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Internal and external quality control system

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Quality Assurance (QA)

Over-all management and systematic actions necessary to provide adequate confidence that a service meets the required quality – ISO/IEC 17025

Quality Control (QC)

Operational techniques and activities that are carried out in a laboratory to ensure the quality of the generated analytical data

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QC system:

should, as far as possible, cover the entire analytical procedure including

- sample preparation, extraction and digestion steps
- measurement or quantitation step

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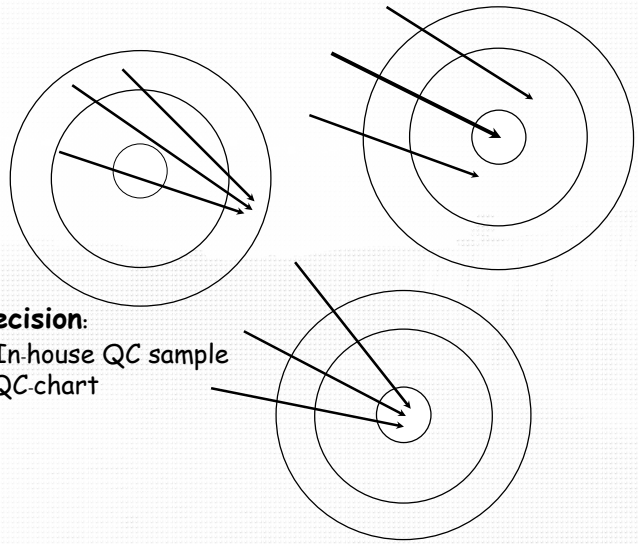
LABORATORY WITH CONSISTENTLY RELIABLE DATA . . . . . .

- Analytical method: validated ⟵ *RM* and fully documented
- Laboratory staff: well-trained and knowledgeable
- Internal quality assessment system: ⟵ *RM*
  - in-house RM/QC sample/control chart
  - using *RM* for checking accuracy
- External quality assessment system :
  - Proficiency testing ⟵ *RM*
- Reported data: traceable to reliable and well documented QC system



Accuracy?:
- Reference material

Precision:
- In-house QC sample
- QC-chart

Reliability (Precision + accuracy)
QC chart + reference materials + lab performance study

Internal and external quality control system

	Intern al QC	External QC: proficiency testing
Sample	In-house QC sample	Test material
- Preparation	✓	✓
- Homogeneity testing - Stability testing	±	✓
Assigned value	Mean ± SD	Robust mean ± robust SD
Performance evaluation criteria	Mean ± 2SD Quality control chart	z-score ≤ 2 Graph or table
Statistical evaluation:	✓ Cochran test, ANOVA	✓✓✓ Cochran test, ANOVA, ISO 13528 – for various purposes

WHAT SAMPLES CAN BE USED FOR TEST MATERIAL PREPARATION?

Any sample which is

- homogeneous
- matrix and analyte stable over time
- similar matrix to the test sample
- readily available, in sufficient quantity
- reasonably cheap

Scheeling P, QHSS, Australia, 1998

POSSIBLE TEST SAMPLES ARE >>>>

1. Certified food reference materials (CRM): highly accurate analytical data *BUT* very (too) expensive for every-day use
2. Reference materials with "consensus" analytical data - can still be reasonably expensive, e.g., from NATA, FAPAS, INMU
3. Laboratory samples (from previous testing): laboratory data - gratis and readily available.
4. For food analysis lab: selected *foods* from supermarkets with data on label - cheap, readily available

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A. Selection of test materials

1. Select a test material that "fit-for-purpose"

General characteristics of PT test material – for nutrient analysis

- Natural sample, fresh or dry form, with various matrices and easily available
- Homogeneous and stable within justified period - *with respect to matrix and components of interest*
- Contains a wide range of components at desirable amounts with minimal or no contamination from other materials
- Reasonable price

Reference: ISO Guide 34: Quality system guideline for the production of reference materials
ISO/IEC 17043: Conformity assessment – General requirements for proficiency testing

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2. Several factors must be considered

Which analytes to be included?

What concentration of the analytes?

Which foods should be selected? ← food composition data (FCD)

What sample matrix will be selected? dry or fresh (perishable)

Stability of sample matrix and nutrients/analytes of interest?

FCD - information on nutritive values of foods? Where to get

- Hard copy: MOPH, INMU
- Electronic database:

MOPH: http://nutrition.anamai.moph.go.th/FoodTable/Html/frame.html

FAO : http://www.fao.org/infoods/directory_en.stm

USDA (U.S. Department of Agriculture): www.ars.usda.gov/ba/bhnrc/ndl

- Food package – nutrition information/Nutrition Facts

Example: test sample

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Selected sample	Target analytes
Soybean flour, weaning food	Proximate composition minerals, vitamins esp., folate, fatty acids
Dry pork, fish powder, milk powder	Proximate composition minerals, vitamins, cholesterol, fatty acids

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3. Where to get the selected food samples?

1) Use laboratory samples from previous testing or in-house RM
Analytical data - gratis and readily available

2) Select foods from a supermarkets* with Nutrition Information on the label *…not expensive, readily available*

**Note: same producing lot*

3. Request from a food producer/distributor

4. Purchase from fresh market……fresh foods

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4. What would be total amount of the test material?

Amount of sample per package ?
- # parameters to be covered
- # amount used per analysis
- # replicate analysis

Number of packages ?
- # expected participants
- # samples used for homogeneity and stability testing
  (cover the whole range of storage period as planned)
- # remaining RMs to be used for internal and external QC system

⟹ Total amount of food sample required

Preparation of test materials

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- How to homogenise the test material?
- Will it require additional process or specific handling?
  Environmental condition? - special care .....type of sample

  Environmental condition control room
  - Humidity
  - Temperature
  - Contamination
- What to be used for packaging and how?....
  normal sealed or under vacuum
- Where to keep the prepared sample?

Ref: ISO Guide 34 , ISO/IEC 17043

B. Preparation of test materials: step by step

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- Preparation of homogenised test material using a proper grinder or mixer (do your best to ensure homogeneity)
- Sieving: passing through a standard sieve with target particle size, e.g., 250 mesh
- Mixing thoroughly again by manual or using a mixer to ensure uniform distribution.
- Packing aliquots of needed amount of the test material into a proper containers/packages
  Food sample: laminated aluminum foil bags,
  screw-cap bottles, vials, ampoules, etc.
- Labelling – e.g., name, date of preparation, code number
- Keeping the test materials at a proper condition, e.g., 4°C

Instrument for preparation of test materials

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Dry foods ↓ Grinder/milling machine

Fresh foods ↓ Food processor/mincer/blenders

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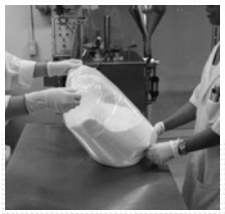
Instrument for mixing test materials



Manual mixing

Rotating mixer

V-shape mixer

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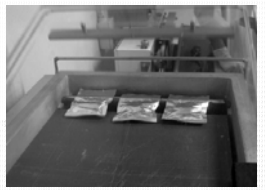
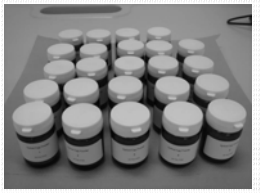
Instrument for packing & containers

Plastic sealer



Vacuum sealing machine

Samples packed in can and laminated aluminum foil



Screw-cap glass bottle

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Sample - Preparation - Homogeneity testing - Stability testing	In-house QC sample ✓ ±	Test material ✓ ✓
Assigned value	Mean ± SD	Robust mean ± robust SD
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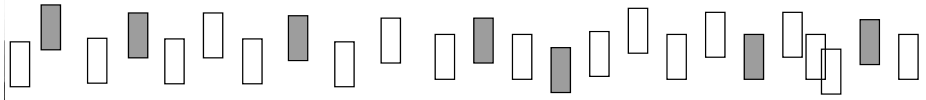
Statistical evaluation:	✓ Cochran test, ANOVA	✓✓✓ Cochran test, ANOVA, ISO 13528 – for various purposes

A. Testing for sample homogeneity

Homogenised test materials, packed in laminated aluminum foil bags or in bottles



selected 10 representative samples at random

↓

Analyse 2 test portions for representative components, in a random order under repeatability conditions* by appropriate methods

↓

Statistical evaluation without exclusion of any value

** each, measured on the same day(s), using the same conditions, by the same competent analyst, using same method, instrument and calibrants*

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Testing for sample homogeneity: 2 steps

Step 1. Checking for <u>within sample variation</u> (precision of the analyst): using Cochran' s maximum range test

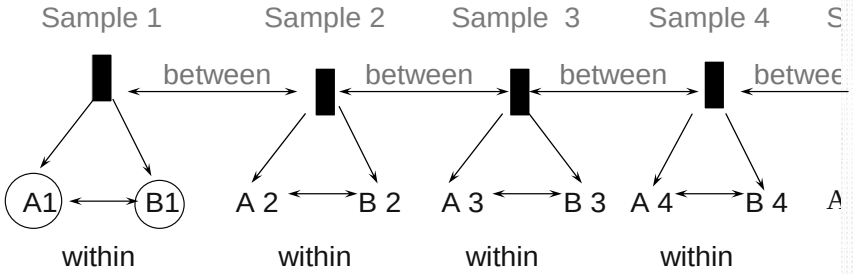
Step 2. Checking for <u>between sample variation</u> using
- One Way ANOVA
- ISO 13528:2005
- %RSD, HORRAT approach

1) Thompson M, Ellison SLR, Wood R. The International harmonised protocol for the proficiency testing of analytical chemistry laboratories (IUPAC Technical Report). *Pure Appl Chem* 78: 145-196, 2006.

2) ISO 5725-1981: Precision of test methods – Determination of repeatability and reproducibility by interlaboratory tests.

3) ISO 13528: 2005 Statistical methods for use in proficiency testing of interlaboratory comparisons.

Check homogeneity of the test materials



Check precision of testing laboratory (analyst who does the work)

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B. Stability testing

1) Classical stability study: i.e., vitamins

- Store samples at storage conditions which represent conditions of the entire process of transportation and storage
- Re-analyse the suspected component in 5 randomly single samples at specified storage period
- If the obtained values are in the range of mean ± 2SD derived from the homogeneity study or at 0 day storage; the component is stable

2) Isochronous stability study: i.e., minerals

- The sub-samples collected at different periods and kept at -40°C to -80°C
- The analytes in the sub-samples are measured at the same time ⟶ reduce measurement variation ⟶ smaller uncertainty

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How to get assigned value for in-house QC system?

1. Analyse required component(s) in 10 samples (or at least 7), *on different days*, under repeatable conditions
2. Calculate the mean and standard deviation (N=10)
3. Record mean and standard deviation on a graph paper, including warning (±2SD) and control lines (± 3SD)
4. Use the control sample in subsequent analysis and record the level of analyte on the control chart
5. Decide if the result of the control sample is acceptable, if not, do corrective action and repeat the whole set of samples.

Internal QC system: control chart
Example: total N analysis

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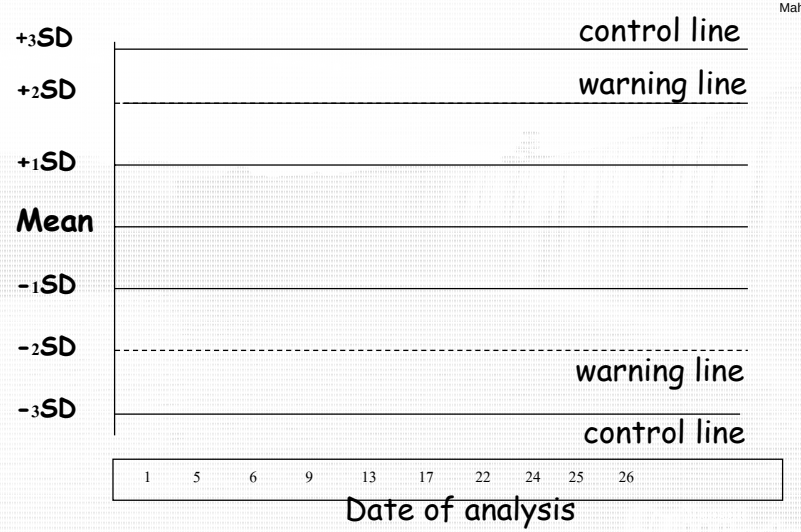

External QC system: establishing assigned values of nutrients in test material

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1) Formulation: known value
2) Certified reference values
3) Reference values
4) Consensus from expert laboratories
5) Consensus from participants in laboratory performance study (laboratory proficiency study, PT)

4) Consensus values by expert laboratories

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- best method of establishment
- obtained from a group of expert laboratories*
  using recognised reference methods
- information should be disclosed: identities of the expert laboratories, the methods used for analysis and calculation for the consensus values and a statement of the traceability and the uncertainty of the values
- Number of laboratories: complexity of measurement procedures
  1. 2-3 laboratories: primary methods – i.e., isotope dilution MS
  2. 6-8 laboratories: lower metrological quality – i.e., AAS, ICP
  3. >10 laboratories: less level of the quality - i.e., total N by Kjeldahl

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5) Consensus from participants in a round of PT scheme

Estimation of assigned values:

Algorithm A in Annex C of ISO 13528

Assigned value: Robust mean (x^*) ± robust SD (s^*),

Robust mean (x^*) ± uncertainty (u_x)

Standard uncertainty $u_X = \frac{1.25 \times s^*}{\sqrt{p}}$

s^* = the robust standard deviation of the results calculated using Algorithm A in Annex C.

p = number of participants report a measurement

Limitations:

- there may be no real consensus amongst the participants;
- the consensus may be biased by the general use of faulty methodology and this bias will not be reflected in the standard uncertainty of the assigned value

Case study from INMU: two approaches for establishing assigned values

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1. From expert laboratories: 10-15 lab using recognised reference methods: OCEANIAFOODS (Australia, New Zealand, Fiji), Europe (Austria, Holland), USA, Canada

2. From PT participants (with statistical accepted)

Estimation of assigned values: ISO 13528*

Robust average (x^*) ± Robust SD (S^*)

Robust average (x^*) ± Uncertainty (u_x)

⇨ See example

Ref: ISO 13528: 2005. Statistical methods for use in proficiency testing by interlaboratory comparisons. Geneva, Switzerland.

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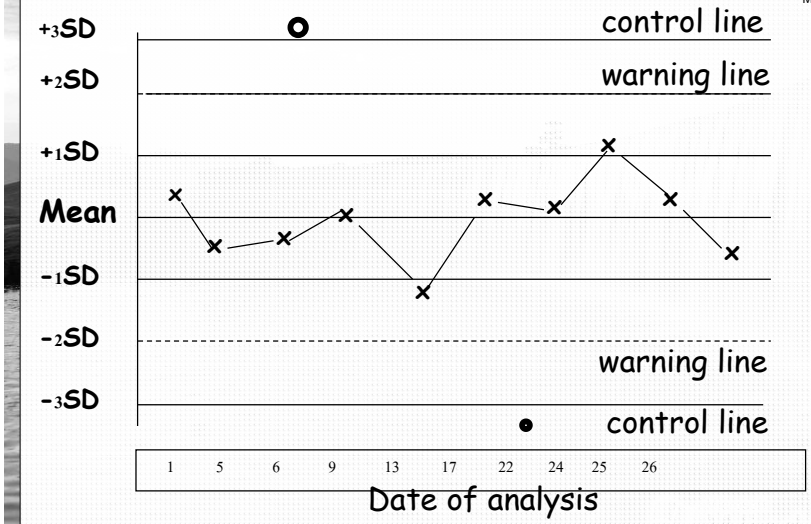

Shewhart Control Charts: ISO Approach

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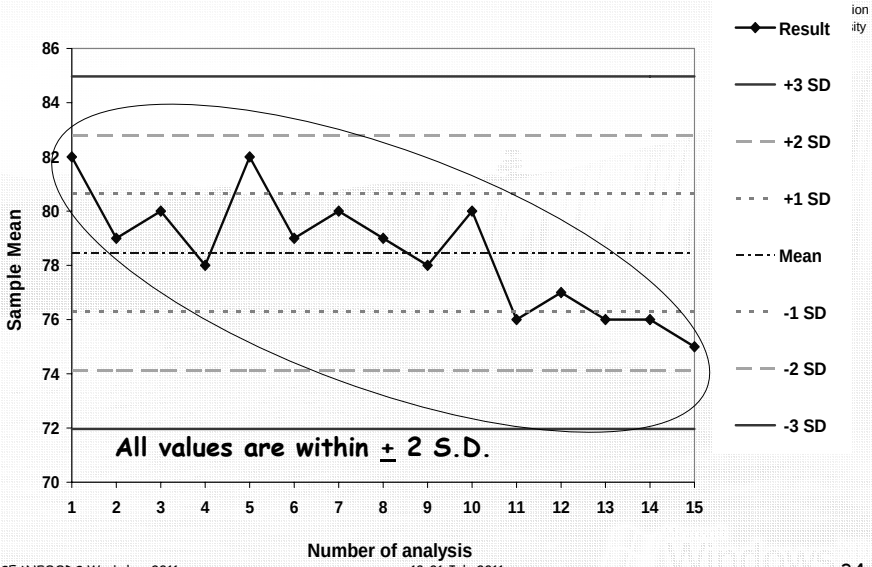
A control chart:

a means in applying statistical principle to control a process of analytical measurement

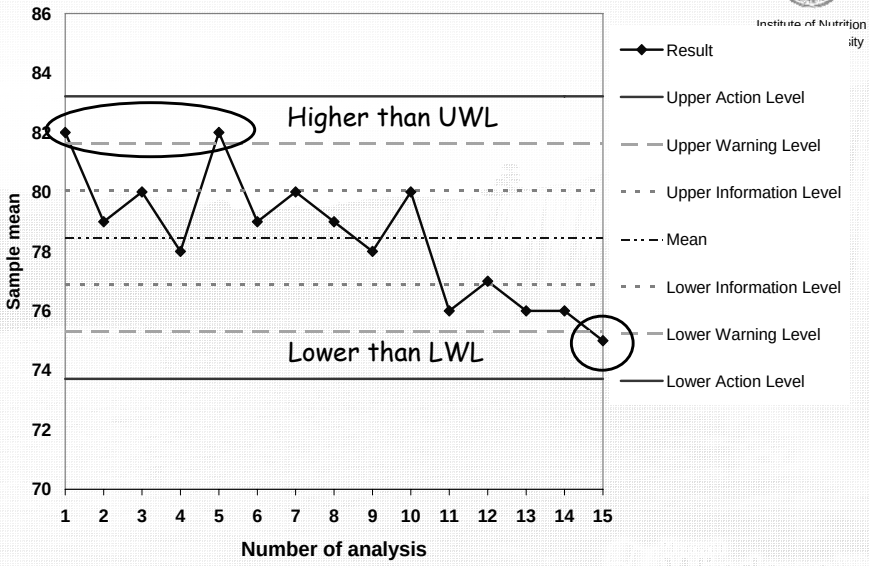
References:

- **ISO Standard 8258:1991, Shewchart Control Charts**
- **ISO Standard 7870:1993, Control Charts-General Principles**

Shewhart Control Chart: use Mean±SD



Shewhart Control Chart: ISO approach



Internal and external quality control system

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Process for laboratory performance study: PT

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Test materials with sufficient homogeneity, with or without assigned values

- Instruction to participants
- Report form
- Questionnaire on methods used
- Questionnaire on internal and external quality system

+ documents

Participating laboratories (private and governmental)

Analysis of components (measurands)

Assigned values

Evaluation of results using z-score

Good performance lab ← Technical meeting → Unacceptable performance Lab

Training/consultation, discussion/exchange experience& information

Establishing assigned values of measurands in PT test materials and reference values of measurands in Reference Materials for long-term QC Programme

Individual data of protein in Rice-1 (N=50) (with uncertainty values from 21 lab)

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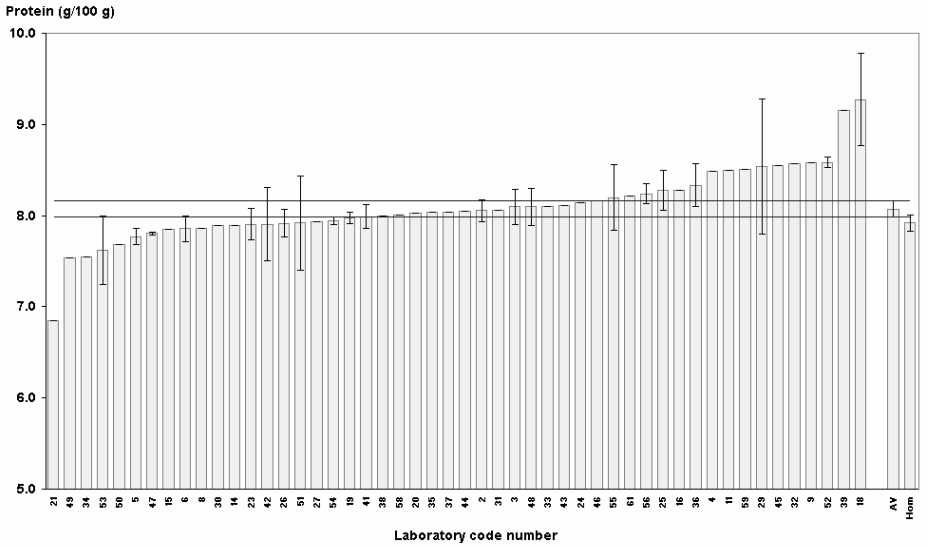

Laboratory performance by estimation of robust z-score

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1. Within laboratory variation (see example)

$$z\text{-score}_{within} = \frac{x - median_{(diff)}}{NIQR_{(diff)}}$$

x = Difference between reported values of A and B from each laboratory/SQRT 2

Median = Median of the difference between reported values of A and B (x)

NIQR = Normalised Inter-quartile Range

= (Quartile 3-Quartile 1) x 0.7413

Interpretation

$|z| \leq 2$ Satisfactory result ("a")

$2 < |z| < 3$ Questionable result ("w")

$|z| \geq 3$ Unsatisfactory result ("ww")

w: within laboratory z-score

2. Between laboratory variation (see example)

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$$\text{Robust z-score} = \frac{x_i - \overline{X}}{SD}$$

where x = average value of reported data from each participant

$\overline{X}$ = robust mean (x^*) obtained by ISO 13528 or median of the mean values obtained from participants

SD = robust standard deviation (s^*) obtained by ISO 13528 or predicted SD of Horwitz

Interpretation

$|z| \leq 2$ Satisfactory (acceptable) result ("a")

$2 < |z| < 3$ Questionable (warning) result ("b")

$|z| \geq 3$ Unsatisfactory (unacceptable) result ("bb")

b: between laboratory z-score

LABORATORY PERFORMANCE STUDY: evaluate against assigned value of test material

Assigned value (robust mean + robust SD) of protein in Rice-1 = 8.07±0.25 g/100g (% CV = 3.1)

Laboratory	Result		*Di*	*Si*	z- score		z-score	Conclusion	Values for
Code No	A	B	(B-A)/SQRT2	(A+B)/SQRT2	Within	Between(1)	Between(2)	based on (2)	consensus
2	8.05	8.06	0.01	11.39	0.29	0.05	-0.06	*a*	8.06
3	8.10	8.09	-0.01	11.45	0.00	0.20	0.10	*a*	8.10
4	8.47	8.51	0.03	12.01	0.73	1.68	1.68	*a*	8.49
8	7.97	7.99	0.01	11.29	0.44	-0.24	-0.36	*a*	7.98
18	9.24	9.30	0.04	13.11	1.02	**4.62**	**4.80**	***bb***	
19	7.97	7.98	0.01	11.28	0.29	-0.25	-0.38	*a*	7.98
20	8.04	8.01	-0.02	11.35	-0.29	-0.07	-0.18	*a*	8.03
31	8.09	8.02	-0.05	11.39	-0.88	0.05	-0.06	*a*	8.06
32	8.54	8.6	0.04	12.12	1.02	1.98	2.00	*a*	8.57
33	8.11	8.1	-0.01	11.46	0.00	0.24	0.14	*a*	8.11
34	7.67	7.43	-0.17	10.68	**-3.35**	-1.85	***-2.08***	***wwb***	
35	8.03	8.04	0.01	11.36	0.29	-0.03	-0.14	*a*	8.04
36	8.35	8.32	-0.02	11.79	-0.29	1.10	1.06	*a*	8.34
37	8.02	8.06	0.03	11.37	0.73	-0.01	-0.12	*a*	8.04
38	8.03	7.97	-0.04	11.31	-0.73	-0.16	-0.28	*a*	8.00
39	9.23	9.07	-0.11	12.94	***-2.19***	**4.16**	**4.32**	***wbb***	
41	8.00	7.98	-0.01	11.30	-0.15	-0.20	-0.32	*a*	7.99
42	7.91	7.9	-0.01	11.18	0.00	-0.52	-0.66	*a*	7.91
43	8.12	8.1	-0.01	11.47	-0.15	0.25	0.16	*a*	8.11
44	7.76	8.33	0.40	11.38	**8.46**	0.01	-0.10	***ww***	
45	8.57	8.52	-0.04	12.08	-0.58	1.89	1.90	*a*	8.55
50	7.70	7.67	-0.02	10.87	-0.29	-1.34	-1.54	*a*	7.69
51	7.98	7.86	-0.08	11.20	-1.60	-0.46	-0.60	*a*	7.92
52	8.56	8.61	0.04	12.14	0.88	**2.04**	***2.06***	***b***	
53	7.57	7.67	0.07	10.78	1.60	-1.59	-1.80	*a*	7.62
54	7.95	7.94	-0.01	11.24	0.00	-0.37	-0.50	*a*	7.95
55	8.21	8.18	-0.02	11.59	-0.29	0.57	0.50	*a*	8.20
61	7.82	8.61	0.56	11.62	**11.67**	0.65	0.58	***ww***	
No of results	50	50	50	50				N	40
Median	8.03	8.04	**-0.01**	**11.37**				Mean	8.07
Q1	7.90	7.91	-0.03	11.18				SD	0.23
Q3	8.29	8.31	0.03	11.69				%CV	2.88
IQR (Q3-Q1)	0.39	0.40	0.07	0.51					
Normalised IQR			**0.05**	**0.38**					
Robust CV				**3.31**					

<--- show only some data

"a" = accepted value; z-score ≤2

"w" or "b" = questionable; 2 < z-score > 3

"ww" or "bb" = outliers; z-score ≥ 3

(w = within lab, b = between lab)

Final consensus value for protein in Rice-1

calculated from laboratories with accepted values

=**8.07±0.23 g/100g** (mean±SD), with N=40,%CV=2.9

(1) based on median±NIQR; (2) based on assigned value estimated according to ISO 13528

Conclusion

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- Users of food composition database needs reliable data
- Food composition data generator should conduct daily in-house quality control system and regularly participate in proficiency testing
- Laboratory accreditation: ISO 17025 requires both internal and external quality control system
- Thus, PT providers and food reference materials producers, to cover various matrices and nutrients, are urgently needed in ASEAN Network of Food Data System.
- Present status of PT provider: Thailand, Indonesia, Vietnam, Philippines, Singapore, Malaysia?