TRƯỜNG ĐẠI HỌC NÔNG LÂM TP. HỒ CHÍ MINH
KHOA MÔI TRƯỜNG VÀ TÀI NGUYÊN

PHÂN TÍCH MÔI TRƯỜNG

Mã môn học: 212930
(3 tín chỉ: 30 tiết lí thuyết và 30 tiết thực hành)

Giảng viên: TS. Ngô Vy Thảo
Email: ngovythao@hcmuaf.edu.vn

1

1

CHƯƠNG 4
PHÂN TÍCH CÁC CHỈ TIÊU MÔI TRƯỜNG KHÍ

2

4.1 Những điều lưu ý

- Đây là bước quan trọng nhất trong suốt quá trình quan trắc.
- Có vẻ dễ, nhưng …
- Mẫu không tốt: địa điểm thu mẫu không hợp lí, pp lấy và bảo quản mẫu không đúng, trữ mẫu quá lâu.
- Vật liệu thiết bị lấy/chứa mẫu: nhựa, Teflon, thép không rỉ, tùy thuộc vào chất cần phân tích.

3

4.2 Các thiết bị lấy mẫu không khí

- Water samples (rain, snow, and ice): buckets (plastic or stainless steel), automated samplers.

Wet-only rainwater sampler Ecotech Pty Ltd, Australia.

Low particulate volume sampler Ecotech

PM2.5, PM10, TSP sampler, Ecotech

Q-MAX Tube Holder

Supelco Q-Max pump

4

CHƯƠNG 5
KĨ THUẬT PHÒNG THÍ NGHIỆM

5

CONTENTS

1. How to keep a legally defensible laboratory notebook
2. Statistical analysis

6

PART 1-1: LAB NOTEBOOK

1. Lab notebook is a legal document

- Your data may have to be explained, defended, reconstructed or repeated without your assistance, so others must be able to understand what you did.

7

PART 1-1: LAB NOTEBOOK

2. Recordkeeping guidelines

- Complete the title page when the notebook is issued
  - All persons recording in the notebook must also sign the title page and give an example of initials used.

8

Cover and Title Page

Laboratory Record
Wastewater Treatment by Modified Activated Sludge Process
Ngo Vy Thao
Department of Environmental Sciences, FERN
Nong Lam University
Quarter 6, Linh Trung Ward, HCMC

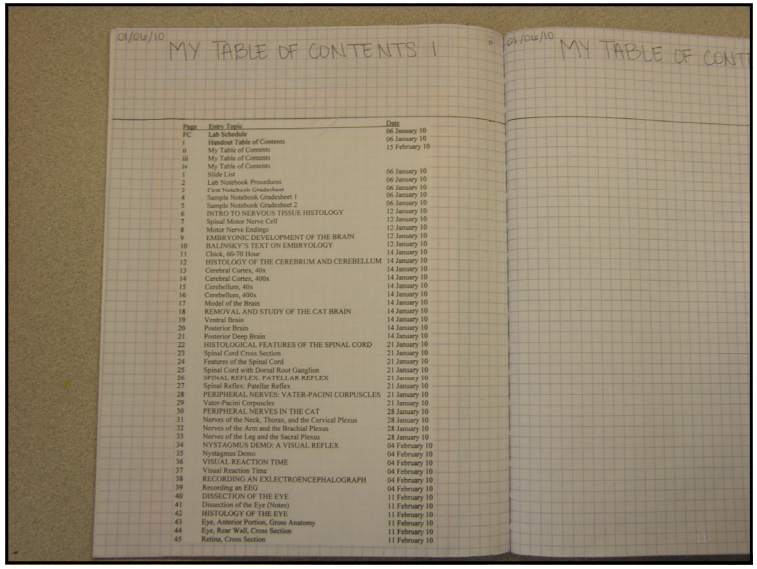
9

PART 1-1: LAB NOTEBOOK

2. Recordkeeping guidelines

- Table of contents
  - Record only the first page number of each multi-page experiment.

10



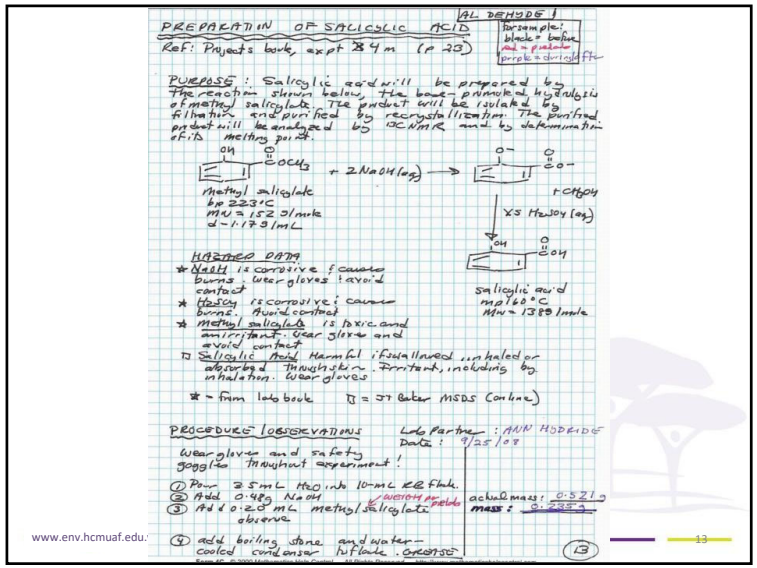
11

PART 1-1: LAB NOTEBOOK

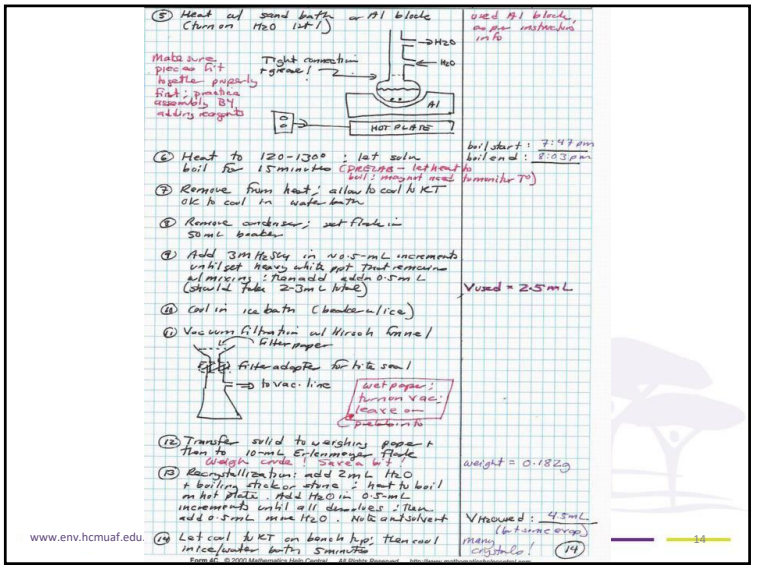
2. Recordkeeping guidelines

- Each recorded lab should have the following parts:
  - Objective or purpose of the lab
  - Plan, outline or flow diagram of lab
  - Step by step procedure
  - Raw data
  - Results, including graphs, tables, figures, photos and/or drawings
  - Conclusion: include the biological and chemical concepts involved, whether the objective was met, any problems encountered, and suggestions for future experiments.

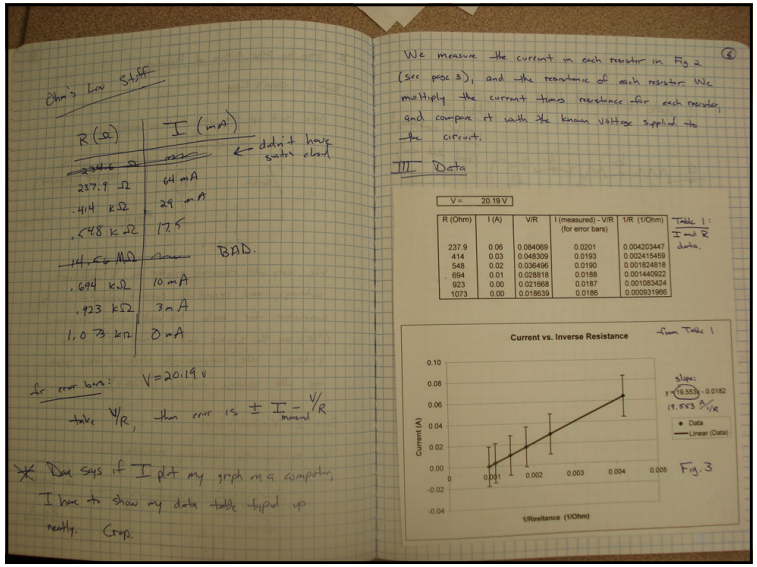
12



13



14



15

PART 1-1: LAB NOTEBOOK

2. Recordkeeping guidelines

- More typically, notebook entries are written as a *diary*. It should include *everything* about the work so that another person can read the notebook and know exactly what was done.
- If procedures or other information are copied from a source, the source must be identified in the lab notebook.

16

PART 1-1: LAB NOTEBOOK

3. When are data recorded?

- Entries should be made as the experiment proceeds, but no later than as soon as it is done or when an idea is conceived.

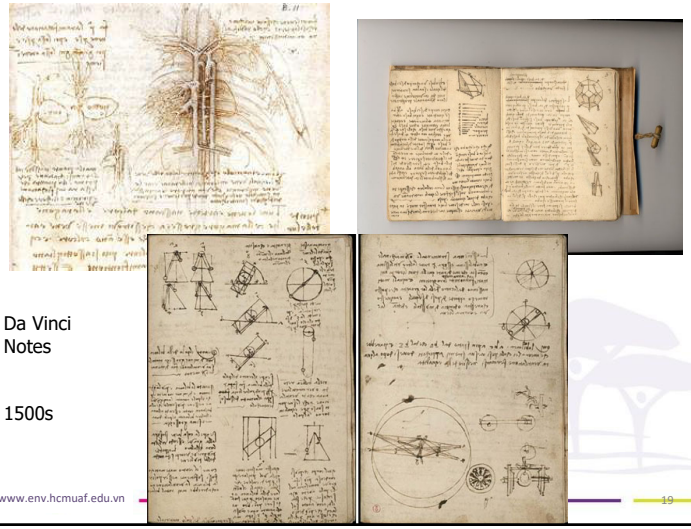
17

PART 1-1: LAB NOTEBOOK

4. What are data?

- Raw data: original of handwritten information or a printout from equipment.
  - Descriptions of observations, procedures, events, for example
- Calculated data: derived from a calculation or statistical evaluation of the raw data.
- Transcribed data: copied raw or calculated data; should indicate "exact copy of original" or where the original data is located.

18



Da Vinci Notes

1500s

19

PART 1-1: LAB NOTEBOOK

5. What do you record?

- Objectives, ideas, experimental plans or outlines, preparations, procedures, data, observations, calculations, discussions, conclusions, future plans and potential uses
- What actually happened
- Results

20

PART 1-1: LAB NOTEBOOK

5. What do you record?

- Notes of unexpected results or observations
- Deviations to a planned protocol
- All measurements and important test conditions (weights, volumns, temperature, etc.)
- All units (ul, g, mg, etc.)

21

PART 1-1: LAB NOTEBOOK

5. What do you record?

- Indicate if the numbers are estimated, rather than measured
- Indicate if the numbers were calculated and provide the equation
- If using Excel, print out the formulas
- Indicate if the number has been rounded or truncated
- Document critical events to prove compliance with SOPs

22

PART 1-1: LAB NOTEBOOK

6. How do you record the data?

- Directly into the notebook; not on post-its, paper towels, scraps of paper, etc.
- In black or blue, indelible ink; no gel pens
- Make entries only in the ruled areas of the numbered pages
- Unnumbered pages can not be used
- Only one experiment per page
- Attach forms or printouts

23

PART 1-1: LAB NOTEBOOK

7. What is the procedure for attaching forms and printouts?

- Attach only to numbered pages within the ruled area only
- Taped on at least 2 sides
- Fully exposed, not folded
- Not covering any previously recorded entries
- With hash marks on at least two corners
- Write the notebook and page number on the attachment
- Sign and date along the edge

24

PART 1-1: LAB NOTEBOOK

8. Who generated the data and where?

- Record the data on the same day it is generated, not after the fact.
- A single page can cover events from more than one day, by the dates must be indicated on each event
- The person making the entry must sign the page.

25

PART 1-1: LAB NOTEBOOK

9. What materials and equipment did you use?

- Important materials must be noted:
  - Related to the reconstructability and repeatability of the experiment
  - Variability between batches and lots
  - Be specific: not just "the buffer"; name it.
  - List the purity, concentration, etc.
  - List the source, catalogue number, etc.
  - Record the recipes

26

PART 1-1: LAB NOTEBOOK

10. Equipment?

- Reactors
- Analytical instruments
- Hardware
- Software and version number
- Unique equipment ID numbers
- Date of last maintenance
- Date of last calibration
- Date of last performance check

27

PART 1-1: LAB NOTEBOOK

11. What routine or repeated procedures did you use?

- Previously run procedures recorded in your notebook
- SOPs
- Must record deviations

28

PART 1-1: LAB NOTEBOOK

12. Conclusions: fact vs. opinion

- Fact: No reaction was observed; vs Opinion: these two chemicals don't react.
- Fact: Expected results were not obtained; vs. Opinion: No good
- Fact: Under these circumstances, the reaction was unsuccessful; vs. Opinion: failed.

29

PART 1-1: LAB NOTEBOOK

13. Do I need a witness?

- Notebook pages must be witnessed within 2 weeks of an experiment or procedure.
- Witness must be someone on your team.

30

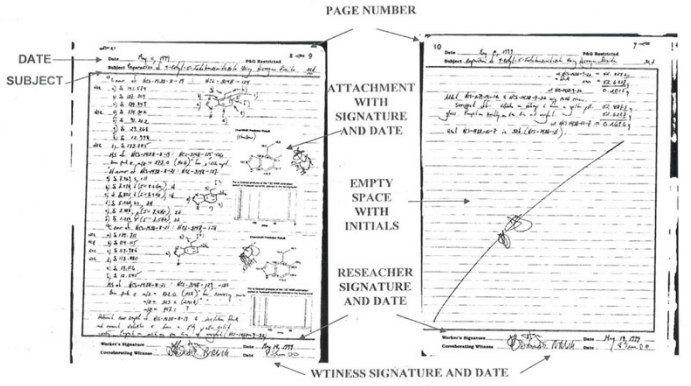
PART 1-1: LAB NOTEBOOK

14. How clear or understandable is your data?

- Legible to others?
- Clear, detailed so someone else in your discipline could understand it and repeat it?
- Include drawings and flow charts to improve clarity?
- Are abbreviations defined and obvious?

31

EXAMPLE NOTEBOOK PAGES



32

PART 1-1: LAB NOTEBOOK

15. What portions of the notebook get X'ed out?

- Unused portions > 3 lines
- Unused fields in forms and tables should be lined out or marked NA

33

PART 1-1: LAB NOTEBOOK

16. Mistakes?

- Never use white-out
- Never erase
- Never write-over
- Never discard or replace attached supplementary data
- Always record a defensible reason for the correction/edit
- Always circle the reason
- Always add your dated initials to the corrected/edited data after the circled reason

34

PART 1-1: LAB NOTEBOOK

17. Where should the notebook be kept?

- In a company or university lab, the lab notebook belongs to the company or university, and should NOT be removed from the premises.

35

References

- **GLP Recordkeeping** http://users.stlcc.edu/departments/fvbio/Lab_Practices_GLP_STLCC.htm
- **Good Laboratory Notebook Practice** http://www.mddionline.com/article/good-laboratory-notebook-practice-0
- **Laboratory Notebook Guidelines** http://www.bookfactory.com/special_info/lab_notebook_guidelines_A4.html
- **Advice on keeping a laboratory notebook** http://www.swarthmore.edu/NatSci/cpurrin1/notebookadvice.htm
- **Guidelines for Keeping a Laboratory Record** http://www.ruf.rice.edu/~bioslabs/tools/notebook/notebook.html#entry

36

PART 1-2: STATISTICAL ANALYSIS

1. Linear least squares analysis

- Instruments read signals, but concentrations directly.
- Calibration curve (line) is use to relate the signal to concentration.
- Least squares regression: does one quantitative variable explain or cause changes in another variable?

37

PART 1-2: STATISTICAL ANALYSIS

1. Linear least squares analysis

- Explanatory variable: explains or causes changes in the other variable; the x variable. (independent variable)
- Response variable: the y-variable; it responds to changes in the x-variable. (dependent variable)

38

PART 1-2: STATISTICAL ANALYSIS

1. Linear least squares analysis

- Fertilizer (x) → corn yield (y)
- Advertising $ (x) → store income (y)
- Drug dose (x) → blood pressure (y)
- Daily temperature (x)
  → natural gas demand (y)
- Change in min wage(x)
  → unemployment rate (y)

39

PART 1-2: STATISTICAL ANALYSIS

1. Linear least squares analysis

- Simplest relationship
- Simplest equation that describes the dependence of variable y on variable x

$$y = mx + b$$

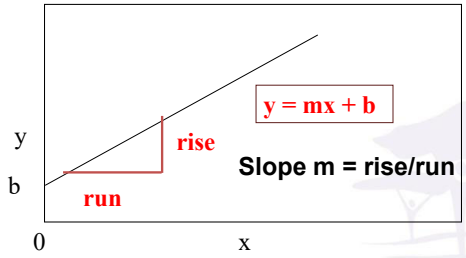
- Linear equation
- Graph is line with slope m and y-intercept b

40

PART 1-2: STATISTICAL ANALYSIS

1. Linear least squares analysis

Graph



41

PART 1-2: STATISTICAL ANALYSIS

1. Linear least squares analysis

- $(x_1, y_1), (x_2, y_2), \ldots, (x_n, y_n)$
- Draw the line y = mx + b through the scatterplot , the point on the line corresponding to x_i is

$\hat{y}_i = mx_i + b$; $\hat{y}_i$ is the value of y predicted by the line $y = mx + b$ when $x = x_i$;

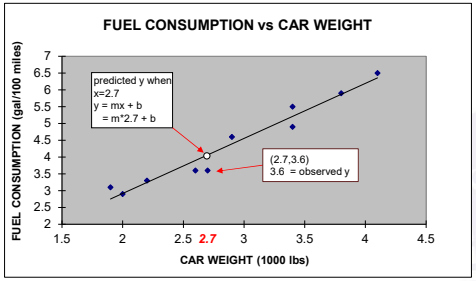
y_i is the observed value of y when $x = x_i$.

42

PART 1-2: STATISTICAL ANALYSIS

1. Linear least squares analysis

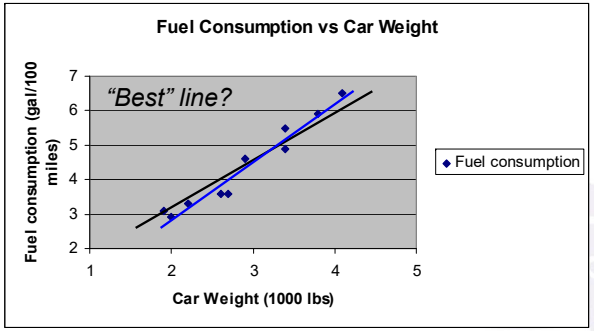
- Observed y, Predicted y



43

PART 1-2: STATISTICAL ANALYSIS

1. Linear least squares analysis

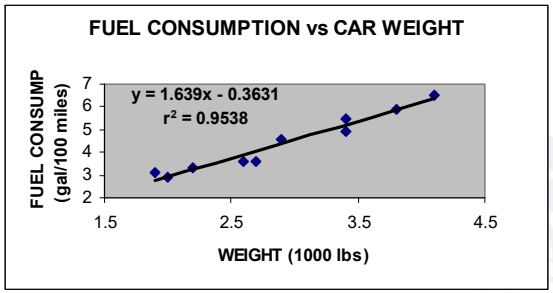


44

PART 1-2: STATISTICAL ANALYSIS

1. Linear least squares analysis

- Scatterplot with least squares prediction line



45

PART 1-2: STATISTICAL ANALYSIS

1. Linear least squares analysis

How do we draw the line? Residuals

the i^{th} residual is the vertical deviation of the i^{th} data point from the line:

i^{th} residual = observed y − predicted y

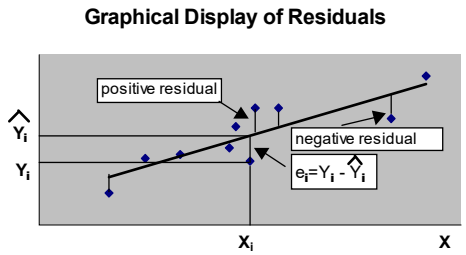
$= y_i - \hat{y}_i$

$= y_i - (mx_i + b)$

46

PART 1-2: STATISTICAL ANALYSIS

1. Linear least squares analysis



47

PART 1-2: STATISTICAL ANALYSIS

1. Linear least squares analysis

Criterion for choosing what line to draw: method of least squares

- The method of least squares chooses the line that makes the sum of squares of the residuals as small as possible.
- This line has slope m and intercept b that minimizes $\sum_{i=1}^{n}[y_i - (mx_i + b)]^2$ for the given observations (x_i, y_i).

48

PART 1-2: STATISTICAL ANALYSIS

1. Linear least squares analysis

For calculation purposes we use the following equations (S's are the sum of squared error or residuals):

$$S_{xx} = \sum (x_i - \bar{x})^2 = \sum (x_i^2) - \frac{(\sum x_i)^2}{N}$$

$$S_{yy} = \sum (y_i - \bar{y})^2 = \sum (y_i^2) - \frac{(\sum y_i)^2}{N}$$

$$S_{xy} = \sum (x_i - \bar{x})(y_i - \bar{y}) = \sum x_i y_i - \frac{\sum x_i \sum y_i}{N}$$

where x_i and y_i are individual observations, N is the number of data pairs, and $\bar{x}$ and $\bar{y}$ are the average values of the observations. Six useful quantities can be computed from these.

49

PART 1-2: STATISTICAL ANALYSIS

1. Linear least squares analysis

1. The slope of the line (m) is $m = S_{xy}/S_{xx}$.
2. The y intercept (b) is $b = y - mx$.
3. The standard deviation of the residuals (s_y) is given by

$$s_y = \sqrt{\frac{S_{yy} - m^2 S_{xx}}{N-2}}$$

4. The standard deviation of the slope is

$$s_m = \frac{s_y}{\sqrt{S_{xx}}}$$

5. The standard deviation of the intercept (s_b) is

$$s_b = s_y \sqrt{\frac{\sum (x_i^2)}{N \sum (x_i^2) - (\sum x_i)^2}} = s_y \sqrt{\frac{1}{N - (\sum x_i)^2 / \sum (x_i^2)}}$$

50

PART 1-2: STATISTICAL ANALYSIS

1. Linear least squares analysis

6. The standard deviation for analytical results obtained with the calibration curve (s_c) is

$$s_c = \frac{s_y}{m} \sqrt{\frac{1}{L} + \frac{1}{N} + \frac{(\bar{y}_c - \bar{y})^2}{m^2 S_{xx}}}$$

where $\bar{y}_c$ is the mean signal value for the unknown sample, L the number of times the sample is analyzed, N the number of standards in the calibration curve, and $\bar{y}$ is the mean signal value of the y calibration observations (from standards). Thus, the final result will be a value (the analytical result) plus or minus another value (the standard deviation, s_c).

Most calculators have an r or r^2 key and you may know that the closer this value is to 1.00, the better. This number comes from

$$r = \frac{\sum x_i y_i}{\sqrt{\sum (x_i^2) \sum (y_i^2)}}$$

r (and r^2) is called the *coefficient of regression* or *regression coefficient*.

51

PART 1-2: STATISTICAL ANALYSIS

1. Linear least squares analysis

r^2 : The Variation Accounted For

- The square of the correlation coefficient r gives important information about the usefulness of the least squares line

52

PART 1-2: STATISTICAL ANALYSIS

1. Linear least squares analysis

r^2: important information for evaluating the usefulness of the least squares line

$-1 \le r \le 1$ implies $0 \le r^2 \le 1$

The square of the correlation coefficient, r^2, is the fraction of the variation in y that is explained by the least squares regression of y on x.

The square of the correlation coefficient, r^2, is the fraction of the variation in y that is explained by the variation in x.

53

PART 1-2: STATISTICAL ANALYSIS

1. Linear least squares analysis

Example: car weight, fuel consumption

- x=car weight, y=fuel consumption

  $r^2 = (.9766)^2 \approx .95$

  About 95% of the variation in fuel consumption (y) is explained by the linear relationship between car weight (x) and fuel consumption (y).
- What else affects fuel consumption?
  - Driver, size of engine, tires, road, etc.

54

PART 1-2: STATISTICAL ANALYSIS

1. Linear least squares analysis

- Limit of Detection (LOD) is taken as the lowest concentration of an analyte in a sample that can be detected, but not necessarily quantified, under the stated conditions of the test.

$$LOD = 3*s_{blk}/m$$

55

PART 1-2: STATISTICAL ANALYSIS

1. Linear least squares analysis

- Limit of Quantitation (LOQ) is the lowest concentration of an analyte in a sample that can be determined with acceptable precision and accuracy under the stated conditions of test.

$$LOQ = 10* s_{blk}/m$$

56

PART 1-2: STATISTICAL ANALYSIS

2. Propagation of uncertainty

- The final calculation, s_c, only incorporates error associated with the linear least squares regression.
- An equally important value is the propagation of uncertainty (POU) resulting from multiple dilutions and weighing events.

57

PART 1-2: STATISTICAL ANALYSIS

2. Propagation of uncertainty

TABLE 2-3. Tolerances for Laboratory Balance Weights

Denomination (g)	Tolerance (mg) Class 1	Tolerance (mg) Class 2	Denomination (mg)	Tolerance (mg) Class 1	Tolerance (mg) Class 2
500	1.2	2.5	500	0.010	0.025
200	0.50	1.0	200	0.010	0.025
100	0.25	0.50	100	0.010	0.025
50	0.12	0.25	50	0.010	0.014
20	0.074	0.10	20	0.010	0.014
10	0.050	0.074	10	0.010	0.014
5	0.034	0.054	5	0.010	0.014
2	0.034	0.054	2	0.010	0.014
1	0.034	0.054	1	0.010	0.014

Source: Harris (1999).

58

PART 1-2: STATISTICAL ANALYSIS

2. Propagation of uncertainty

TABLE 2-4. Tolerances of Class A Burets

Buret Volume (mL)	Smallest Graduation (mL)	Tolerance (mL)
5	0.01	±0.01
10	0.05 or 0.02	±0.02
25	0.1	±0.03
50	0.1	±0.05
100	0.2	±0.10

Source: Harris (1999).

59

PART 1-2: STATISTICAL ANALYSIS

2. Propagation of uncertainty

TABLE 2-5. Tolerances of Class A Volumetric Flasks

Flask Capacity (mL)	Tolerance (mL)	Flask Capacity (mL)	Tolerance (mL)
1	±0.02	100	±0.08
2	±0.02	200	±0.10
5	±0.02	250	±0.12
10	±0.02	500	±0.20
25	±0.03	1000	±0.30
50	±0.05	2000	±0.50

Source: Harris (1999).

TABLE 2-6. Tolerances of Class A Transfer Pipets (Harris, 1999)

Volume (mL)	Tolerance (mL)	Volume (mL)	Tolerance (mL)
0.5	±0.006	10	±0.02
1	±0.006	15	±0.03
2	±0.006	20	±0.03
3	±0.01	25	±0.03
4	±0.01	50	±0.05
5	±0.01	100	±0.08

Source: Harris (1999).

60

PART 1-2: STATISTICAL ANALYSIS

2. Propagation of uncertainty

TABLE 2-7. Error Propagation in Arithmetic Calculations

Type of Calculation	Example	Standard Deviation of x
Addition or subtraction	$x = p + q - r$	$s_x = \sqrt{s_p^2 + s_q^2 + s_r^2}$
Multiplication or division	$x = p(q/r)$	$\frac{s_x}{x} = \sqrt{\left(\frac{s_p}{p}\right)^2 + \left(\frac{s_q}{q}\right)^2 + \left(\frac{s_r}{r}\right)^2}$
Exponentiation	$x = p^y$	$\frac{s_x}{x} = y\frac{s_p}{p}$
Logarithm	$x = \log_{10} p$	$s_x = 0.434\frac{s_p}{p}$
Antilogarithm	$X = \text{antilog}_{10} p$	$\frac{s_x}{x} = 2.303 s_p$

Source: Skoog et al. (1998).

61

PART 1-2: STATISTICAL ANALYSIS

2. Propagation of uncertainty

The use of these and other tolerances is illustrated in the following example. We weigh out 10.00 g of sample, extract it into 100 mL of solvent, make a 1 : 10 dilution, inject 1.0 µL into a GC, and calculate the concentration.

Operation	Raw Value of Operation	Error Associated with Each Operation (as $\pm s$)
Weighing	10.00 g	0.05
Extraction efficiency	0.95	0.02
Extraction volume	100.00 mL	0.02
Dilution 1	10.00	0.01
Injection volume	1.00×10^{-6} L	0.01×10^{-6}
Calculation of concentration (from linear least squares analysis)	1.14 pg/µL	0.05

62

PART 1-2: STATISTICAL ANALYSIS

3. Student's *t* test

- Purpose:
  - To know if the average is within an acceptable range of the true value

$$\bar{x} - \mu = \pm\frac{t \cdot s.d.}{\sqrt{N}}$$

$\bar{x}$: mean of your measurements
µ : known or true value of the sample
t : the value from the t table
s.d. : the standard deviation
N: the number of replicates

If $\bar{x} - \mu > \pm\frac{t \cdot s.d.}{\sqrt{N}}$, the experimental and true values are statistically different.

Degree of freedom: N - 1

63

PART 1-2: STATISTICAL ANALYSIS

3. Student's *t* test

- Purpose:
  - To compare mean values obtained from two different techniques

$$\overline{x_1} - \overline{x_2} = \pm\frac{t \cdot s.d._{pooled}}{\sqrt{\frac{n_1 n_2}{n_1 + n_2}}}$$

$s1$, $s2$: respective standard deviations about each mean
$n1$, $n2$: number of observations in each mean

$$s_{pooled} = \sqrt{\frac{s_1^2(n_1 - 1) + s_2^2(n_2 - 1)}{(n_1 + n_2) - 2}}$$

If $\overline{x_1} - \overline{x_2} > \pm\frac{t \cdot s.d._{pooled}}{\sqrt{\frac{n_1 n_2}{n_1 + n_2}}}$, the two techniques are different.

Degree of freedom: $N_1 + N_2 - 2$

64