Chapter:

Introduction to Analytical Chemistry

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ANALYTICAL CHEMISTRY

✓ CHAP#1
Introduction to Analytical chemistry

Applications of Analytical CHEMISTRY

① Pharmacy:

- Shelf life of a compound → medicine is stored in the Refrigerator or darker places to keep its shelf life.
- Determining Adulterants → Mixing | impurity | doping of something.
- Dissolution in Drugs → whether medicine is released in it form, which can be tablet.
- qualitative & quantitative Analysis:
  ↓ to check the composition and quality of drugs.
  - Derivilization of drugs.

② Agriculture:

- soil testing* → to test the inorganic and organic minerals
- water* testing: → Right water → for perfectly grow your crops
  ↳ composition of water. (mineral contents for plants growth)
- Harvested* Crop testing:
  ↳ After harvesting, undergo quality test inorder to check the impurities, pesticides Residues
- pesticides* study → toxicity of pesticides / Effectiveness

③ Medical technology:
↳ Diagnosis process

- Medical test:
  ↳ cholesterol (serum) → proteins / carbohydrates test
  ↳ ketones in urine

④ Food and Beverages:

- Food Contamination test:
  - ↳ pesticides are contaminents ✓
  - ↳ To detect the contaminent.
- Flavours & Quality
  - → pH, EC, specific gravity, sugars test.
  - → HMF test
  - → Reducing / Non-Reducing Sugars.
- Toxicity

  Study of toxic compounds / Detection
- chemicalsstudy in;

  Cigratte, Alcoholic compounds.

⑤ Spectroscopic Related Studies;

UV, IR, NMR, MS, CV etc

⑥ chromatographic studies:

paper, TLC, LC, GC etc
SPE

⑦ Chips industry:

- scientists are trying to invent the Analysis methods For Reducing the chip size. (Reduction of chemicals)
- which Reagants are used for Reduction
- silicon properties / Functionalities etc

⑧ Forensic science:

- → Study of toxic chemicals
- → Quantification / Quality
- → criminals identification / DNA Testing | poisnous substances

⑨ Nano industry:

- Study of nano-size Structures / CNT's, Graphene etc.
- Characterization techniques

RD, X-Ray, SEM, TEM, Atomic Spectroscopy, FTIR, UV-visible

⑩ Environmental Analysis:

- Detection of different metals
- Soil Analysis / water Analysis
- toxic Metals- identification.

⑪ ~~livestock~~ industry:

- → Livestock
- → Food
- → Beverages
- → Fats/oils
- → glass manufacturers.
- → cement
- → Fertilizers
- → Chemicals

* livestock industry → to test the quality of Animal Feed, identifying various species / products in livestock

* oil and Fats industry ⟶ to check all qualitative & Quantitative Analysis on oils.

⑫ Metrology:

- Mass / volume measurements for medical devices, chemical sensors / sensing technology

⑬ Scientific Officer / Research Officer / Lab Expert

↳ Diff. Research labs in Pakistan

↓ (Quality Assurance)

⑭ University Research / Professors / PH.D's

⑮ Teaching Field:

⑯ Nuclear technology:

ANALYTICAL CHEMISTRY

Analytical chemistry deals with the qualitative and quantitative Analysis

Qualitative

- What chemicals are present.
- What type of composition.
- identification of elements, ions or compounds.

Examples;

① e.g Qualitative study of identification of Gunpowder Residue on hands. No need of how much Amount

② * UV-visible → Quantitative study
* MS → Quantitative study
* NMR → both Qualitative & Quantitative
* volumetric Analysis → Titrations
* Electro-Analysis

Quantitative

- How much amount is present
- % age of substances

Examples;

① • price of coal is defined By the % of impurity present in it e.g Sulphure present in coal. Quantification of S

② IR-Study → Qualitative
Chromatographic techniques
↓
Qualitative study

Sample: The substance which is selected for the experiment known As sample. e.g Blood sample, water sample, soil sample etc.

Analyte: specific desired substance present in our sample which is understudy

e.g * Glucose level in blood
* Toxic metals e.g As, Cr, Sb in water
* metals in the soil

Analyze / Analysis:

To characterize / identify / Quantify some thing

Selectivity

- Selectivity is related to the sample
- To select some thing

Example;

★ Blood Sample

Specificity

- Specificity is Related to the Analyte
- To study the specific / desired substance

Example;

★ proteins in blood.

ANALYTICAL PROCESS

↳ Steps Adopted to Analyze something.

Steps;

① Define the problem.

② Select a Method.

③ Sampling.

④ preparation of Samples.

⑤ Necessary chemical Separations

⑥ perform the Measurement / Method development

⑦ Results and Report

1) Define the problem: → The most critical stage.

- what is problem?
- What you want to study/Analze?
- what a study Qualitative/Quantitative?
- How it will Accurate & precise?
- What a cost on it?

2) Selection of Method:

Selection of Method depends upon the No. of factors.

★ Sample type → solid → liquid → Gas → volatile → Non-volatile

★ Size of Sample → macrosize → micro → nano

★ Sample preparation → Needed → Not Needed

★ Sensitivity → concentration detectable or not → LOD → LOQ

★ Selectivity → How much selective is method? for better Results.

★ Experience,/Equipment availability/cost / time involved

★ Availability of Method in literature?

★ Availability of Standard Method

★ Standard Regulations.

3) Sampling: → obtaining a Representating Sample.

Samples

- Gross Samples → Several portion of material to be tested
- Laboratory Samples → small portion to be tested
- Analysis Sample → Actually Analyzed

Selectivity of Samples depends upon following factors;

- Sample type- ✓
- Homogenity of Samples ✓
- Sampling tools-
- Suitable place for Samples.
- Handling tools for Sampling
- Geographically location.

⇒ Necessary precautions/conditions For Biological/Blood Samples:

★ B/c composition of blood varies before and after eating meals ★ **Blood Sample**

⊛ Example;
12h Fasting is Necessary, before Analyzing the cholesterol level in blood.

⊛ preservation for blood Sample is done by NaF otherwise glucose ~~Nature~~ ~~may change~~ may degrade
↓
of glucose/anticoagulants

⊛ When blood is taken → Add Small amount of Anticoagulant such as heparin or citrate Salt. If not add Anticoagulant soluble protein fibrinogen ⟶ converted to insoluble protein fibrin which forms the blood clot.

⇒ Handling and Storing samples: / Storage of Samples

It is necessary to store the Sample. It should be prevented from

- Atmosphere
- Heat/T
- Light

★ **Urine Sample** ⇒ It should kept in Acidic environment
↓ $pH = 4.5$
∴ usually by adding 1 or 2 mL glacial Acetic Acid

Otherwise calcium phosphate present in urine will precipitate, entrapping metal ions.

4) Preparation of Samples:

Factors;

- What type of Sample → solid / → liquid / → gas
- Dissolvation Necessary or Not?
- Which Method used → Acid digestion method / → wet digetion method
- interferences Removal → using masking Agent
- Need to concentrate our Analyte or not?
- Derivitization Necessary OR Not for detection?

⇒ Solids → Dried in oven At 105-110°c for 3h

OR

Solid Samples usually must be put into solution

⇒ inorganic materials;

↳ May be dissolved in various Acids

⇒ organic materials;

Dry Ashing

↓

Sample is burnt 400-700°c

↓

Leaving behind inorganic Residue that is soluble in dil. Acid

Wet digestion

↓

organic matter is destroyed by heating with oxidizing Acids

↓

(mix. of HNO_3 & H_2SO_4)

OR

perchloric Acid.

5) Necessary Chemical Separations:

- Distillation- , Sublimation
- precipitation- $AgNO_3 + NaCl \rightarrow AgCl\downarrow + NaNO_3$
- Solvent Extraction→
- SPE ⟶
- Chromatographic techniques
- Electrophoresis.

6) performing Measurements | Method validation:

- Volumetric Analysis → $\boxed{\text{titrations}}$
- Gravimetric Analysis → precipitations
- instrumental Analysis
  - Spectrophotometric
    - X-RAY
    - UV-visible
    - IR
    - NMR
    - mass
    - SEM, TEM, XRD
  - Electroanalytical
    - potentiometric
    - voltametric

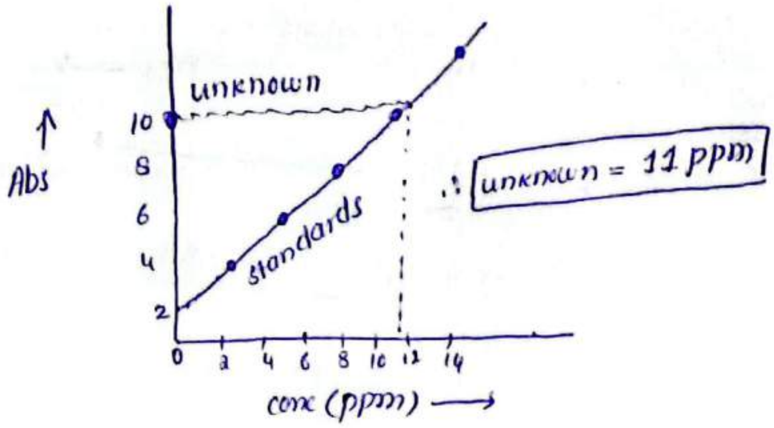
★ instrument standardization:
- calibration curve
- standard Addition
- internal standard method

★ validation of method

↳ compare with the standards values./References material

* Calibration Curve method:

- Different known Amounts of standards are run on instrument → Give absorbance
- Then our unknown Analyte is run give the absorbance.



* Standard Addition Method:

* in this method we determine the Amount of Analyte in an unknown where known Quantity of same standard (Analyte) is added.

* This method is used when our Analyte is in very low Quantity and can not be detectable thus we add the known amount of same type of Analyte standard so that it can be detected. Thus then, it is subtracted from standard.

$$\boxed{\underset{\text{(conc)}}{\text{Analyte}} = \underset{\substack{\text{(conc)}\\ \text{known}}}{\text{Standard}} - \underset{\text{Analyte}}{\text{unknown}}}$$

* Internal Standard Method:

in this method, we add another known analyte (not same) to supress the ionization (increase the Atomization)

e.g Addition of K in Na (unknown)

- K easiey ionized → Thus prevent Na from ionization → thus Sod. Atoms increases

7) Results & Reports

- ⋆ Statistical Analysis is done
- ⋆ Results of mean valus ± S. Deviation.
- ⋆ ANNOVA is applied.
- ⋆ T-test / F-test / paired T-test / student t-test
- ⋆ softwares → mini-lab software to compile the Results
- ⋆ Report is prepared / thesis

CHAPTER 2

Data Handling

Errors → Diff b/w true & Calculated value from the experiment

Errors can be observed while computing Results.

two main types of Errors

Errors

Determinate Errors

- Non Random error
- It can be determined
- It can be Resolved
- This is Systematic error

Examples;

- personal error → due to unexperienced person
  ↓
  Also called operative error
  - teach the person
- Instrumental error
  ↓
  due to uncalibrated instrument | Faulty instrument
  - Removed by calibrating it.
- Method Errors
  ↓
  unvalid Method
  So validity of Method by comparing with Standard Method.

Indeterminate Errors

→ Random Errors

→ Accidental Errors

→ These are complex and it is difficult to Resolve these.

Examples;

→ Naturally occuring errors

→ Environmental factors e.g T, Humidity Volume & concentration changing.

* Fill

Accuracy and Precision

* **Accuracy:**

Difference b/w calculated & Actual value

precesion:

Diff b/w Replicate measurements.

Difference;

Accuracy	precision
1. How close the measurements with true value	1. How close the calculated values to each other
2. More accurate measurements, ~~may not~~ are ~~more~~ precise	2. More precise measurements may not be more Accurate
3. Single measurement	3. Multiple measurements
4. Accuracy is affected due to systemic errors.	4. precision is affected with Random errors.

Ways OF Expressing Accuracy

Accuracy is defined & expressed by the Absolute error & Relative Error.

Absolute Error: ⇒ Difference b/w Actual value & calculated value with respect to sign known as Absolute Error

Absolute Error = Actual value − Measured value

$= 1.0 - 0.9 = +0.1$

Relative Error:

or

$$\text{Relative Accuracy} = \frac{\overbrace{\text{Measured value} - \text{Expected value}}^{\text{Absolute Error}}}{\text{Actual value}} \times 100$$

$$\%\ \text{Error} = \text{Relative Error} \times 100\%$$

Rounding off the data:

- Rounding of data is also important to make your results precise.

① * If last digit is greater than 5 → then number is rounded up to the next higher number.

Example:

$7.47 \Rightarrow 7.5$

$5.66 \Rightarrow 5.7$

② If last digit is less than 5 then nearest digit retained

$3.72 = 3.7$

$9.43 = 9.4$

③ If last digit is 5 then two cases

If nearest digit even
↓
Retained.

If nearest digit odd
↓
increase the digit one value

Example;

7.45 = 7.4

8.75 = 8.8

———————

Significant Figures:

→ The digits in measurement which are known with certainity.

↳ Significant figures show the Accuracy in measurements. We can also understand the precision of a measurement, if we know the Significant figures.

⇒ "more will be the no. of Significant figures, more will be the Accuracy"

Example;

two measurements

2.3769
↓
more S.Fig
↓
more Accurate

2.37
↓
less sig. Fig
↓
less Accurate

Rules:

(i) All the Non-Zero digits are sig. figures

e.g 3.357 → 4 sig. Fig

11.3456 → 6 s. Fig

(ii) Zero b/w Non-zero digits are Significant

e.g 2408 → 4 S. Fig

e.g 10,014 → 5 S. Fig

(iii) Zeros to the Right of decimal are Sig. Figures

e.g $\overset{=2}{2.000}$ → 4 Sig. Fig

e.g 2001.00 → 6 Sig Fig

(iv) Zeros locating in decimal, no. of magnitude less than One → Not Significant

e.g 0.1235 4 sig. Fig

0.0000034 2 sig. Fig

(v) Zeros located in decimal, no. of magnitude greate than one → not Significant

e.g $\overset{3 \times 10^4}{30000}$ → 1 Sig. Fig

120000 → 2 S. Fig

↳ 12×10^4

* * *

Accuracy of Analytical process

9t is important to checque the Accuracy OF some Analytical process such as;

<u>sampling</u>, <u>weighing</u>, <u>volume measurements</u>, <u>precipitations</u>, <u>washing</u>, <u>filleration</u> & <u>ignition</u>.

1) Sampling: (obtaining a Representative Sample)

- proper Handling of Samples is very important

∴ types of Samples
- → Gross Samples → large portion
- → Laboratory Sample → small portion
- → Analysis Sample → to be Analyzed

* Homogenity of Sample is good criteria for Accuracy of Sampling.

What types of problems are faced while obtaining Sample?

Solids: ① → Heterogenity of Sample
② → variation in particle size (↓ Monodisperse)
↓ So Representative Sample difficult to get
③ preservation
④ proper preparation

} → so Random Sampling should done (solutions:)
↓ from 2 or 3 places

Liquids:
- ↳ Homogeneous → Representative Sample easy
- ↳ Here single Random Sample enough
- ↳ 9f liquid Samples are not homogeneous → Then shake these Sample immediately

* { * Blood Sample, * Urine Sample } ⇒ How to preservation? ⇒ Effect The Accuracy

GASES: → Sampling done in evacuated containers
→ usually stainless steel or innert PV Fluoride bag is commonly used.

★ Sample preparation is Also Important factor to evaluate the Accuracy.

- Drying the Sample
- Sample Dissolution → Acid digestion ~~wet~~ Dry digestion
  → wet digestion

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2) Weighing:

Standard laboratory weighings are typically made to 3 or 4 significant figures.

- Accuracy of weighning is done by comparison of weight with the Standard weight.
- Zero point drift Also important for Accuracy.

~~General Rules~~;

Sources of Errors during weighing:

- Zero point drift
- Temperature of object being weighed
- Hygroscopic Nature of Sample
- Air Sensitive Sample

General Rules for weighing:

- Never weighed with fingers → use piece of clean paper
- weigh at room T → avoid Air convection current
- Never put on pan, weighed on vessel / weighing Bottle, weighing dish / paper
- Close the balance cover door.

⇒ Solids;

Hygroscopic

★ Solids Non-metallic materials are usually weighed and dried in a weighing bottle.

★ You can use direct weighing / weighing Dish for Non-Hygroscopic Samples.

liquids;

→ usually done by direct weighing/ weighing bottle with stopper.

* If you are using pipette for volume transferring → rinsed the inside of pippete.

Types of weighing:

- Rough (2 or 3 Sig.fig)
  - Needed when weight is not involved in the computation of the analytical Results.
  - → Not done on Analytical Balance / May completed on top-loading Balance
- Accurate
  - → Results to 4 Significant figures
  - * only performed by Analytical Balance
  - * Accuracy → Near to 0.1mg

3) Volume Measurements:
→ volumetric flask
→ pipets / syringe pipettes
→ Biurette

(i) Volumetric Flasks: → to measure the more Accurate volume
→ used for dilution of Solutions to a certain volume
→ 1mL to 2L

calibration:

To Calibrate a volumetric flask

- First weigh the clean, dry flask & stopper
- Fill it to the mark with distilled water
- There should be no droplets on the neck, If there, blot them with tissue paper.
- Flask & water should be equilibrated to room temperature.

* Weigh the filled Flask and then record the temperature of the water to 0.1 °C. The increase in weight represents the weight in air of water contained by the flask.

(ii) **pipets:** used to transfer of a volume of a Solution.

* 100 — 0.5 mL or less available

precautions

- Not Suck by mouth b/c of moisture adding changes.
- pipette Sucker should be used
- Should be calibrated.
- its tip should touch the flask to exit the last remaing drops.

* Syringe pipets are useful for the delivering microliter volumes.

(iii) **Biurets:** → used for accurate dilivery of a variable amount of a Solution

→ used for titrations. (standard Solutions taken)

precautions:

- Keep eye on lower minscus during experiment
- use stopcock properly → secure tightly to prevent leakage
- tip should touch on the wall of glassware
- it should be calibrated.
- Should be rinsed at least twice with the solution with which they are to be filled. If ~~they~~ it wet they should be rinsed with water first

* ———————— *

4) Precipitations: (Gravimetric ANALYSIS)

Gravimetric Analysis is used for the precipitation. (element/compounds precipitated from its solution by using suitable reagent)

precautions
- precipitates should be practically ★complete
- precipitates must be free from ★contamination.
- Must be of known★ composition.
- Avoid over-washing as no precipitate is quite insoluble
- let the precipitates★ settle before filtering
- The mass of precipitate★ gives the amount of Analyte in the original Sample.

5) Washing of precipitates

★ After the formation of precipitats, they are transferred to the filterpaper, it is washed with the five or six ~~or~~ small ★portions of wash liquid.
↓ more effective than one large volume

★ ~~preci~~ There should be perform a washing★ test to check the completness of washing.

★ Avoid over★ washing as no precipitate is quite insoluble

6) Filteration of precipitates

precautions
- size★ of Filter paper should be proper According to desired substance
- Filterpaper when folded somewhat smaller★ than Funnel
- If possible, liquid should be Filtered Hot★

Filters used for precipitates;

(i) Gooch Crucible (ii) sintered glass crucible
(iii) porcelein F.paper

- Care must be taken in the decanting & transfering of precipitates to avoid losses. This is properly done by using of stirring rod and a wash bottle.

7) Ignition of precipitates

- To Remove the moisture, precipitates are ignited in a filter crucible.
- Ignition is done in Muffle* furnace / Burner*
- Dry & weigh the crucible before adding ~~ce~~ precipitate.
- Ignition temperature* should be maintained.

⇒(CH # Laboratory Tools)

Laboratory Materials & Reagents

Laboratory glasswares specifications:—

★ Borosilicate glass, the most commonly used material for laboratory apparatus such as Beakers pipetts, Biuretts, Flasks, Measuring cylinder etc.

★ These glasswares are majorly manufactured by two companies like pyrex & Kimax.

★ Borosilicate glass are very stable, it tolerate the very high temperature (Hot solutions)-

Some important Materials & their characteristics:

Materials	(sensitivity)	chemicals innertness
★ Borosilicate glass	200 °c	→ Attacked by some Alkali-solutions on heating.
★ Fused Quartz glass	1050°c	→ Resistant to most Acids, halogens & Alkalies than Borosilicate glass
★ porcelain glass	1100°c	→ Excellent innert
★ Stainless steel	400-500°c	→ Not attacked by Alkalies & Acids except conc. HCl, dil. H_2SO_4, Boiling conc. HNO_3
★ polystyrene	70°c	→ Not attacked by HF. but Attacked by → many organic solvent
★ Teflon	250°c	→ innert to most chemicals

= Specifications of chemicals / Reagents

- Reagents and chemicals specification is a key factor for the specific Analysis.

- Different grades of chemicals are available in different laboratories. USE OF these chemicals depends their specification

- purification of chemicals is very important Factor. chemicals quality & purity is highly checked by standard & societies.

- American chemical Society (ACS) → checked the purity OF chemicals.

TYPES OF Reagents

Reagent grade chemicals

- They have minimum purity Requirement
- Less expensive
- Not Good Quality
- Generally used to make the secondary Standard Solutions. (common solutions)

e.g NaOH, KOH, dil HCl $Ca(OH)_2$ etc.

OR

Always most used for the Analysis

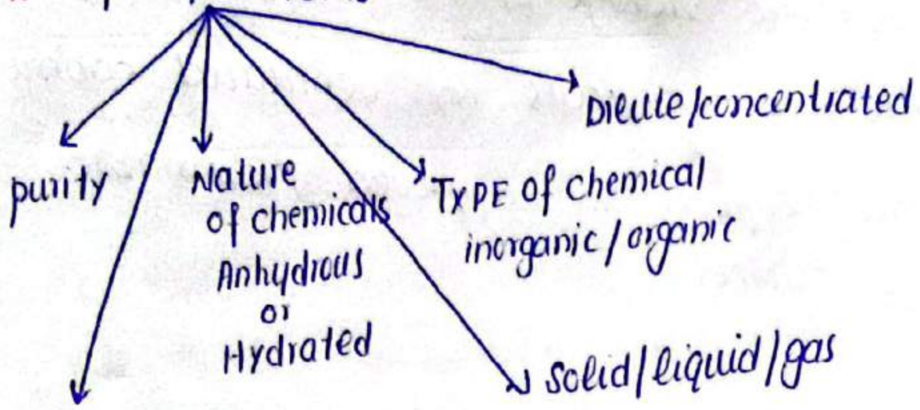
primary standards chemicals

- Also known as American chemical society grade chemicals
  - They Required high purity (About 99.95% purity)
- Expensive
- Good Quality
- Generally used in Analytical laboratory
- Used to make the primary standard solutions or volumetric

e.g $KMnO_4$, $K_2Cr_2O_7$ H_2SO_4 etc

* chemicals specification → are mentioned / Labelled
↓
printed on the label

* specifications



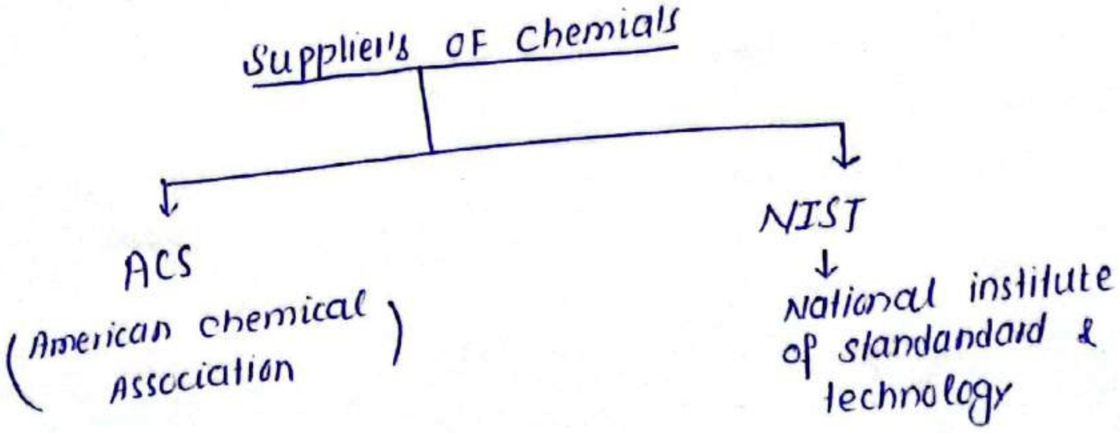
Characteristics

- powder / crystalline
- M.P / B.P
- Density
- Mass
- % purity

* Chemicals Also used as a solvent - purity OF Solvent depends upon the use OF solvent. For what Analysis solvent being to used.

* pure Solvents should used For highly accurate Measurements / techniques

e.g NMR, FTIR, UV-visible, chromatographic techniques.



Storage/Handling of chemicals:

- ★ All chemicals containers should be labelled fully.
- ★ provide a specific storage space for each chemical
- ★ store volatile toxic chemicals in ventilated cabinets.
- ★ Store Flammable liquids in approved Flammable liquid storage cabinets.
- ★ Keep chemical container closed.

CH# Statistical Analysis:

★ Standard Deviation:

$$S.D = S = \sigma = \sqrt{\frac{\Sigma (x_i - \bar{x})^2}{N-1}}$$ ✓

∴ N-1 Degree of Freedom

OR

$$S.D = \sqrt{\frac{\Sigma x_i^2 - (\Sigma x_i)^2/N}{N-1}}$$ ✓

★ Standard Deviation of Mean / Standard Error:

$$S_{mean} = \frac{S}{\sqrt{N}}$$

★ % Relative S.D: / coefficient of variance

$$\% R.S.D = \frac{S}{\bar{x}} \times 100$$

★ variance: (S^2)

$$S^2 = \frac{\Sigma (x_i - \bar{x})^2}{N-1}$$

∴ coefficient of varience

★ Confidence limit:

↳ tells us the Range within which our true value falls.

Formula,

$$\boxed{\text{conf. limit} = \bar{x} \pm \frac{t \cdot s}{\sqrt{N}}}$$

∴ t = statistical factor, depends upon the degree of freedom
↳ choosed by confidence level given in the table either 90%, 95%, 99%.

∴ Degree of freedom = N−1
↳ one Less than the no. of measurements
↳ Represented by 'ν'

Q: Calculate the confidence limit at 95% conf. level.
Following Results are obtained by the Analytical process: 93.50, 93.58, 93.43 wt ?

ν	90%	95%	99%	99.5%
1	—	—	—	—
2	—	—	—	—
3	—	—	—	—
4	—	—	—	—

Tests of Significance:

↳ To develop a new Method, It is necessary to compare it with the accepted method.

★ F-Test: →(used to compare the S.D of two methods) / Precision
↳ indicates the significant difference b/w two methods based on standard deviation.
↳ Based on variance

$$F = \frac{S_1^2}{S_2^2}$$

your method (ν_1) (mg/L) 1st	standard method (mg/L) 2nd
127	130
125	128
123	131
130	129
131	127
126	125
129	

$\rightarrow \nu_2$ $(N-1)$

N = 7
$\nu = 6$

$\rightarrow$ N = 6
$\nu = 5$

varience of two methods is calculated

$S_1^2 = ?$, $S_2^2 = ?$

values of F at 95% confidence level

	$\nu_1 = 2$	3	4	5
$\nu_2 = 2$				
3				
4				
5				
6				

* If calculate value of F is < the tabulated/critical value then it means there is no sig- difference in precision of two methods.

* If calculate F value > tabulated value
↳ significant Difference in precision.

*** T- test :** compare the means OF two populations/metho

↳ determines is there any sig. Difference b/w means OF two groups.

$$t = \frac{\overset{\text{mean of sample}}{\bar{x}} - \mu \rightarrow \text{Theoretical mean of population}}{\underset{\text{S.D}}{s}/\sqrt{\underset{\text{no of observations}}{n}}}$$

↳ AT confi- level 95%.

∴ $t_{cal} < t_{table}$ NO significant Difference

∴ $t_{cal} > t_{table}$ significant Difference.

*** Q-test:**

↳ Quotient test

↳ Decide wether to retain or reject the suspected value.

$$Q = \frac{a \rightarrow \text{Difference b/w suspect No. \& its neighboor/closED}}{w \rightarrow \text{Range}}$$

Example;

103, 107 , 106, 114

Soln: Arrange the data in order.

$103, 106, \underset{\text{closed}}{107}, \underset{\text{suspect}}{\boxed{114}}$

$$Q = \frac{a}{w} = \frac{114-107}{114-103} = \frac{7}{11} = 0.64 \text{ calculated}$$

Tabulated value =

* If $Q_{calculated} \geq$ tabulated value → Reject the suspected value

* If $Q_{cal} <$ tabulated ⟶ NO Rejection of Suspected value.

* **Correlation coefficient: (r)** → measure the strength of the linear relationship b/w two variables.

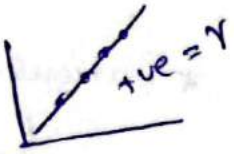
Correlation cofficient is used as a measure of correlation b/w two variables (x & y)

$$r = \frac{\sum x_i y_i - n\overline{xy}}{\sqrt{(\sum x_i^2 - n\bar{x}^2)(\sum y_i^2 - n\bar{y}^2)}}$$

OR

$$r = \frac{n\sum x_i y_i - \sum x_i \sum y_i}{\sqrt{[n\sum x_i^2 - (\sum x_i)^2][n\sum y_i^2 - (\sum y_i)^2]}}$$

If
- * its maximum value is ① → Fully dependent
- * r = 0 → complete independence of variables.
- * minimum value of r = (−1)

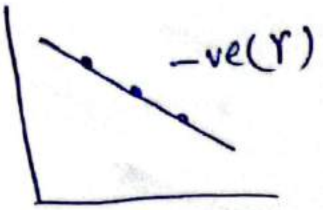


$r = +1 \longrightarrow -1$

*** General Rule for linearity;**
↳ Research

- * If $r \leq 0.95$ Fair curve
- * If $r \leq 0.99$ Good curve
- * If $r > 0.99$ Excellent linearity

- * $r > 0$ +ve Relationship
- * $r < 0$ −ve Relationship



* coefficient of determination: (R^2 / r^2)
↳ Better measure of Fit as compared to correlation cofficient

*** LOD :** ↷ conc that gives a signal that is above the background signal by 3 times the S.D of the Background signal

$$\boxed{LOD = 3.3 \times \frac{S.D}{Slope}}$$

*** LOQ:** lowest conc. of Analyte that can be measured in the sample matrix at an acceptable level of precision and Accuracy.

$$\boxed{LOQ = 10 \times \frac{S.D}{Slope}}$$

* Quality Assurance & METhOD validation :

- * GLP → Good Laboratory practices
- * method validation
- * Quality Assurance

* various Government Agencies → provide guidlines for method validation
- ↳ EPA (Environmental protection Agency)
- ↳ FDA (Food & Drug Administration)
- ↳ AOAC (Association of American chemists)

* Good- Laboratory practices:
- ↳ GLP ensures correct Results are Reported
- ↳ GLP → Should have SOP's
  - ↳ standard operating procedures
- ↳ Analytical method
- ↳ instrument maintenance
- ↳ Sample handling & practices
- ↳

MEthOD Validation:

There is some need of method validation

* Following are elements For method validatio

- Selectivity
- Sensitivity
- LOD
- LOQ
- Linearity
- Accuracy
- precision
- Robustness

***Selectivity:** → Either method is selective or not to measure all Analyte conc.
Extent of method to measure the Analyte other than matrix

*** Linearity:**
Linear Respons b/w Analyte conc. v/s response
$R^2 \approx 1$

*** Accuracy:**
closEness of obtained value with true value

*** precision:**
Repeated values closeness

*** Sensitivity:** → How much the developed method is sensitive-
↳ Determined by the slope of calibration curve

*** Range:**
↳ working Range of method For concentration over which acceptable Accuracy and precision obtained

*** LoD:** $= 3.3 \times \frac{S.D}{Slope}$

*** LoQ:** $= 10 \times \frac{SD}{slope}$

Good Luck