HPLC is used in a variety of industrial and scientific applications, such as pharmaceutical, environmental, forensics, and chemicals.
Sample retention time will vary depending on the interaction between the stationary phase, the molecules being analyzed, and the solvent, or solvents used. As the sample passes through the column it interacts between the two phases at different rate, primarily due to different polarities in the analytes. Analytes that have the least amount of interaction with the stationary phase or the most amount of interaction with the mobile phase will exit the column faster.

HILIC



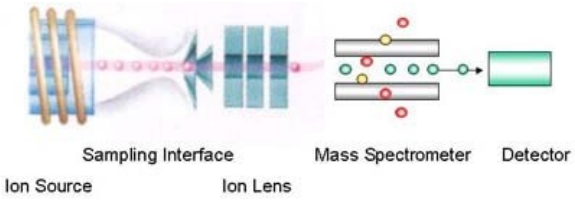
Hydrophilic interaction chromatography : Hydrophilic interaction chromatography (or hydrophilic interaction liquid chromatography, HILIC) is a variant of normal phase liquid chromatography that partly overlaps with other chromatographic applications such as ion chromatography and reversed phase liquid chromatography. HILIC uses hydrophilic stationary phases with reversed-phase type eluents. The name was suggested by Dr. Andrew Alpert in his 1990 paper on the subject. He described the chromatographic mechanism for it as liquid-liquid partition chromatography where analytes elute in order of increasing polarity.

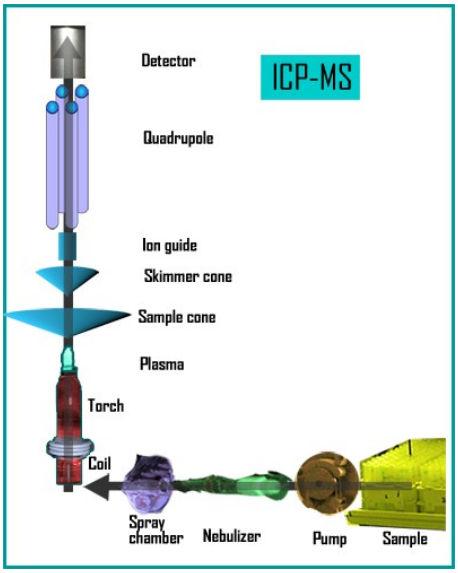
ICP-MS

Over twenty years have passed since ICP Mass Spectrophotometers (ICP-MS) was first introduced by R.S. Houk, A.L. Gray et al. in 1980, then put on the market in 1983, and is now widely used in various fields. This is especially so in the semi-conductor industry, as ICP mass spectrometry is used as analysis method for quality control of high-purity material, where demands increase with the times. Also, it is expected that the method can be applied to analysis of trace amounts of hazardous metals, and recently with various legislations in the environmental field, ICP-MS is used to respond to the stricter environmental and drainage standards. ICP-MS offers the following features:

1. High sensitivity analysis - lower detection limits of most elements are in ppt to ppq-order.
2. Simultaneous multi-element analysis possible.
3. Can determine quality and quantity quickly.
4. Wide dynamic range with 8 figures.
5. Isotope comparison possible.

Structure of ICP-MS
ICP-MS consists of an ion source (ICP), a sampling interface, ion lens, a mass spectrophotometer and a detector.





The ion source, ICP is an ideal ionization source for mass spectrometry, and can ionize over 90% of many elements. Ions produced in the ICP are led through the sampling interface to the mass analysis unit. The sampling interface unit consists of two metallic cones, the sampling cone (orifice radius about 0.5 to 1mm) and the skimmer cone (orifice radius about 0.5 to 1mm), and a rotary

gear pump ventilates between the two into several hundreds Pa condition. The path of the ions pulled through by the sampling cone and the skimmer cone converge into the mass spectrophotometer through the ion lens. The ion lens and the mass spectrophotometer unit are ventilated to 10-3 and 10-4 Pa respectively, by the turbo molecular pump. The ions sorted by mass with the mass spectrophotometer are detected by the ion detector.

ICP-OES

Ion Chromatography (IC / IC-MS)
IC-MS and IC-ICP/MS coupling

Combining IC with an electrospray ionization mass spectrometer (ESI-MS) or an inductively coupled plasma mass spectrometer (ICP-MS) solves even complex separation problems while achieving outstanding sensitivities and selectivities. These so-called hyphenated techniques provide valuable information for unambiguous peak identification and are less prone to matrix influences than IC with conductivity detection.

LC-MS

Liquid chromatography - Mass spectrometry (LC-MS)
This technique combines the physical separation capabilities of liquid chromatography with the mass analysis capabilities of mass spectrometry.

Liquid Chromatography - Mass Spectrometry (LC-MS) is a powerful analytical technique used in industries requiring very low detection limits of sometimes unknown samples, such as food analysis and pharmaceutical drug development. The efficient physical separation of chemical substances dissolved in a mobile phase, performed by liquid chromatography, is combined with the mass spectrometer being able to sort and identify the components (gaseous ions) in electric and magnetic fields according to their mass-to-charge ratios. The samples analysed by LC-MS are often complex mixtures.

Liquid Chromatography (LC)

Liquid chromatography
Liquid chromatography (LC) is a separation technique in which the mobile phase is a liquid, where sample ions or molecules are dissolved.

It is carried out either in a column or a plane. The sample with the mobile liquid will pass through the column or the plane, which is packed with a stationary phase composed of irregularly or spherically shaped particles. Due to the differences in ion-exchange, adsorption, partitioning, or size, different solutes will interact with the stationary phase to different degrees, and therefore the separation of the compounds can be achieved and the transit time of the solutes through the column can be determined by utilizing these differences.

Conventional LC is commonly used in preparative scale work to purify and isolate some components of a mixture. Nowadays liquid chromatography generally utilizes very small packing particles and a relatively high pressure for analytical separations of solutions, detection & quantification, referred to as high performance liquid chromatography (HPLC). HPLC can provide a very high resolution (up to parts per trillion) and a fast analysis time.

MALDI-TOF

Mass spectrometry is an analytical technique in which chemical compounds are ionized into charged molecules and ratio of their mass to charge (m/z) is measured. Though MS was discovered in the early 1900s, its scope was limited to the chemical sciences. However, the development of electron spray ionization (ESI) and matrix assisted laser desorption ionization (MALDI) in 1980s increased the applicability of MS to large biological molecules like proteins. In both ESI and MALDI, peptides are converted into ions by either addition or loss of one or more than one protons. Both are based on “soft ionization” methods where ion formation does not lead to a significant loss of sample integrity. MALDI-TOF MS has certain advantages over ESI-MS viz. (i) MALDI-TOF MS produces singly charged ions, thus interpretation of data is easy comparative to ESI-MS, (ii) for analysis by ESI-MS, prior separation by chromatography is required which is not needed for MALDI-TOF MS analysis. Consequently, the high throughput and speed associated with complete automation has made MALDI-TOF mass spectrometer an obvious choice for proteomics work on large-scale.

Mass Spectrometry (MS) : An analytical technique that is used to measure the mass-to-charge ratio of ions. The results are typically presented as a mass spectrum, a plot of intensity as a function of the mass-to-charge ratio. Mass spectrometry is used in many different fields and is applied to pure samples as well as complex mixtures.

Proteomics

The large-scale study of proteins. Proteins are vital parts of living organisms, with many functions. The proteome is the entire set of proteins that is produced or modified by an organism or system. Proteomics has enabled the identification of ever increasing numbers of protein. This varies with time and distinct requirements, or stresses, that a cell or organism undergoes. Proteomics is an interdisciplinary domain that has benefitted greatly from the genetic information of various genome projects, including the Human Genome Project. It covers the exploration of proteomes from the overall level of protein composition, structure, and activity. It is an important component of functional genomics.

Reversed-Phase Chromatography

Reversed-phase chromatography has become the most widely-used method for purification and analysis of peptides owing to its powerful resolving capability, reproducibility and recovery [1,2] even at ultramicroanalytical levels. The purification factor achievable with reversed-phase chromatography ranges from 2 to 200. In comparison the purification factor of size exclusion ranges from 2 to 20, of ion exchange chromatography from 2 to 40 and of hydrophobic interaction chromatography from 2 to 30. The major advantage of reversed-phase chromatography, apart from its resolving capability, is the availability and use of volatile mobile phases (e.g. aqueous trifluoracetic acid–acetonitrile systems) which do away with the need for sample desalting.

SIFT-MS

SIFT-MS is an ideal tool for high-throughput and real-time gas analysis applications.

Selected ion flow tube mass spectrometry (SIFT-MS) is a form of direct mass spectrometry that analyzes trace amounts of volatile organic compounds (VOCs) and inorganic gases in air. Real-time, quantitative analysis is achieved by applying precisely controlled soft chemical ionization and eliminating sample preparation, pre-concentration and chromatography.

There are three elements of the SIFT-MS technique:

Reagent ion generation and selection. The eight SIFT-MS reagent ions – H_3O^+, NO^+, O_2^+, O^-, O_2^-, OH^-, NO_2^-, and NO_3^- – are all formed by microwave discharge through moist or dry air. The reagent ion of choice is then selected using a quadrupole mass filter.

Analyte ionization. The selected reagent ion is injected into the flow tube and excess energy is removed through collisions with the carrier gas. The sample is then introduced at a known flow rate and the reactive compounds it contains are ionized by the reagent ion to form well-characterized product ions.

Analyte quantitation. Product ions and unreacted reagent ions are sampled into a second quadrupole mass spectrometer. Utilizing a compound library, the software instantaneously calculates each analyte's absolute concentration.

BENEFITS OF SIFT-MS INCLUDE:

- Instantaneous, quantitative analysis of air and headspace with very high sensitivity and selectivity.
- Simultaneous analysis of chemically diverse VOCs (e.g. aldehydes, amines and organosulfurs).
- Direct analysis of high humidity samples.
- Simplicity of operation.
- Simple integration with existing infrastructure.
- Low maintenance and long-term stabilit

Spectroscopy Atomic

Atomic spectroscopy includes a number of analytical techniques used to determine the elemental composition of a sample (it can be gas, liquid, or solid) by observing its electromagnetic spectrum or its mass spectrum. Element concentrations of a millionth (ppm) or one billionth part (ppb) of the sample can be detected. There are different variations of atomic spectroscopy, emission, absorption, fluorescence, and mass spectroscopy. Determination of an appropriate technique requires a basic understanding of each technique since each has its individual strengths and limitations. This chapter is designed to provide a basic overview to the atomic spectroscopy techniques and how can you select the one that best suits our analytical problems.

Spectroscopy ICP

Principle of ICP Optical Emission Spectrometry (ICP-OES)

It has been 25 years since ICP optical emission spectrophotometers (ICP-OES) began to be widely used, and is now one of the most versatile methods of inorganic analysis. Its features are often compared to atomic absorption spectrophotometers. Compared to atomic absorption spectrophotometers, in which the excitation temperature of air-acetylene flame measures 2000 to 3000 K, the excitation temperature of argon ICP is 5000 to 7000 K, which efficiently excites many elements. Also, using inert gas (argon) makes oxides and nitrides harder to be generated.

ICP Optical Emission Spectrometry Principle

ICP, abbreviation for Inductively Coupled Plasma, is one method of optical emission spectrometry. When plasma energy is given to an analysis sample from outside, the component elements (atoms) are excited. When the excited atoms return to low energy position, emission rays (spectrum rays) are released and the emission rays that correspond to the photon wavelength are measured. The element type is determined based on the position of the photon rays, and the content of each element is determined based on the rays' intensity.
To generate plasma, first, argon gas is supplied to torch coil, and high frequency electric current is applied to the work coil at the tip of the torch tube. Using the electromagnetic field created in the torch tube by the high frequency current, argon gas is ionized and plasma is generated. This plasma has high electron density and temperature (10000K) and this energy is used in the excitation-emission of the sample. Solution samples are introduced into the plasma in an atomized state through the narrow tube in the center of the torch tube.

Analytical Chemical Features of ICP-OES

The following features of ICP-OES distinguish it from atomic absorption spectrophotometers used for similar purposes.

- Simultaneous, sequential analysis of multiple elements possible

- Wide linear region of analytical curve
- Few chemical interference or ionization interference, making analysis of high-matrix samples possible
- High sensitivity (low limit of detection for majority of elements is 10ppb or lower)
- High number of measurable elements - elements that are difficult to analyze in atomic absorption spectrometry such as Zr, Ta, rare earth, P and B can be easily analyzed
- Stable

The majority of the above features are derived from the structure and characteristics of the light source plasma.

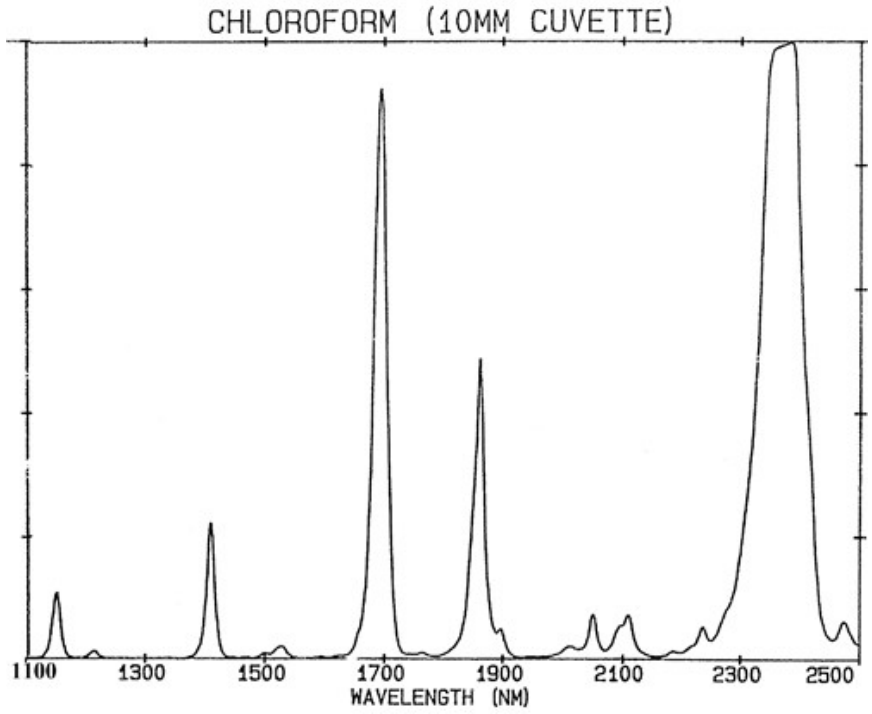
Spectroscopy IR NIR

The first (near) infrared spectra were measured in 1881 by Abney and Festing using photographic plates. Not only did they produce the first spectra but they also suggested, correctly, that the absorptions were related to the chemical composition of the liquids they investigated. The most important pioneer of IR spectroscopy was William W. Coblentz. In 1905 he published the result of a large study of compounds whose spectra he had recorded from 1000 nm to 16,000 nm. Coblentz's work was a breakthrough in that researchers were able to relate the character of groups of atoms within molecules as being related to specific absorptions in the mid-IR (2500–50,000 nm). These absorptions are the result of interactions with the fundamental vibrations of the chemical bonds associated with the atoms of the groups. We can think of chemical bonds as weak springs holding together two or more atoms, these springs will vibrate naturally and when energy is added to the system then they will vibrate more energetically. However, atoms in molecules are constrained by quantum mechanics so that only a few specific energy levels are allowed. If we have only two atoms then the only vibration will be seen as a stretching. When three or more atoms are involved then bonds can also bend, giving rise to a whole series of different vibrations. Stretch vibrations require more energy than bending vibrations but there will also be variation in the energy requirements of the bending vibrations. Different chemical bonds (like O–H, C–H and N–H) vary in strength and hence the amount of energy required for the bond vibration to move from one level to the next. This variation in energy will be seen in a spectrum as a series of absorptions at different wavelengths. By looking at the spectrum we can deduce what vibrations are occurring and hence work out the structure of the molecule (or groups of atoms present).

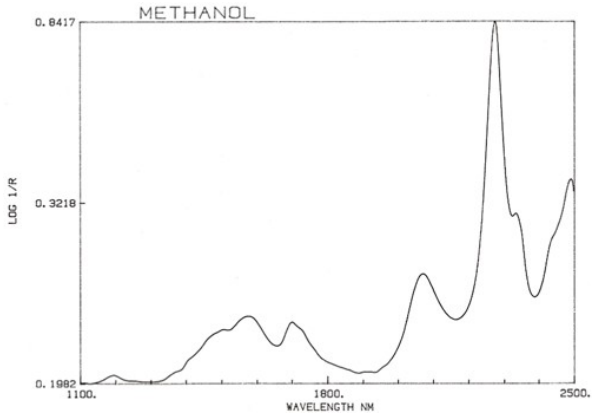
One of the very useful properties of mid-IR spectra is that the region from 8500 nm to 12,500 nm is very characteristic for the molecule measured and this region is known as the "finger-print" region because it can be used to confirm the identity of many pure substances. While the study of mid-IR spectroscopy continued to grow, especially after World War II, interest in the NIR extended to quantitative measurements of water, a few simple organic compounds and a very few studies of specific proteins. No one considered it useful for characterising samples and it was considered too complex for use in quantitative analysis.

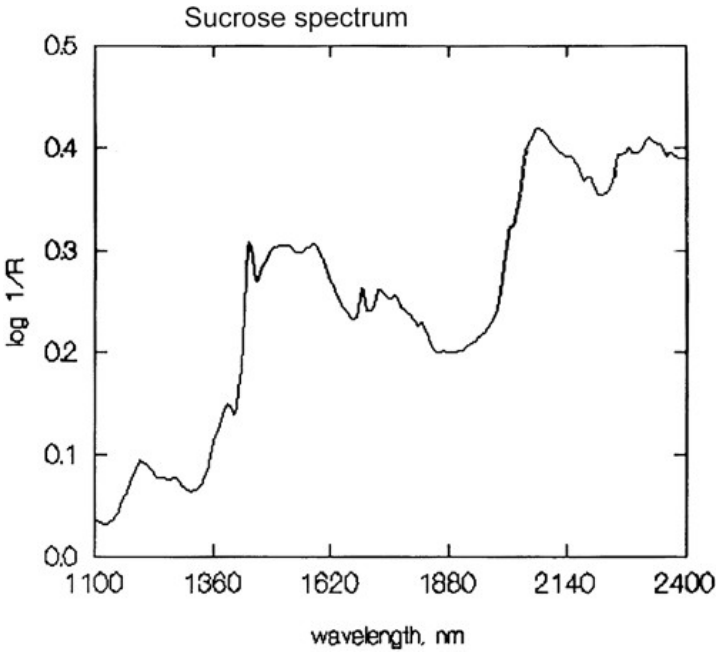
Absorptions in the NIR region

If chemical bonds behaved exactly like weak springs then quantum mechanics would restrict their vibration to just two states and there would be very few absorptions in the NIR region. Absorptions in the NIR region (780–2500 nm) are generated from fundamental vibrations by two processes; overtones and combinations. Overtones can be thought of as harmonics. So every fundamental will produce a series of absorptions at (approximately integer) multiples of the frequency (frequency is the reciprocal of wavelength). Combinations are rather more complex. NIR absorptions are at a higher state of excitement so they require more energy than a fundamental absorption. Combinations arise from the sharing of NIR energy between two or more fundamental absorptions. While the number of possible overtones from a group of fundamental absorptions in a molecule are limited to a few, a very large number of combinations will be observed. The effect of all these absorptions combine to make many NIR spectra to look rather uninteresting and to consist of only a few rather broad peaks. Figure 2 is an NIR spectrum of chloroform, $CHCl_3$, the molecule contains only one hydrogen atom but all the absorption in its spectrum are caused by this single atom.

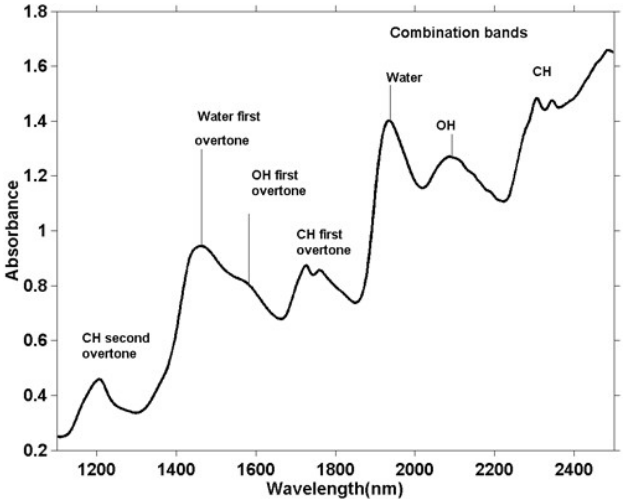


It is an important generalisation that NIR spectroscopy is dominated by hydrogen. Figure 3 is a spectrum of methanol, CH_3OH, which contains four hydrogen atoms (but three are equivalent) and this spectrum is much more like a typical NIR spectrum with broad peaks. Figure 4 is a spectrum of sucrose, $C_{12}H_{24}O_{12}$, which shows very broad areas of absorption but also some quite narrow peaks. It is important to realise that all of these broad absorptions are caused by multiple narrow, over-lapping absorptions. NIR spectra are much more complex than they appear.





Although NIR spectra are more complicated it is possible to make some general observations. As a fundamental O–H stretching absorption is different to a fundamental C–H stretch then the series of overtones generated by these absorptions will also be different. The same goes for combination bands. The most common (and energetic) combination bands arise from stretch and bend combinations in the same group. So we see absorptions due the combination of O–H stretch with O–H bend and C–H stretch with C–H bend and these occur in different positions in the spectrum. Figure 5 is the NIR spectrum of a sample of biscuit dough. Biscuit dough contains several ingredients each of which contain many different molecules so this spectrum contains hundreds if not thousands of absorptions but we see the integration of them all and there appear to be just a few broad absorptions. From their position we can say in general terms the cause of the absorption, as indicated on the figure.



When the complexity of NIR absorption was first realised and compared to the relatively more easily understood mid-IR spectra, it was thought by most researchers that there was little to be gained by studying NIR spectroscopy. The region became neglected and students were wrongly instructed that there was nothing to be gained by studying the NIR region. Many students are still being taught the same opinion. The requirements were: very low noise spectrometers, the electronic computer, the application of mathematical techniques (chemometrics) and a genius to bring it all together. The man was Karl Norris; an engineer working for the USDA at Beltsville. He had not been taught spectroscopy so he did not know that there was nothing to be gained in the NIR region. So, rather like Herschel who looked for something where there was nothing, Norris developed the instruments and utilised computers to demonstrate that the NIR region **was** very useful for quantitative analysis, particularly of agricultural samples. One of the reasons why NIR analysis is so useful is that it can use

reflected energy and this means that NIR analysis can be done with little or no sample preparation. Reflected energy is complex. First, because there are two components, specular (or mirror-like) and diffuse. In the context of NIR spectroscopy, the specular component does not give any information. The diffuse component depends on the physical nature of the sample; particle size being particularly important. Variation of the physical parameters of a sample causes changes in the spectrum so that the observed spectrum is a mixture of chemical and physical information.

The use of reflected energy was forced on Karl Norris. While it makes possible the NIR analysis of a very much wider range of samples, it does add another layer of complication. A complete mathematical theory of reflection spectroscopy is not yet available but it has been found possible by good experimental practice and the utilisation of mathematical techniques to use NIR reflection spectroscopy for analytical chemistry. Because the technique can be applied with little or no sample preparation, analysis times are reduced from hours to minutes and furthermore several analytical results can be obtained from the same NIR data while the conventional analysis would often require another technique and more hours of work. It is, however, necessary to develop calibrations which require many samples, many hours of work and thousands (or probably millions) of computer calculations. With these sorts of attributes it is not surprising that 40 years after the ground-breaking research, a very wide range of analysis can be achieved by NIR spectroscopy.

Spectroscopy NMR

Nuclear Magnetic Resonance Spectroscopy

Over the past fifty years nuclear magnetic resonance spectroscopy, commonly referred to as nmr, has become the preeminent technique for determining the structure of organic compounds. Of all the spectroscopic methods, it is the only one for which a complete analysis and interpretation of the entire spectrum is normally expected. Although larger amounts of sample are needed than for mass spectroscopy, nmr is non-destructive, and with modern instruments good data may be obtained from samples weighing less than a milligram. **To be successful in using nmr as an analytical tool, it is necessary to understand the physical principles on which the methods are based**.

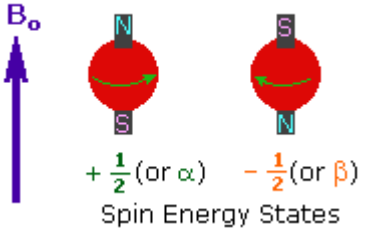
The nuclei of many elemental isotopes have a characteristic spin (**I**). Some nuclei have integral spins (e.g. I = 1, 2, 3 ....), some have fractional spins (e.g. I = 1/2, 3/2, 5/2 ....), and a few have no spin, I = 0 (e.g. ^{12}C, ^{16}O, ^{32}S, ...). Isotopes of particular interest and use to organic chemists are ^{1}H, ^{13}C, ^{19}F and ^{31}P, all of which have I = 1/2. Since the analysis of this spin state is fairly straightforward, our discussion of nmr will be limited to these and other I = 1/2 nuclei.

The following features lead to the nmr phenomenon:

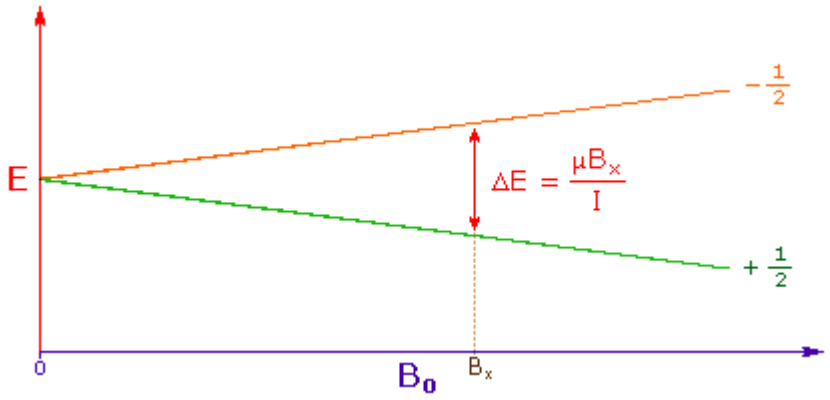
1. A spinning charge generates a magnetic field, as shown by the animation on the right. The resulting spin-magnet has a magnetic moment (**μ**) proportional to the spin.

2. In the presence of an external magnetic field ($\mathbf{B_0}$), two spin states exist, **+1/2** and **-1/2**.

The magnetic moment of the lower energy +1/2 state is aligned with the external field, but that of the higher energy -1/2 spin state is opposed to the external field. Note that the arrow representing the external field points North.



3. The difference in energy between the two spin states is dependent on the external magnetic field strength, and is always very small. The following diagram illustrates that the two spin states have the same energy when the external field is zero, but diverge as the field increases. At a field equal to B_x a formula for the energy difference is given (remember I = 1/2 and μ is the magnetic moment of the nucleus in the field).

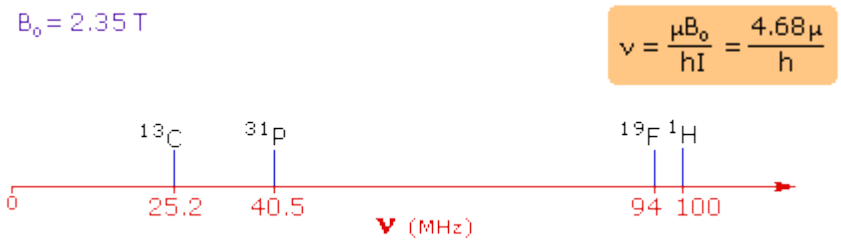


Strong magnetic fields are necessary for nmr spectroscopy. The international unit for magnetic flux is the tesla (**T**). The earth's magnetic field is not constant, but is approximately 10^{-4} T at ground level. Modern nmr spectrometers use powerful magnets having fields of 1 to 20 T. Even with these high fields, the energy difference between the two spin states is less than 0.1 cal/mole. To put this in perspective, recall that infrared transitions involve 1 to 10 kcal/mole and electronic transitions are nearly 100 time greater.

For nmr purposes, this small energy difference (ΔE) is usually given as a frequency in units of MHz (10^6 Hz), ranging from 20 to 900 Mz, depending on the magnetic field strength and the specific nucleus being studied. Irradiation of a sample with radio frequency (rf) energy corresponding exactly to the spin state separation of a specific set of nuclei will cause excitation of those nuclei in the +1/2 state to the higher -1/2 spin state. Note that this electromagnetic radiation falls in the radio and television broadcast spectrum. Nmr spectroscopy is therefore the energetically mildest probe used to examine the structure of molecules.

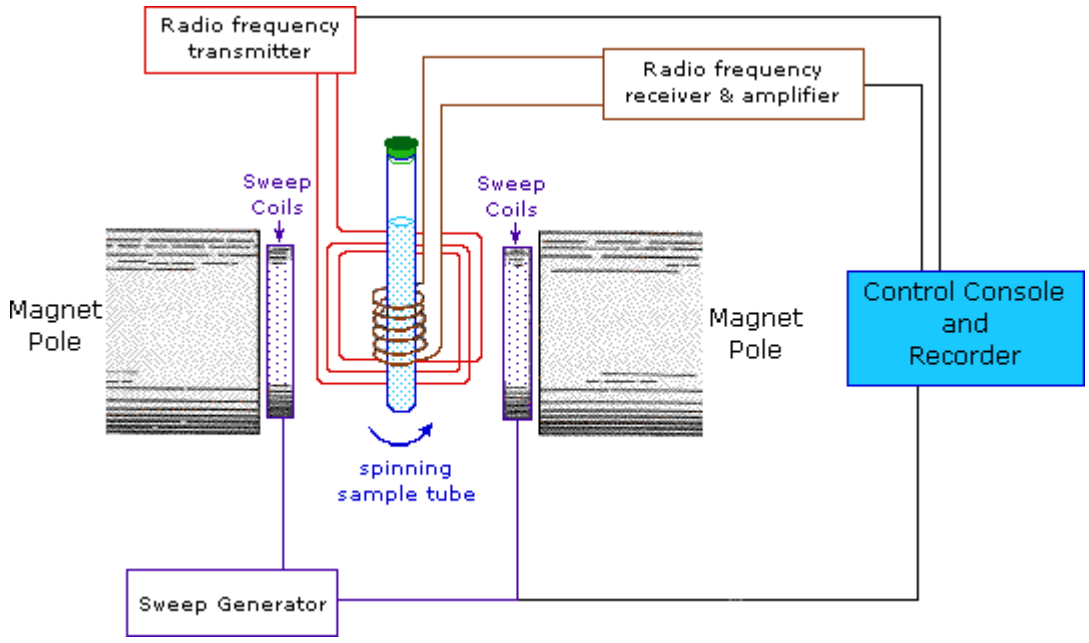
The nucleus of a hydrogen atom (the proton) has a magnetic moment μ = 2.7927, and has been studied more than any other nucleus. The previous diagram may be changed to display energy differences for the proton spin states (as frequencies) by mouse clicking anywhere within it.

4. For spin 1/2 nuclei the energy difference between the two spin states at a given magnetic field strength will be proportional to their magnetic moments. For the four common nuclei noted above, the magnetic moments are: ^{1}H μ = 2.7927, ^{19}F μ = 2.6273, ^{31}P μ = 1.1305 & ^{13}C μ = 0.7022. These moments are in nuclear magnetons, which are $5.05078 \cdot 10^{-27}$ JT^{-1}. The following diagram gives the approximate frequencies that correspond to the spin state energy separations for each of these nuclei in an external magnetic field of 2.35 T. The formula in the colored box shows the direct correlation of frequency (energy difference) with magnetic moment (h = Planck's constant = $6.626069 \cdot 10^{-34}$ Js).

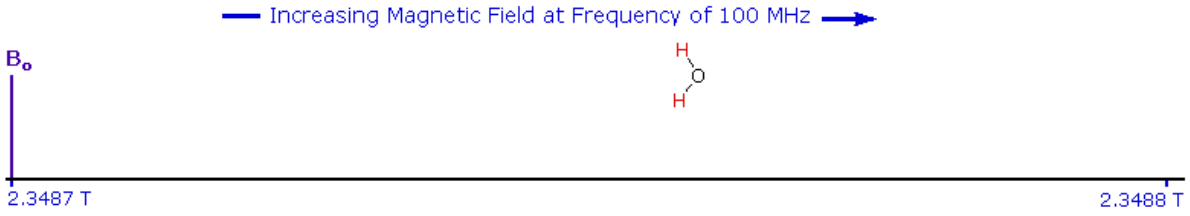


Proton NMR Spectroscopy

This important and well-established application of nuclear magnetic resonance will serve to illustrate some of the novel aspects of this method. To begin with, the nmr spectrometer must be tuned to a specific nucleus, in this case the proton. The actual procedure for obtaining the spectrum varies, but the simplest is referred to as the **continuous wave** (CW) method. A typical CW-spectrometer is shown in the following diagram. A solution of the sample in a uniform 5 mm glass tube is oriented between the poles of a powerful magnet, and is spun to average any magnetic field variations, as well as tube imperfections. Radio frequency radiation of appropriate energy is broadcast into the sample from an antenna coil (colored red). A receiver coil surrounds the sample tube, and emission of absorbed rf energy is monitored by dedicated electronic devices and a computer. An nmr spectrum is acquired by varying or sweeping the magnetic field over a small range while observing the rf signal from the sample. An equally effective technique is to vary the frequency of the rf radiation while holding the external field constant.



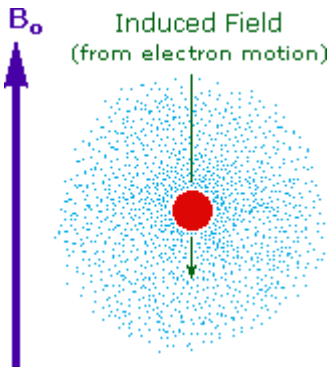
As an example, consider a sample of water in a 2.3487 T external magnetic field, irradiated by 100 MHz radiation. If the magnetic field is smoothly increased to 2.3488 T, the hydrogen nuclei of the water molecules will at some point absorb rf energy and a resonance signal will appear. An animation showing this may be activated by clicking the **Show Field Sweep** button. The field sweep will be repeated three times, and the resulting resonance trace is colored red. For visibility, the water proton signal displayed in the animation is much broader than it would be in an actual experiment.



Since protons all have the same magnetic moment, we might expect all hydrogen atoms to give resonance signals at the same field / frequency values. Fortunately for chemistry applications, this is not true. By clicking the **Show Different Protons** button under the diagram, a number of representative proton signals will be displayed over the same magnetic field range. It is not possible, of course, to examine isolated protons in the spectrometer described above; but from independent measurement and calculation it has been determined that a naked proton would resonate at a lower field strength than the nuclei of covalently bonded

hydrogens. With the exception of water, chloroform and sulfuric acid, which are examined as liquids, all the other compounds are measured as gases.

Why should the proton nuclei in different compounds behave differently in the nmr experiment ?

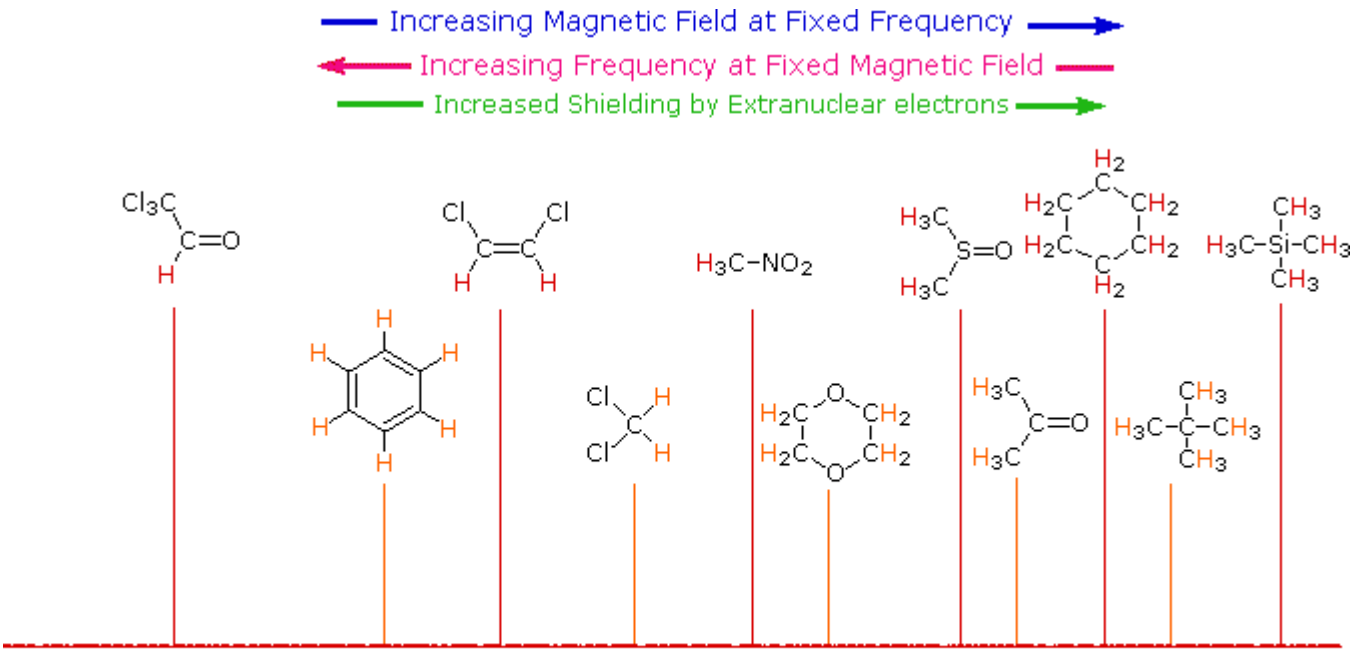


The answer to this question lies with the electron(s) surrounding the proton in covalent compounds and ions. Since electrons are charged particles, they move in response to the external magnetic field (B_0) so as to generate a secondary field that opposes the much stronger applied field. This secondary field **shields** the nucleus from the applied field, so B_0 must be increased in order to achieve resonance (absorption of rf energy). As illustrated in the drawing on the right, B_0 must be increased to compensate for the induced shielding field. In the upper diagram, those compounds that give resonance signals at the higher field side of the diagram (CH_4, HCl, HBr and HI) have proton nuclei that are more shielded than those on the lower field (left) side of the diagram.

The magnetic field range displayed in the above diagram is very small compared with the actual field strength (only about 0.0042%). It is customary to refer to small increments such as this in units of **parts per million** (ppm). The difference between 2.3487 T and 2.3488 T is therefore about 42 ppm. Instead of designating a range of nmr signals in terms of magnetic field differences (as above), it is more common to use a frequency scale, even though the spectrometer may operate by sweeping the magnetic field. Using this terminology, we would find that at 2.34 T the proton signals shown above extend over a 4,200 Hz range (for a 100 MHz rf frequency, 42 ppm is 4,200 Hz). Most organic compounds exhibit proton resonances that fall within a 12 ppm range (the shaded area), and it is therefore necessary to use very sensitive and precise spectrometers to resolve structurally distinct sets of hydrogen atoms within this narrow range. In this respect it might be noted that the detection of a part-per-million difference is equivalent to detecting a 1 millimeter difference in distances of 1 kilometer.

Chemical Shift

Unlike infrared and uv-visible spectroscopy, where absorption peaks are uniquely located by a frequency or wavelength, the location of different nmr resonance signals is dependent on both the external magnetic field strength and the rf frequency. Since no two magnets will have exactly the same field, resonance frequencies will vary accordingly and an alternative method for characterizing and specifying the location of nmr signals is needed. This problem is illustrated by the eleven different compounds shown in the following diagram. Although the eleven resonance signals are distinct and well separated, an unambiguous numerical locator cannot be directly assigned to each.



^{1}H NMR Resonance Signals for some Different Compounds

One method of solving this problem is to report the location of an nmr signal in a spectrum relative to a reference signal from a standard compound added to the sample. Such a reference standard should be chemically unreactive, and easily removed from the sample after the measurement. Also, it should give a single sharp nmr signal that does not interfere with the resonances normally observed for organic compounds. **Tetramethylsilane**, $(CH_3)_4Si$, usually referred to as **TMS**, meets all these characteristics, and has become the reference compound of choice for proton and carbon nmr.

Since the separation (or dispersion) of nmr signals is magnetic field dependent, one additional step must be taken in order to provide an unambiguous location

unit. This is illustrated for the acetone, methylene chloride and benzene signals by clicking on the previous diagram. To correct these frequency differences for their field dependence, we divide them by the spectrometer frequency (100 or 500 MHz in the example), as shown in a new display by again clicking on the diagram. The resulting number would be very small, since we are dividing Hz by MHz, so it is multiplied by a million, as shown by the formula in the blue shaded box. Note that ν_{ref} is the resonant frequency of the reference signal and ν_{samp} is the frequency of the sample signal. This operation gives a locator number called the **Chemical Shift**, having units of parts-per-million (ppm), and designated by the symbol **δ** Chemical shifts for all the compounds in the original display will be presented by a third click on the diagram.

The compounds referred to above share two common characteristics:

- The hydrogen atoms in a given molecule are all structurally equivalent, averaged for fast conformational equilibria.
- The compounds are all liquids, save for neopentane which boils at 9 °C and is a liquid in an ice bath.

The first feature assures that each compound gives a single sharp resonance signal. The second allows the pure (neat) substance to be poured into a sample tube and examined in a nmr spectrometer. In order to take the nmr spectra of a solid, it is usually necessary to dissolve it in a suitable solvent. Early studies used carbon tetrachloride for this purpose, since it has no hydrogen that could introduce an interfering signal. Unfortunately, CCl_4 is a poor solvent for many polar compounds and is also toxic. Deuterium labeled compounds, such as deuterium oxide (D_2O), chloroform-d ($DCCl_3$), benzene-d_6 (C_6D_6), acetone-d_6 (CD_3COCD_3) and DMSO-d_6 (CD_3SOCD_3) are now widely used as nmr solvents. Since the deuterium isotope of hydrogen has a different magnetic moment and spin, it is invisible in a spectrometer tuned to protons.

Spectroscopy Raman

Raman Spectroscopy is a non-destructive chemical analysis technique which provides detailed information about chemical structure, phase and polymorphy, crystallinity and molecular interactions. It is based upon the interaction of light with the chemical bonds within a material.
Raman is a light scattering technique, whereby a molecule scatters incident light from a high intensity laser light source. Most of the scattered light is at the same wavelength (or color) as the laser source and does not provide useful information – this is called Rayleigh Scatter. However a small amount of light (typically 0.0000001%) is scattered at different wavelengths (or colors), which depend on the chemical structure of the analyte – this is called Raman Scatter.

A Raman spectrum features a number of peaks, showing the intensity and wavelength position of the Raman scattered light. Each peak corresponds to a specific molecular bond vibration, including individual bonds such as C-C, C=C, N-O, C-H etc., and groups of bonds such as benzene ring breathing mode, polymer chain vibrations, lattice modes, etc.

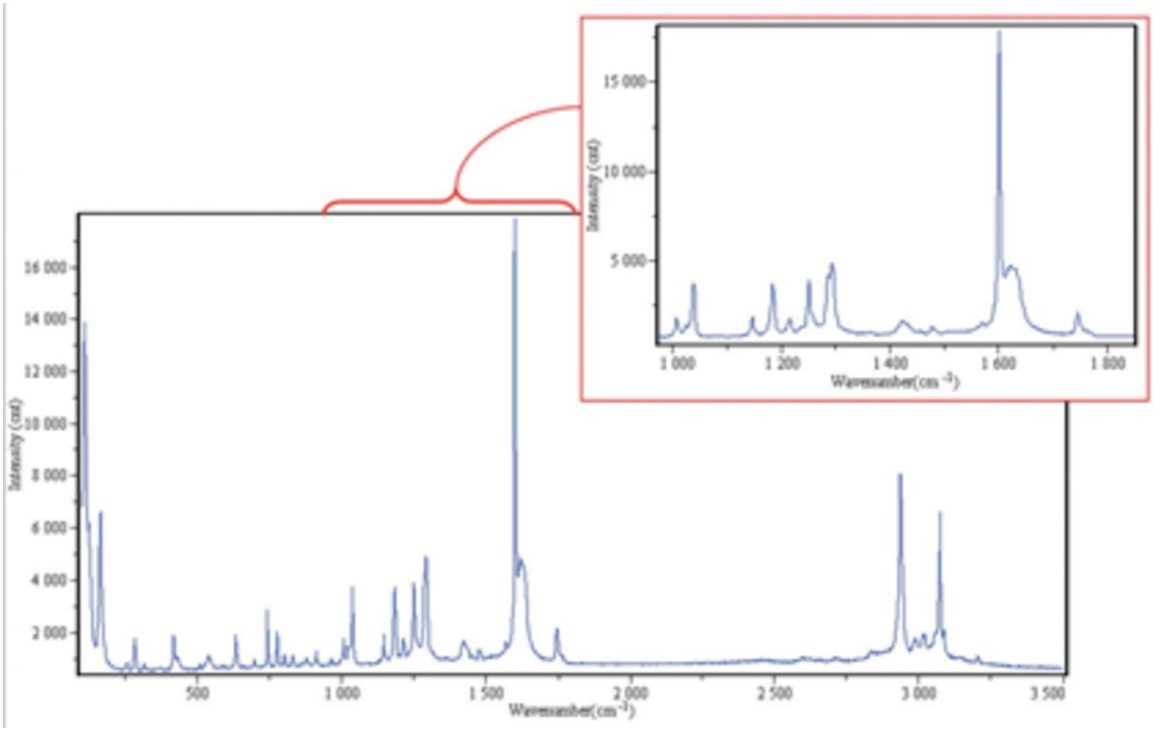


Fig. 2: A typical Raman spectrum, in this case, of aspirin (4-acetylsalicylic acid). The inset image shows the detail which is present in the spectrum

Information provided by Raman spectroscopy

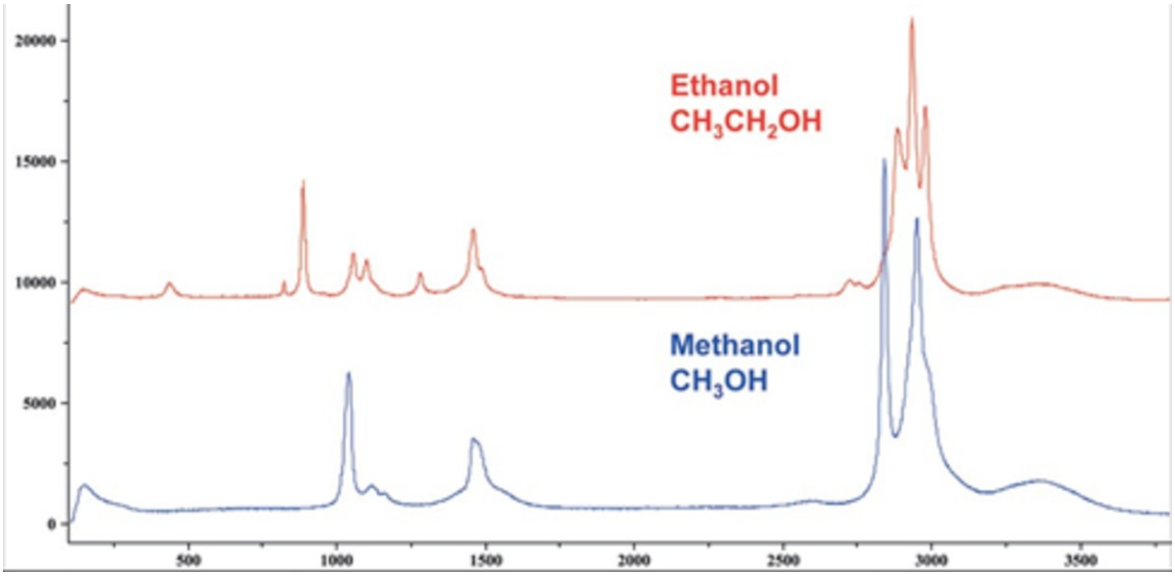


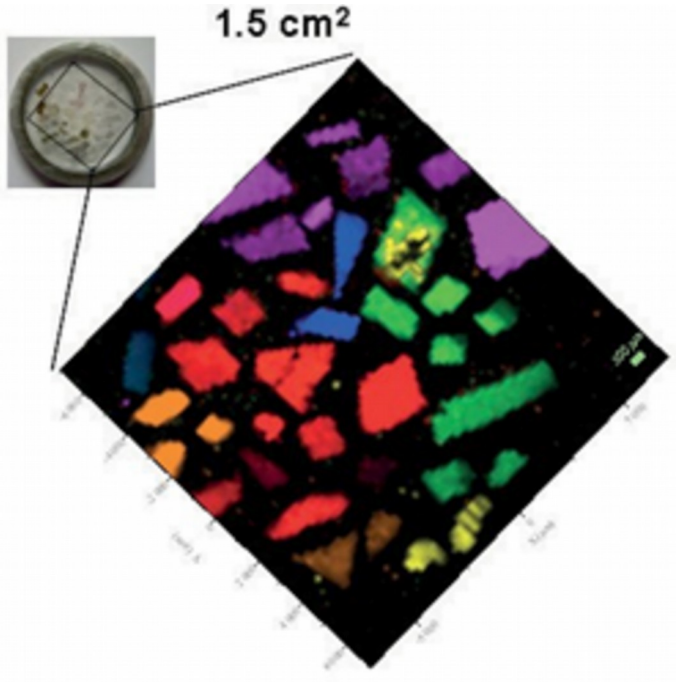
Fig. 3: Raman spectra of ethanol and methanol, showing the significant spectral differences which allow the two liquids to be distinguished.

Raman spectroscopy probes the chemical structure of a material and provides information about:

- Chemical structure and identity
- Phase and polymorphism
- Intrinsic stress/strain
- Contamination and impurity

Typically a Raman spectrum is a distinct chemical fingerprint for a particular molecule or material, and can be used to very quickly identify the material, or distinguish it from others. Raman spectral libraries are often used for identification of a material based on its Raman spectrum – libraries containing thousands of spectra are rapidly searched to find a match with the spectrum of the analyte.

Fig. 4: Mineral distribution



In combination with mapping (or imaging) Raman systems, it is possible to generate images based on the sample's Raman spectrum. These images show distribution of individual chemical components, polymorphs and phases, and variation in crystallinity.

Raman spectroscopy is both qualitative and quantitative.
The general spectrum profile (peak position and relative peak intensity) provides a unique chemical fingerprint which can be used to identify a material, and distinguish it from others. Often the actual spectrum is quite complex, so comprehensive Raman spectral libraries can be searched to find a match, and thus provide a chemical identification.

The intensity of a spectrum is directly proportional to concentration. Typically, a calibration procedure will be used to determine the relationship between peak intensity and concentration, and then routine measurements can be made to analyze for concentration. With mixtures, relative peak intensities provide information about the relative concentration of the components, while absolute peak intensities can be used for absolute concentration information.

Spectroscopy UV VIS

UV-Vis Spectroscopy (or Spectrophotometry) is a quantitative technique used to measure how much a chemical substance absorbs light. This is done by measuring the intensity of light that passes through a sample with respect to the intensity of light through a reference sample or blank. This technique can be used for multiple sample types including liquids, solids, thin-films and glass.

Spectrophotometry is a quantitative measurement of the absorption/transmission or reflection of a material as a function of wavelength. Despite being termed UV-Vis, the wavelength range that is typically used ranges from 190 nm up to 1,100 nm in the near-infrared.

Using a spectrophotometer and carrying out absorption/transmission measurements we can determine the amount (or concentration) of a known chemical substance simply, by studying the number of photons (light intensity) that reach the detector. The more a material absorbs light at a specific wavelength, the higher the concentration of the known substance.

Supercritical Fluid Chromatography (SFC)

The discovery of supercritical fluids led to novel analytical applications in the fields of chromatography and extraction known as supercritical fluid chromatography (SFC) and supercritical fluid extraction (SFE). Supercritical fluid chromatography is accepted as a column chromatography methods along with gas chromatography (GC) and high-performance liquid chromatography (HPLC). Due to to the properties of supercritical fluids, SFC combines each of the advantages of both GC and HPLC in one method. In addition, supercritical fluid extraction is an advanced analytical technique.

Definition and Formation of Supercritical Fluids

A supercritical fluid is the phase of a material at critical temperature and critical pressure of the material. Critical temperature is the temperature at which a gas cannot become liquid as long as there is no extra pressure; and, critical pressure is the minimum amount of pressure to liquefy a gas at its critical temperature. Supercritical fluids combine useful properties of gas and liquid phases, as it can behave like both a gas and a liquid in terms of different aspects. A supercritical fluid provides a gas-like characteristic when it fills a container and it takes the shape of the container. The motion of the molecules are quite similar to gas molecules. On the other hand, a supercritical fluid behaves like a liquid because its density property is near liquid and, thus, a supercritical fluid shows a similarity to the dissolving effect of a liquid.

The characteristic properties of a supercritical fluid are density, diffusivity and viscosity. Supercritical values for these features take place between liquids and gases.

Table 3.3.1 demonstrates numerical values of properties for gas, supercritical fluid and liquid.

Table 3.3.1

	Gas	Supercritical fluid	**Liquid**
Density (g/cm^3)	0.6×10^{-3}-2.0×10^{-3}	0.2-0.5	0.6-2.0
Diffusivity (cm^2/s)	0.1-0.4	10^{-3}-10^{-4}	0.2×10^{-5}-2.0×10^{-5}
Viscosity (cm/s)	1×10^{-4}-3×10^{-4}	1×10^{-4}-3×10^{-4}	0.2×10^{-2}-3.0×10^{-2}

The formation of a supercritical fluid is the result of a dynamic equilibrium. When a material is heated to its specific critical temperature in a closed system, at constant pressure, a dynamic equilibrium is generated. This equilibrium includes

the same number of molecules coming out of liquid phase to gas phase by gaining energy and going in to liquid phase from gas phase by losing energy. At this particular point, the phase curve between liquid and gas phases disappears and supercritical material appears.
In order to understand the definition of SF better, a simple phase diagram can be used. Figure 3.3.1
displays an ideal phase diagram. For a pure material, a phase diagram shows the fields where the material is in the form of solid, liquid, and gas in terms of different temperature and pressure values. Curves, where two phases (solid-gas, solid-liquid and liquid-gas) exist together, defines the boundaries of the phase regions. These curves, for example, include sublimation for solid-gas boundary, melting for solid-liquid boundary, and vaporization for liquid-gas boundary. Other than these binary existence curves, there is a point where all three phases are present together in equilibrium; the triple point (TP).

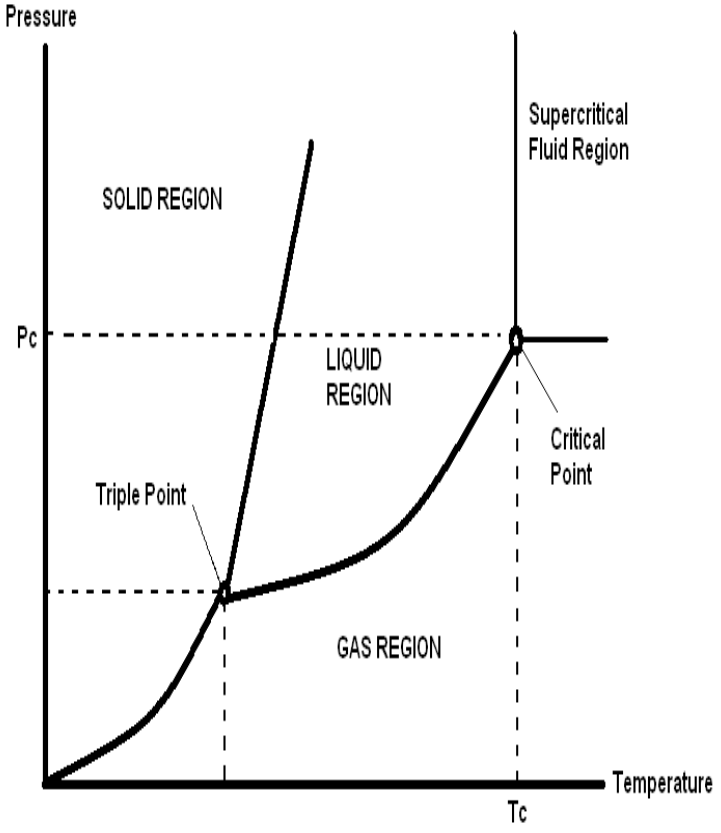


Figure 3.3.1

There is another characteristic point in the phase diagram, the critical point (CP). This point is obtained at critical temperature (T_c) and critical pressure (P_c). After the CP, no matter how much pressure or temperature is increased, the material cannot transform from gas to liquid or from liquid to gas phase. This form is the supercritical fluid form. Increasing temperature cannot result in turning to gas, and

increasing pressure cannot result in turning to liquid at this point. In the phase diagram, the field above T_c and P_c values is defined as the supercritical region.

In theory, the supercritical region can be reached in two ways:

- Increasing the pressure above the P_c value of the material while keeping the temperature stable and then increasing the temperature above T_c value at a stable pressure value.
- Increasing the temperature first above T_c value and then increasing the pressure above P_c value.

The critical point is characteristic for each material, resulting from the characteristic T_c and P_c values for each substance.

Physical Properties of Supercritical Fluids

As mentioned above, SF shares some common features with both gases and liquids. This enables us to take advantage of a correct combination of the properties.

Density

Density characteristic of a supercritical fluid is between that of a gas and a liquid, but closer to that of a liquid. In the supercritical region, density of a supercritical fluid increases with increased pressure (at constant temperature). When pressure is constant, density of the material decreases with increasing temperature. The dissolving effect of a supercritical fluid is dependent on its density value. Supercritical fluids are also better carriers than gases thanks to their higher density. Therefore, density is an essential parameter for analytical techniques using supercritical fluids as solvents.

Diffusivity

Diffusivity of a supercritical fluid can be 100 x that of a liquid and $^1/_{1,000}$ to $^1/_{10,000}$ x less than a gas. Because supercritical fluids have more diffusivity than a liquid, it stands to reason a solute can show better diffusivity in a supercritical fluid than in a liquid. Diffusivity is parallel with temperature and contrary with pressure. Increasing pressure affects supercritical fluid molecules to become closer to each other and decreases diffusivity in the material. The greater diffusivity gives supercritical fluids the chance to be faster carriers for analytical applications. Hence, supercritical fluids play an important role for chromatography and extraction methods.

Viscosity

Viscosity for a supercritical fluid is almost the same as a gas, being approximately $^1/_{10}$ of that of a liquid. Thus, supercritical fluids are less resistant than liquids towards components flowing through. The viscosity of supercritical fluids is also distinguished from that of liquids in that temperature has a little effect on liquid viscosity, where it can dramatically influence supercritical fluid viscosity. These properties of viscosity, diffusivity, and density are related to each other. The change in temperature and pressure can affect all of them in different

combinations. For instance, increasing pressure causes a rise for viscosity and rising viscosity results in declining diffusivity.

Super Fluid Chromatography (SFC)

Just like supercritical fluids combine the benefits of liquids and gases, SFC bring the advantages and strong aspects of HPLC and GC together. SFC can be more advantageous than HPLC and GC when compounds which decompose at high temperatures with GC and do not have functional groups to be detected by HPLC detection systems are analyzed.

There are three major qualities for column chromatographies:

- Selectivity.
- Efficiency.
- Sensitivity.

Generally, HPLC has better selectivity that SFC owing to changeable mobile phases (especially during a particular experimental run) and a wide range of stationary phases. Although SFC does not have the selectivity of HPLC, it has good quality in terms of sensitivity and efficiency. SFC enables change of some properties during the chromatographic process. This tuning ability allows the optimization of the analysis. Also, SFC has a broader range of detectors than HPLC. SFC surpasses GC for the analysis of easily decomposable substances; these materials can be used with SFC due to its ability to work with lower temperatures than GC.

Instrumentation for SFC

As it can be seen in Figure 3.3.2

SFC has a similar setup to an HPLC instrument. They use similar stationary phases with similar column types. However, there are some differences. Temperature is critical for supercritical fluids, so there should be a heat control tool in the system similar to that of GC. Also, there should be a pressure control mechanism, a restrictor, because pressure is another essential parameter in order for supercritical fluid materials to be kept at the required level. A microprocessor mechanism is placed in the instrument for SFC. This unit collects data for pressure, oven temperature, and detector performance to control the related pieces of the instrument.

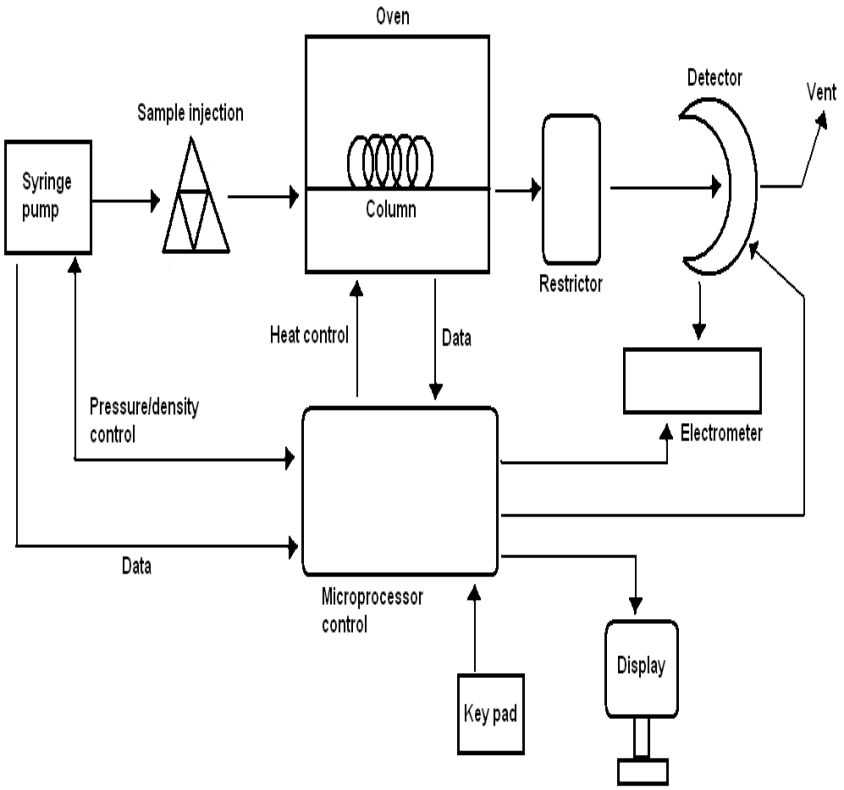


Figure 3.3.2

Stationary Phase

SFC columns are similar to HPLC columns in terms of coating materials. Open-tubular columns and packed columns are the two most common types used in SFC. Open-tubular ones are preferred and they have similarities to HPLC fused-silica columns. This type of column contains an internal coating of a cross-linked siloxane material as a stationary phase. The thickness of the coating can be 0.05-1.0 μm. The length of the column can range from of 10 to 20 m.

Mobile Phases

There is a wide variety of materials used as mobile phase in SFC. The mobile phase can be selected from the solvent groups of inorganic solvents, hydrocarbons, alcohols, ethers, halides; or can be acetone, acetonitrile, pyridine, etc. The most common supercritical fluid which is used in SFC is carbon dioxide because its critical temperature and pressure are easy to reach. Additionally, carbon dioxide is low-cost, easy to obtain, inert towards UV, non-poisonous and a good solvent for non-polar molecules. Other than carbon dioxide, ethane, n-butane, N_2O, dichlorodifluoromethane, diethyl ether, ammonia, tetrahydrofuran can be used. Table 3.3.2 shows select solvents and their T_c and P_c values.

Table 3.3.2

Solvent	**Critical Temperature (°C)**	**Critical Pressure (bar)**
Carbon dioxide (CO_2)	31.1	72
Nitrous oxide (N_2O)	36.5	70.6
Ammonia (NH_3)	132.5	109.8
Ethane (C_2H_6)	32.3	47.6
n-Butane (C_4H_{10})	152	70.6
Diethyl ether (Et_2O)	193.6	63.8
Tetrahydrofuran (THF, C_4H_8O)	267	50.5
Dichlorodifluoromethane (CCl_2F_2)	111.7	109.8

Detectors

One of the biggest advantage of SFC over HPLC is the range of detectors. Flame ionization detector (FID), which is normally present in GC setup, can also be applied to SFC. Such a detector can contribute to the quality of analyses of SFC since FID is a highly sensitive detector. SFC can also be coupled with a mass spectrometer, an UV-visible spectrometer, or an IR spectrometer more easily than can be done with an HPLC. Some other detectors which are used with HPLC can be attached to SFC such as fluorescence emission spectrometer or thermionic detectors.

Advantages of working with SFC

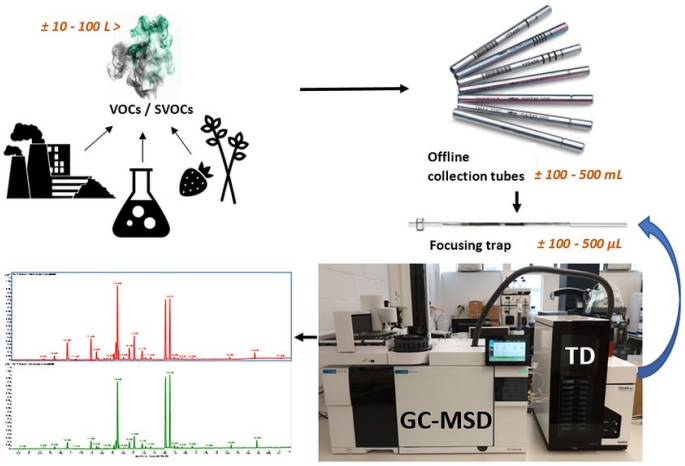
The physical properties of supercritical fluids between liquids and gases enables the SFC technique to combine with the best aspects of HPLC and GC, as lower viscosity of supercritical fluids makes SFC a faster method than HPLC. Lower viscosity leads to high flow speed for the mobile phase.

Thanks to the critical pressure of supercritical fluids, some fragile materials that are sensitive to high temperature can be analyzed through SFC. These materials can be compounds which decompose at high temperatures or materials which have low vapor pressure/volatility such as polymers and large biological molecules. High pressure conditions provide a chance to work with lower temperature than normally needed. Hence, the temperature-sensitive components can be analyzed via SFC. In addition, the diffusion of the components flowing through a supercritical fluid is higher than observed in HPLC due to the higher diffusivity of supercritical fluids over traditional liquids mobile phases. This results in better distribution into the mobile phase and better separation.

Applications of SFC

The applications of SFC range from food to environmental to pharmaceutical industries. In this manner, pesticides, herbicides, polymers, explosives and fossil fuels are all classes of compounds that can be analyzed. SFC can be used to analyze a wide variety of drug compounds such as antibiotics, prostaglandins, steroids, taxol, vitamins, barbiturates, non-steroidal anti-inflammatory agents, etc. Chiral separations can be performed for many pharmaceutical compounds. SFC is dominantly used for non-polar compounds because of the low efficiency of carbon dioxide, which is the most common supercritical fluid mobile phase, for dissolving polar solutes. SFC is used in the petroleum industry for the determination of total aromatic content analysis as well as other hydrocarbon separations.

TD-GC-MS



Analysis of volatile organic compounds using thermal desorption coupled with GC-MSD

The repertoire of mass spectrometry instruments in MC has been expanded by the addition of a gas chromatography mass selective detector (GC-MSD) interfaced with an automated thermal desorption (TD) unit. The instrument will be primarily utilised for monitoring of volatile organic compounds as markers of indoor air quality, as well as conventional GC compatible samples (e.g. volatile, non-polar compounds).
Volatile organic compounds (VOC), semi-volatile organic compounds (SVOC), and very volatile organic compounds (VVOC) refers to any of thousands of organic chemicals that are present as gases at room temperature. VOCs and SVOCs are atmospheric pollutants that can have adverse effects on human health, some of which are known carcinogens. VOC examples include alcohols (cleaning reagents), benzene (petroleum fuel, tobacco smoke), alkanes and alkynes (lubricants) and terpenes (paint and glue). SVOC examples include polycyclic aromatic hydrocarbons (fuel combustion), brominated flame retardants and phthalate esters (consumer products), as well as organochlorine pesticides and pyrethroids (agriculture, insect repellents).

Description	Abbreviation	Boiling point range (°C)	Example compounds
Very volatile (gaseous) organic compounds	VVOC	<0 to 50-100	Propane, butane, methyl chloride
Volatile organic compounds	VOC	50-100 to 240-260	Formaldehyde, d-Limonene, toluene, acetone, ethanol (ethyl alcohol) 2-propanol (isopropyl alcohol), hexanal
Semi volatile organic compounds	SVOC	240-260 to 380-400	Pesticides (DDT, chlordane, plasticizers (phthalates), fire retardants (PCBs, PBB)

Analysis of different VOC classes can be problematic due to their relative low levels of occurrence at sub parts-per-billion (PPB) levels, often in complex mixtures. Thermal desorption (TD) is a very effective technique to up-concentrate these low-level analytes and is therefore often used to monitor a wide range of sample matrices, including environmental monitoring of indoor and outdoor air, water, soil, as well as aroma-profiling in food and drink. The TD analysis is a two-stage process, firstly off-line collecting and concentration of organic vapours from samples onto sorbent-packed collection tubes takes place, after which the compounds are passed on to a secondary focusing trap inside a TD autosampler unit using carrier gas. The secondary trap is then rapidly heated to desorb and pass the volatiles onto the GC column for separation. Large sample volumes, containing a range of target VOCs, are therefore repeatedly extracted/desorbed into smaller volumes of gas, resulting in up to 10^6-fold up concentration, ultimately improving sensitivity and enabling trace-level analysis of target compounds.

Benefits of Thermal Desorption as compared to solvent extraction:

- Efficient extraction – as gas is continuously purging compounds from the sorbent/trap typical desorption, therefore efficiencies of 95% are observed with good reproducibility.
- Minimal sample handling – manual sample preparation is minimised, obviating the need to work with toxic or odorous extraction solvents, and limiting introduction of interfering compounds from pipettes or solvents.

A multitude of versatile configurations/collection sampling methods helps to enable analysis of both liquid- and solid-phase samples. Conventional methods for

sampling onto sorbent tubes includes passive diffusion and active pumping of sampled atmosphere. Sorbent tubes can also be coupled to thermogravimetric analysis (TGA) equipment to allow for the collection of evolved gases during sample heating. Experiments can even be conducted in heated chambers (μ-chambers) specifically designed to collect vapours from samples such as leather, car trim plastics, herbs and spices, and kids' toys, all with the aim of determining what volatile organic compounds are given off by these materials under different environmental conditions.

Absolute quantitation of VOCs can be achieved by injecting authentic gas or liquid standards onto sorbent tubes, allowing for the response factors to be compared with the sample analytes to determine absolute amounts. Moreover, MS/MS spectra can be compared to commercially available fragmentation libraries, such as the National Institute of Standards and Technology (NIST) database, to help identify unknowns. Desorption from the sorbent tubes previously meant only one-shot analyses could be performed, however this shortcoming has been addressed by recollecting vapours from the trap back onto the original or blank sorbent tube, allowing multiple desorption of the same collected sample for multiple analyses using different methods.

Two-dimensional Gas Chromatography (GCxGC)

GCxGC chromatography employs a pair of GC columns each containing a chemical phase that is orthogonal to the other, connected in series via a modulator*. Effluent from the first column is trapped in the modulator for a given period (modulation time) before being focused and released into the second column.
The chromatogram obtained through repeated trapping and injection is rendered in two dimensions using specialized software, with the first and second dimensions on respective axes.

Features

Separation of Overlapping Peaks

Mixtures of compounds with similar boiling points that could not be separated adequately in one-dimensional chromatography may now be separated based on another chemical or physical property, such as polarity. This allows for the analysis of components in complex matrices that previously were difficult to separate.

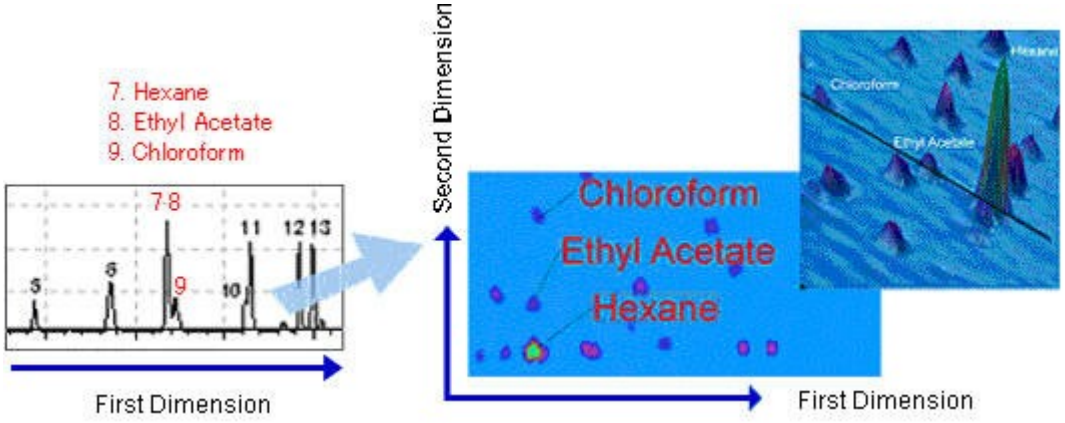
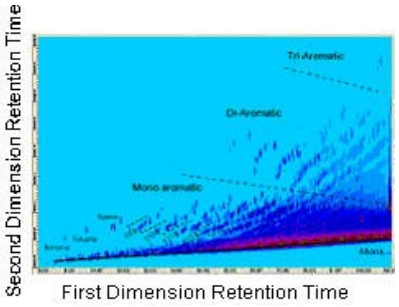
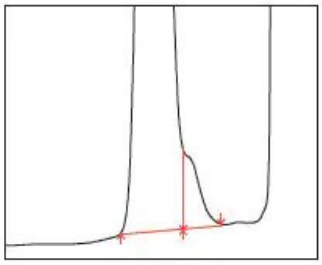


Image Patterns Show Compound Structure

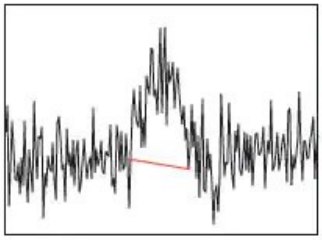
GCxGC provides image patterns that can be correlated to compound class structure. This "fingerprinting" capability is especially useful for quickly characterizing a complex natural product sample such as a crude oil, essential oil or fatty acid extract. These bands of compounds with similar chemical structure are easily grouped using software which allows for the quick determination of the amount of any and all compound classes in a complex sample.



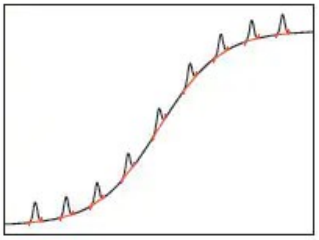
Analysis of Diesel Oil
This image shows the patterns produced by the GCxGC technique. Compounds and organized by carbon number and boiling point on the x axis and by polarity on the y axis.



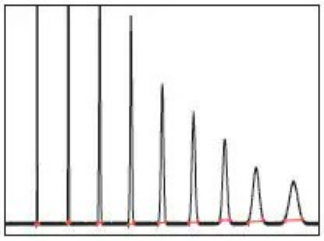
Highly accurate detection of shoulder peaks



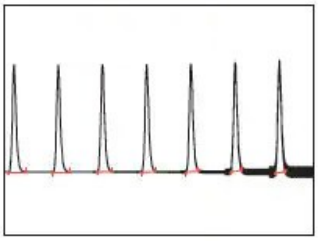
Peak detection at low S/N levels



Automatic determination of baseline undulation and drift



Automatic tracking of peak width variation



Automatic tracking of noise intensity changes

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