Analytical Performance of Laboratories in Asia Pacific Food Analysis Network - APFAN PT2 (2018-2019): Report and Discussion

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Wisdom of the Land

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Final Report of APFAN PT-2 (2019):

Defatted Soybean Flour,
Fishmeal,
Natural Rice Flour,
Spiked Rice Flour

Download final report at
http://www.inmu.mahidol.ac.th/aseanfoods/apfan/program.php

Report, results and discussion are presented only for defatted soybean flour

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Outline

1. Objectives of study
2. Selection and preparation of test materials
3. Materials and methods
4. Testing homogeneity of representative nutrients
5. Establishing assigned values of measurands
6. Evaluation of lab performance
7. Establishing reference values of measurands in the test material, to become RM from PT

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Important steps in the proficiency testing

1. Selection and preparation of test materials

↓ Defatted soybean flour

Sample preparation → sub-samples and vacuum pack

↓

2. Testing homogeneity of representative nutrients*

*Follow ISO Guide 34, 35, ISO 5725 & 13528

↓ Test material + E-documents

Participating laboratories

↓

Analyses of nutrients

↓ results

**Robust statistics and ISO 13528

3. Establishing assigned values of measurands*

↓

4. Evaluation of lab performance: robust z- score

*Follow ISO/IEC 17043

↓

Good performance lab → Unsatisfied performance lab

↑

5. Technical meeting

↓

6. Establishing reference values of measurands in the test material to become RMs

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Objectives of this study

- To assess laboratory performance on analyses of nutrients (Moisture, Total nitrogen, Total fat, Dietary fibre, Ash, Calcium, Magnesium, Phosphorus, Sodium, Potassium, Iron, Zinc, Copper) in defatted soybean flour
- To develop PT food reference materials with reference values of nutrients, from good performance laboratories, to be used for internal quality control system and method validation

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Test material

10 kilograms of the fine particles (<250 μm) defatted soybean flour were purchased from Kasetsart University, Thailand.

↓

Sample preparation

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Materials and methods (cont.)

Sample preparation

Transfer sample to mixing machine & mix for 3 hrs.

Aliquot 1-2 kg

Packing (~50 g/bag)

Vacuum seal

Test material

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Materials and methods (cont.)

Sample preparation

Particle size distribution

10 packages

Sampling

Storage sample
at -20^0 C

Random sampling

Homogeneity testing

Moisture, total nitrogen, dietary fibre, ash

One package provides to each participating laboratory

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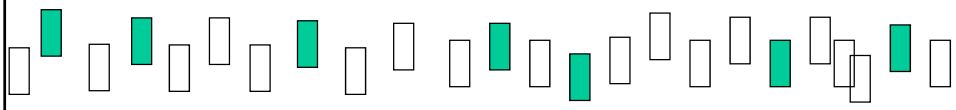
Testing for sample homogeneity

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



selected 10 representative samples at random
(stratified or systematic method)

ISO 17025 Accredited lab

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

Statistical analysis without exclusion of any value

*on the same day using the same reagents, instrument, conditions, and the same analysts for each parameter

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Statistical analysis: homogeneity testing

2 Steps:

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

Step 2. Checking for between sample variation (check homogeneity of the test materials) using
- ISO 13528:2005

$$s_s \leq 0.3\sigma_{pt}$$

The test materials is adequately homogeneous.

"Sample variation will not significantly affect the between laboratory results"

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

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Step 1. Checking for within sample variation: Cochran's test

Duplicate analysis:
Cochran's maximum range test

Calculate the ratio: $\frac{D_{max}^2}{\sum D_i^2}$ or $\frac{R_{max}^2}{\sum R_i^2}$

where
D_{max} = *the maximum difference of the duplicates (or R)*
D_i = *difference of each pair of duplicates*

- Compare the ratio to the critical value from Cochran Table (95% confidence).
- If the ratio is < critical value, there is no evidence of analytical outliers

- An outlier set should not be rejected unless it is significant at 99% level or any permanent analytical errors are found.

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Table 1. Summary sample homogeneity testing of defatted soybean flour: within sample variation (Cochran's test)

No.	Moisture (g/100g)				Total protein (g/100g)				Ash (g/100g)				Dietary fibre (g/100g)			
	A	B	Range² (R)	R/R-total	A	B	Range² (R)	R/R-total	A	B	Range² (R)	R/R-total	A	B	Range² (R)	R/R-total
1	8.09	8.05	0.002	0.11	49.87	49.97	0.010	0.05	6.28	6.12	0.026	0.07	15.75	16.09	0.115	0.04
2	8.00	8.02	0.000	0.03	49.96	49.79	0.029	0.15	6.25	6.12	0.017	0.04	16.34	16.27	0.004	0.00
3	8.05	8.06	0.000	0.01	49.91	49.87	0.002	0.01	6.22	6.22	0.000	0.00	16.42	16.46	0.001	0.00
4	8.09	8.08	0.000	0.01	49.80	49.77	0.001	0.00	6.37	6.19	0.032	0.08	16.35	16.66	0.096	0.03
5	7.99	7.96	0.001	0.06	49.71	49.69	0.000	0.00	6.40	6.26	0.020	0.05	16.16	16.78	0.386	0.13
6	8.12	8.02	0.010	0.67	49.79	49.92	0.017	0.09	6.41	6.17	0.058	0.15	16.19	16.91	0.508	0.17
7	7.99	7.98	0.000	0.01	49.92	49.82	0.010	0.05	6.44	6.32	0.014	0.04	16.41	15.99	0.176	0.06
8	7.91	7.91	0.000	0.00	49.84	49.74	0.010	0.05	6.17	6.34	0.029	0.07	17.16	16.04	1.241	0.42
9	8.01	7.98	0.001	0.06	49.93	49.63	0.090	0.48	5.96	6.40	0.194	0.49	16.65	16.10	0.309	0.10
10	8.04	8.01	0.001	0.06	49.85	49.71	0.020	0.10	6.29	6.34	0.002	0.01	16.35	15.99	0.124	0.04
R Total			0.015	Max =		R Total	0.188	Max =			0.392	Max =			2.961	Max =
N			10	0.67		N	10	0.48			10	0.49			10	0.42
Cochran critical value[a]	95% CI		0.602	Not pass			0.602	Pass			0.602	Pass			0.602	Pass
	99% CI		0.718	Pass			0.718	Pass			0.718	Pass			0.718	Pass

- An outlier set should not be rejected unless it is significant at 99% level or any permanent analytical errors are found.

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Statistical analysis: homogeneity testing

Step 2: Checking for between sample variation (check homogeneity of the test materials) using ISO 13528:2015

$$s_s \leq 0.3\sigma_{pt}$$

The test materials is adequately homogeneous.

"Sample variation will not significantly affect the between laboratory results"

1) ISO 13528: 2015 Statistical methods for use in proficiency testing of interlaboratory comparisons.

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Table 2. Summary sample homogeneity testing of defatted soybean flour: based on ISO 13528 (2005)

Parameter	Mean	Standard deviation (SD_r)	Relative standard deviation (RSD_r), %CV	Sampling standard deviation (*Ss*)	RSDp (%)*	σ_{pt}	0.3 σ_{pt}	Summary
Moisture (g/100g)	8.02	0.06	0.70	0.051	2.92	0.234	0.070	Pass
Protein (g/100g)	49.82	0.10	0.2	0.000	2.22	1.107	0.332	Pass
Ash (mg/kg)	6.26	0.12	1.93	0.000	3.03	0.190	0.057	Pass
Total Dietary fibre (g/100g)	16.35	0.34	2.1	0.000	2.63	0.430	0.129	Pass

* Target standard deviation using general model: "Horwitz' s equation"

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Determination of the assigned value

$$z = \frac{(x_i - x_{pt})}{\sigma_{pt}}$$

- 7.3 Formulation
- 7.4 Certified reference material
- ✓ **7.5 Results from one laboratory**
- 7.6 Consensus value from expert laboratories
- ✓ **7.7 Consensus value from participant results**

σ_{pt} = standard deviation for proficiency assessment

- 8.2 By perception of experts
- 8.3 By experience from previous rounds of a proficiency testing scheme
- ✓ **8.4 By use of a general model**
- 8.5 Using the repeatability and reproducibility standard deviations from a previous collaborative study of precision of a measurement method
- ✓ **8.6 From data obtained in the same round of a proficiency testing scheme**

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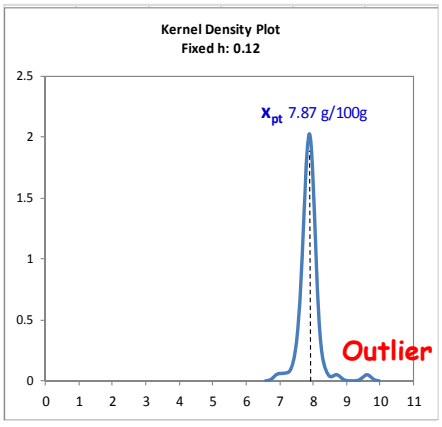
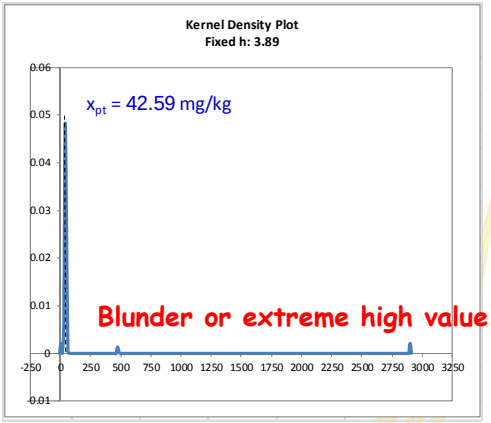
Visual review of data using Kernel density plot

- ✓ **A kernel density plot, one of a visualize tool, often more useful for identifying possible bimodalities or lack of symmetry.**
- ✓ **It should set bandwidth at 0.75 σ_{pt}.**



Total nitrogen



Zinc

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Determination of assigned values using approach of

"Consensus from participants in a round of PT scheme"

following Algorithm A in Annex C of ISO 13528: 2015

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

Robust mean (x^*) ± uncertainty (u_x)

Important steps:

Step 1. Review all data submitted by various laboratories.
This is to leave out the known extreme values due to:
- unaccepted analytical method,
- misplace of decimal point,
- wrong unit of expression, etc.

Step 2. Modify remaining submitted extreme low and high values (if any).
The process is conducted in Excel spread sheet (see next slide)

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Table3. Calculation of robust average and standard deviation (ISO 13528: 2015): Total nitrogen in defatted soybean flour

Iteration	0	|xi-x*|	1	2	3	4	5	6	7	8
δ = 1.5 s*	---		0.2302	0.2303	0.2307	0.2309	0.2311	0.2311	0.2311	0.2311
x* - δ	---		7.6463	7.6422	7.6412	7.6409	7.6407	7.6407	7.6406	7.6406
x* + δ	---		8.1067	8.1029	8.1026	8.1027	8.1028	8.1029	8.1029	8.1029
49	6.94	0.937	7.6463	7.6422	7.6412	7.6409	7.6407	7.6407	7.6406	7.6406
30	7.18	0.695	7.6463	7.6422	7.6412	7.6409	7.6407	7.6407	7.6406	7.6406
25	7.43	0.447	7.6463	7.6422	7.6412	7.6409	7.6407	7.6407	7.6406	7.6406
54	7.51	0.364	7.6463	7.6422	7.6412	7.6409	7.6407	7.6407	7.6406	7.6406
11	7.54	0.337	7.6463	7.6422	7.6412	7.6409	7.6407	7.6407	7.6406	7.6406
93	7.61	0.267	7.6463	7.6422	7.6412	7.6409	7.6407	7.6407	7.6406	7.6406
22	7.66	0.217	7.6600	7.6600	7.6600	7.6600	7.6600	7.6600	7.6600	7.6600
87	7.67	0.208	7.6690	7.6690	7.6690	7.6690	7.6690	7.6690	7.6690	7.6690
90	7.68	0.197	7.6800	7.6800	7.6800	7.6800	7.6800	7.6800	7.6800	7.6800
55	7.70	0.177	7.7000	7.7000	7.7000	7.7000	7.7000	7.7000	7.7000	7.7000
15	8.02	0.144	8.0200	8.0200	8.0200	8.0200	8.0200	8.0200	8.0200	8.0200
23	8.02	0.144	8.0200	8.0200	8.0200	8.0200	8.0200	8.0200	8.0200	8.0200
44	8.02	0.145	8.0210	8.0210	8.0210	8.0210	8.0210	8.0210	8.0210	8.0210
67	8.03	0.153	8.0300	8.0300	8.0300	8.0300	8.0300	8.0300	8.0300	8.0300
6	8.08	0.208	8.0840	8.0840	8.0840	8.0840	8.0840	8.0840	8.0840	8.0840
2	8.09	0.214	8.0900	8.0900	8.0900	8.0900	8.0900	8.0900	8.0900	8.0900
84	8.12	0.243	8.1067	8.1029	8.1026	8.1027	8.1028	8.1029	8.1029	8.1029
50	8.33	0.457	8.1067	8.1029	8.1026	8.1027	8.1028	8.1029	8.1029	8.1029
27	8.70	0.823	8.1067	8.1029	8.1026	8.1027	8.1028	8.1029	8.1029	8.1029
75	9.61	1.734	8.1067	8.1029	8.1026	8.1027	8.1028	8.1029	8.1029	8.1029
New x*	**7.8765**	0.104	**7.873**	**7.872**	**7.872**	**7.872**	**7.872**	**7.872**	**7.872**	**7.872**
New s*	**0.153**		**0.154**	**0.154**	**0.154**	**0.154**	**0.154**	**0.154**	**0.154**	**0.154**
p	66									
% Different of Robust SD =			0.04	0.17	0.10	0.05	0.02	0.01	0.00	0.00

Assigned value (robust mean ± robust SD) = 7.87 ± 0.15 g/100 g (%CV = 1.9%, N = 66)

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Choices of assigned values of test materials

- Robust mean (x*) ± robust SD (s*), estimated according to ISO 13528:2015 e.g. Moisture, Nitrogen, Ash
- Reference mean (X) obtained from NIMT using
  Isotope dilution ICP-MS; Cu, Zn
  Gravimetric standard addition addition IDMS; Ca, Mg, Fe
- If variation of data is too high (indicated by %RSD):
  Robust mean* ± target SD (SDp, Horwitz's predicted SD)
  e.g. Dietary fibre (x* ± 3SDp), Calcium (x* ± 2SDp),
- Median ± Normalised IQR** e.g. Iron

*estimated from ISO 13528 or use median value (not affected by extreme values) of the data obtained from participants

IQR = Q3-Q1

** Normalised IQR = Inter Quartile Range x 0.7413

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Table 4. Assigned values of measurands for evaluation of testing parameters in defatted soybean flour

Parameters	Method of assigned	x_{pt}	σ_{pt}	%RSD	$u_{x(pt)}$	$0.3\sigma_{pt}$	$u_{x(pt)}$ is negligible?
Moisture (g/100g)	x* & s*	7.26	0.71	9.8	0.10	0.21	Yes, use z score
Total nitrogen (g/100g)	x* & s*	7.87	0.16	2.0	0.02	0.05	Yes, use z score
Fat (g/100g)	x* & s*	1.41	0.71	50.4	0.14	0.04	No, Not evaluate
Ash (g/100g)	x* & s*	6.31	0.33	5.2	0.05	0.10	Yes, use z score
Total dietary fibre	x* & $3SD_p$	16.44	1.29	7.9	0.10	0.39	Yes, use z score
Calcium (mg/kg)	x* & $2SD_p$	2031	207	10.2	35.8	62.0	Yes, use z score
(reference value)	X & $2SD_p$	2100	207	9.8	28.6	62.0	Yes, use z score
Magnesium (mg/kg)	x* & s*	2652	343	12.9	62.5	102.8	Yes, use z score
(reference value)	X & SD_p	2650	259	9.8	37.8	102.8	Yes, use z score

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Table 4. Assigned values of measurands for evaluation of testing parameters in defatted soybean flour (con't)

Parameters	Method of assigned	x_{pt}	σ_{pt}	%RSD	$u_{x(pt)}$	$0.3\sigma_{pt}$	$u_{x(pt)}$ is negligible?
Phosphorus (mg/kg)	x* & s*	7787	456	5.9	99.3	136.9	Yes, use z score
Sodium (mg/kg)	x* & $3SD_p$	72.5	18.3	25.2	4.0	5.5	Yes, use z score
Potassium (mg/kg)	x* & $3SD_p$	23133	2447	10.6	437	734	Yes, use z score
Iron (mg/kg)	Med. & NIQR	75.50	8.78	11.6	1.54	2.63	Yes, use z score
(reference value)	X & SD_p	75.1	6.4	8.5	0.9	2.63	Yes, use z score
Zinc (mg/kg)	x* & s*	42.59	5.18	12.2	0.95	1.55	Yes, use z score
(reference value)	X & SD_p	43.1	3.9	9.0	0.6	1.55	Yes, use z score
Copper (mg/kg)	x* & SD_p	12.19	1.34	11.0	0.25	0.40	Yes, use z score
(reference value)	X & SD_p	12.5	1.3	10.7	0.2	0.40	Yes, use z score

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Laboratory performance evaluation: robust *z score*

Between laboratory variation

$$z = \frac{(x_i - x_{pt})}{\sigma_{pt}}$$

where x_i = average value of reported data from each participant

x_{pt} = the assigned value i.e. robust mean (x^*) obtained from ISO 13528 or median of the values obtained from participants

σ_{pt} = standard deviation for proficiency assessment i.e. robust standard deviation (s^*) obtained from ISO 13528 or predicted SD from Horwitz's equation

Interpretation

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

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

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

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Laboratory performance: Zeta score

- Zeta scores can be useful when an objective for the proficiency testing scheme is to evaluate a participant's ability to have results be close to the assigned value within their claimed uncertainty.

$$\zeta_i = \frac{(x_i - x_{pt})}{\sqrt{u_{x_i}^2 + u_{x_{pt}}^2}}$$

where x_i = value of reported data from each participating laboratory;

x_{pt} = the assigned value i.e. robust mean (x^*), median, reference value;

u_{x_i} = the participant's estimate of the standard uncertainty of its result x_i;

$u_{x_{pt}}$ = the standard uncertainty of the assigned value x_{pt}

- ζ scores above 2 or below -2 may be caused by systematically biased methods or by a poor estimation of the measurement uncertainty by the participant.
- ζ scores therefore provide a rigorous assessment of the complete result submitted by the participant.

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RESULTS: DATA DISTRIBUTION AND EVALUATION

"Moisture"

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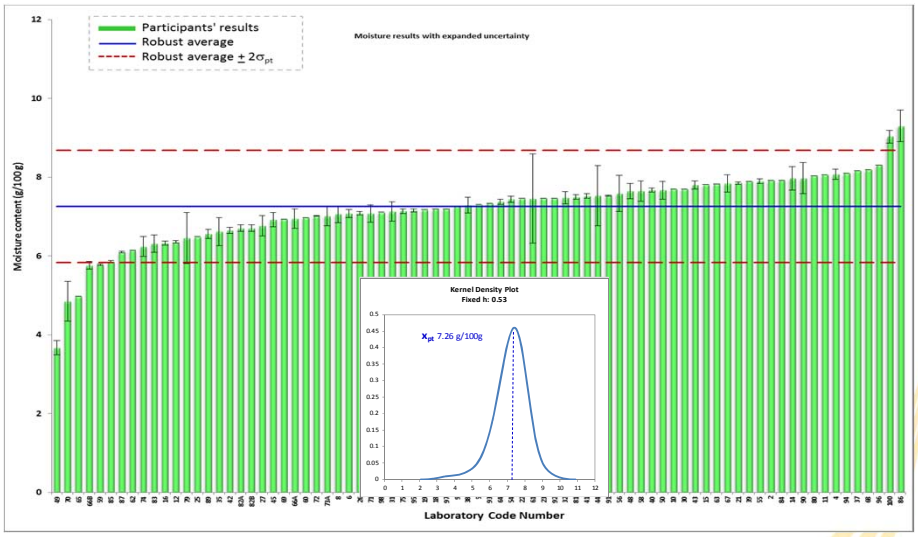

Figure 1. Distribution of moisture results (ascending order) in defatted soybean flour with expanded uncertainty

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Table 5. Example of individual data of Laboratory performance on moisture analysis

Laboratory Number	Moisture g/100g, as received	MU g/100g, as received	z score	Zeta score	Sample weight (g)	Temperature (°C)	Time (Hours)	Method Reference
Assigned value obtained from robust average ± robust SD = 7.26 ± 0.71 g/100 g (CV 9.8%, n= 79)								
Acceptance criteria =			|z score| ≤ 2.00	|ζ score| < 2.00				
2	7.92	-	0.93	-	2.00	135	2	AOAC (2016) 930.15
4	8.07	0.13	1.14	6.79	2.xxxx	130 ± 3	1	Based on AOAC
9	7.28	-	0.03	-	5 to 10	105	4	Based on ISO 6496:1999
10	7.70	-	0.62	-	2	130	1	AOAC 2012, 32.2.09 A, Chapter 32
11	8.06	-	1.13	-	5.0000	103	4	AOAC (2016) 925.10
12	6.36	0.03	-1.27	-8.90	1	100 ± 1	3	AOAC (2016) 952.08
48	7.65	0.19	0.54	2.79	2	130	1	SNI 3549 2009
49	3.67	0.18	-5.06	-26.68	2	130	1	AOAC 20th Ed 2016
50	7.67	0.23	0.58	2.71	2.1804	130	1.0	AOAC 925.10
54	7.44	0.08	0.25	1.67	1	105	5	AOAC 927.05
55	7.90	0.06	0.90	6.11	5	130	1	AOAC (2012) 945.39A
56	7.59	0.46	0.46	1.32	2.03250	130 ± 3	1	AOAC Intl 20th Ed, 2016 925.10
58	7.65	0.26	0.55	2.38	2 to 5	130 / 105	3	Based on AOAC 20th Ed 2016
59	5.78	0.02	-2.08	-14.73	1 to 2	105	3	SNI 01-2891-1992 point 5.1
60	6.98	-	-0.39	-				SNI 01-2891-1992 Butir 5.1
61	7.46	1.13	0.28	0.35	3	130	1.5	A6801 130C Air oven
62	6.15	-	-1.56	-				
63	7.83	-	0.80	-				
64	7.37	0.06	0.16	1.09	2.0577	130	1	AOAC 925.10
65	4.98	-	-3.21	-	4.8006	105	2.5	Oven drying
66A	6.95	0.25	-0.44	-1.94	10.0032	130.0	0.50	AOCS Official Method Ca 2c-25, 7th Ed., 2017
66B	5.76	0.10	-2.11	-13.42	10.0010	130.0	0.50	AOCS Official Method Ca 2c-25, 7th Ed., 2017
67	7.84	0.22	0.82	3.90	2.0xxx	130	1.0	AOAC 925.10

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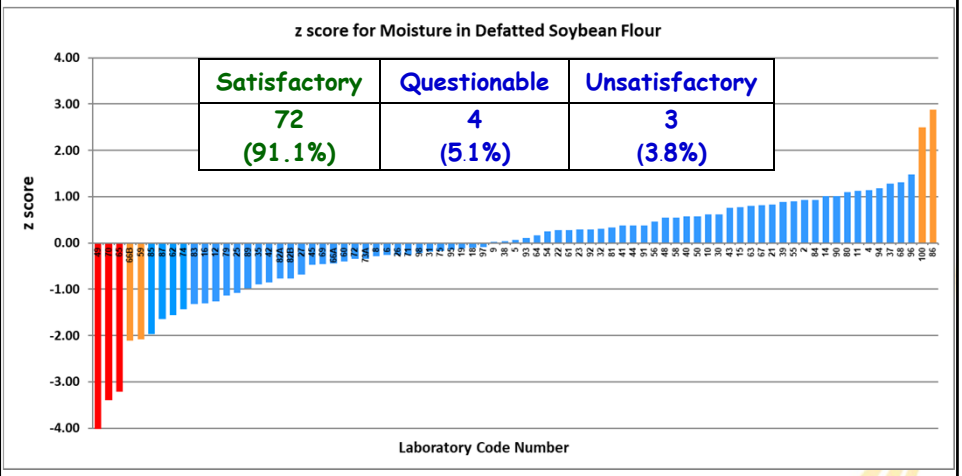
Figure 2: Laboratory performance on **moisture** analysis: Distribution of z-scores:



Satisfactory	Questionable	Unsatisfactory
72 (91.1%)	4 (5.1%)	3 (3.8%)

Unsatisfactory results ($|z| \geq 3$): Lab # 49, 70, 65
Questionable results ($2 < |z| < 3$): Lab # 66B, 59, 100, 86

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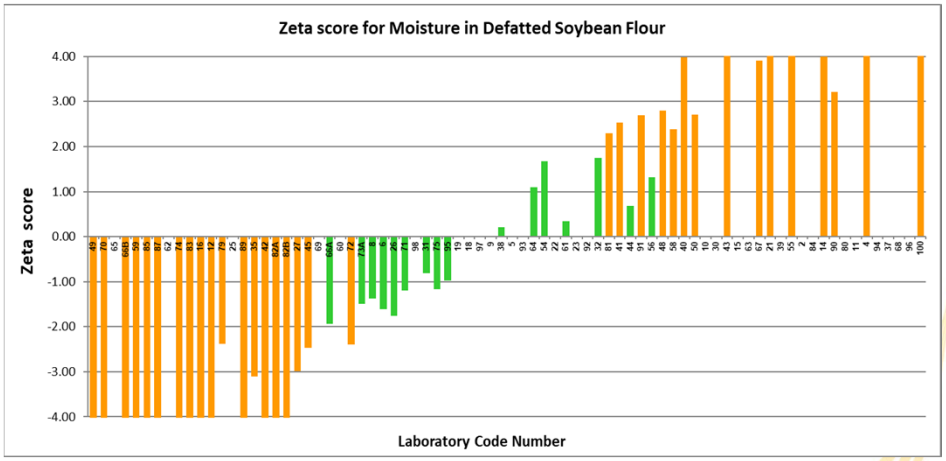
Figure 3. (example): Distribution of **Zeta score; moisture** in defatted soybean flour



Satisfactory	Questionable	Unsatisfactory
16 (20.3%)	35 (44.3%)	25 (31.6%)

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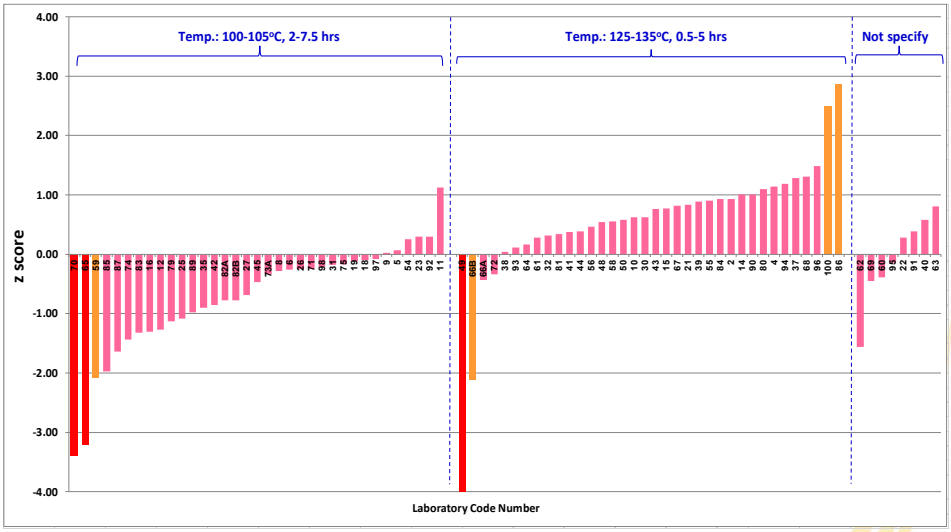

Figure 4. Plot of ordered *z* score: **Moisture** in defatted soybean flour, - categorised in groups according to drying temperatures

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Discussion and Interpretation:

"Moisture"

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Discussion and Interpretation: moisture determination in defatted soybean flour

Participants: **78 Lab**

- Participants followed three standard methods
  - **51% dried the sample in an oven at 125-135°C, for 1-5 h (AOAC 925.10, 2016).**
  - **Others dried at 100-105°C for 2 to 7.5 h (ISO 6496 and SNI 01-2891-1992).**
  - Drying the sample at difference temperatures has generally no effect on the performance of moisture analysis based on |z-scores| ≤ 2 but those who dried the sample at 100-105°C reported lower levels of moisture
- Three laboratories who reported extreme low levels of moisture applied the same drying temperatures (105°C for 2.5-5 h and 130°C for 1 h) and used the sample of 2-5 g which are the common practices of 91% of the good performance laboratories; thus, the reported values are real extreme values.

Recommendation: drying the sample until constant weight is strongly recommended for moisture determination.

The reference value of moisture content in the defatted soybean flour obtained from 70 good performance laboratories is 7.27 ± 0.57 g /100 g (Mean ± SD, %CV= 7.8)

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Results, Data distribution and Evaluation:

"Total Nitrogen"

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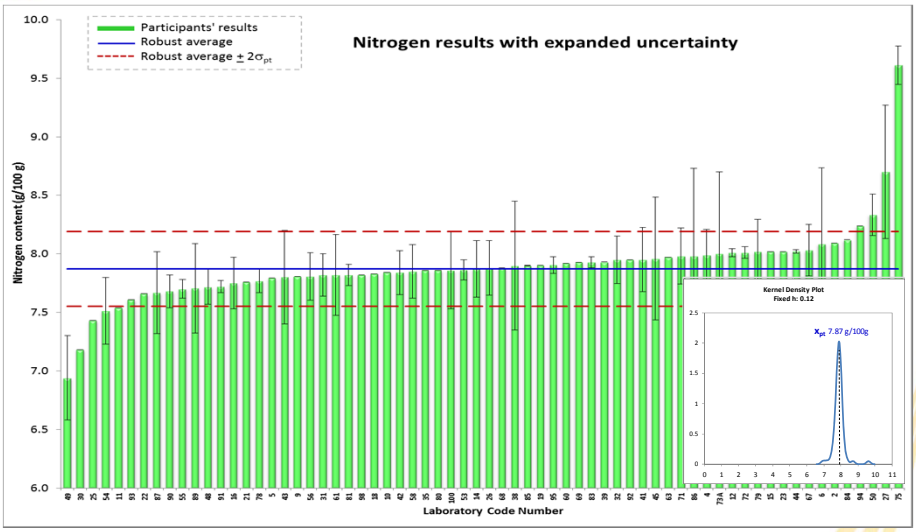

Figure 5: Distribution of total nitrogen results (ascending order) in defatted soybean flour with expanded uncertainty

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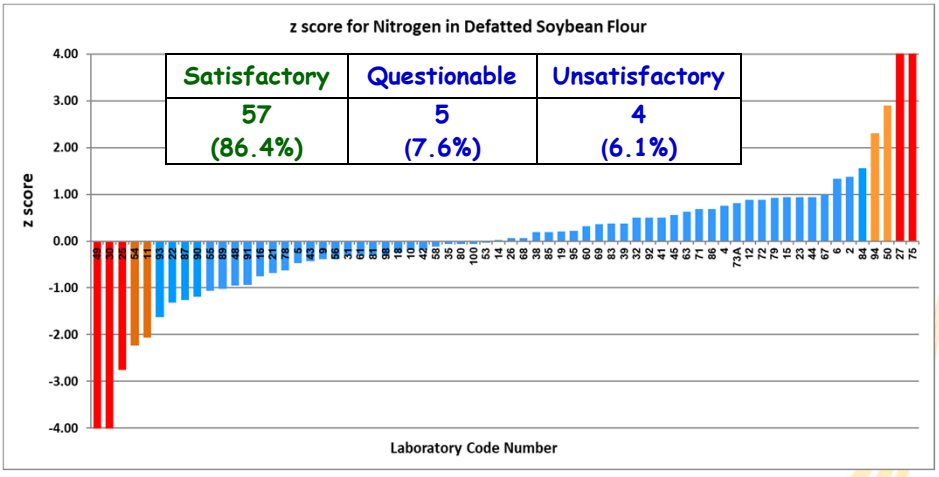
Figure 6: Laboratory performance on **total nitrogen** analysis



Unsatisfactory results (| z | ≥ 3): Lab # 49, 30, 25, 27, 75
Questionable results (2 < | z | < 3): Lab # 54, 11, 94, 50

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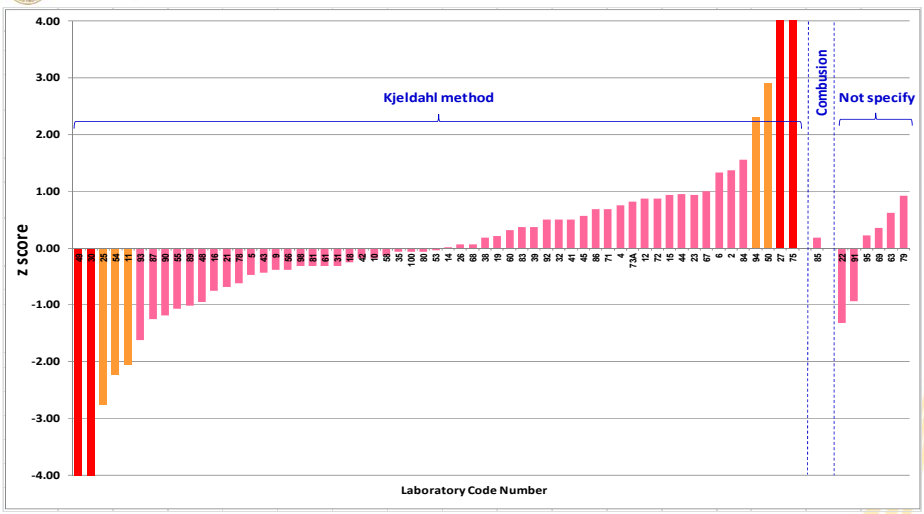

Figure 7. Plot of ordered z score for total nitrogen in defatted soybean flour, categorised in groups according to analytical methods/parameters used

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Discussion and Interpretation: Total nitrogen determination: 66 Lab

- To avoid an error in using different conversion factors to calculate protein from total N; this PT study evaluated performance on protein analysis from reported total N
- Laboratories use Kjeldahl method for total N determination, except one use combustion method (DuMaster Protein Analyzer, Buchi).
- Results: as shown in Figure – performance on Total N Determinatiom
  - 56 out of 66 lab (86%) were identified as good lab performance
  - Nine out of 66 Lab (14%) reported extremely high or low or questionable values of total N.

    ‹ These lab used Kjeldahl method for total N analysis, following the standard methods of AOAC, ISO and SNI (Soy Nutrition Institute).
    ‹ Used similar range of sample weight, 0.2 to 2 g (except one lab used 0.05 g reported questionable low value).
    ‹ Thus, each step on total N analysis must be critically reviewed and corrective action must be performed at these laboratories.

- From the Thai FCTs 2015 (adjusted moisture content): defatted soybean flour contains 8.31 g of total N/100 g

The reference value of total N content in the defatted soybean flour obtained from 56 good performance laboratories is 7. 88 ± 0.12 g /100 g (Mean ± SD, %CV= 1.5)

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Note to PTP on total protein determination:

- Although total N was used in this study for assessment of analytical performance, some PT studies may request to report total protein.
- The usual conversion factors used to convert total N to protein for defatted soybean reported by 27 laboratories are:

Total No. of Lab (n= 27)	N Conversion Factor used
18 (67%)	6.25
6 (22%)	5.71
3 (11 %)	5.95

- This evidence emphasises that the difference in N conversion factor used among participants could be one of the important factors for the discrepancy of the reported levels of protein in some PT programs.
- Several N-conversion factors for soy protein among various references, i.e., AOAC, ISO, and SNI are specified. Thus, the factors for soybean and other protein foods should be harmonised based on the scientific evidence from research.
- In 2010, the International Network of Food Data System (INFOODS) published a book – FAO/INFOODS "Guidelines for Checking Food Composition Data prior to the Publication of a User Table/Database" - Version 1.0 <http://www.fao.org/3/ap810e/ap810e.pdf> - which includes the N conversion factors used for various foods.

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Results, Data distribution and Evaluation:

"Total Fat"

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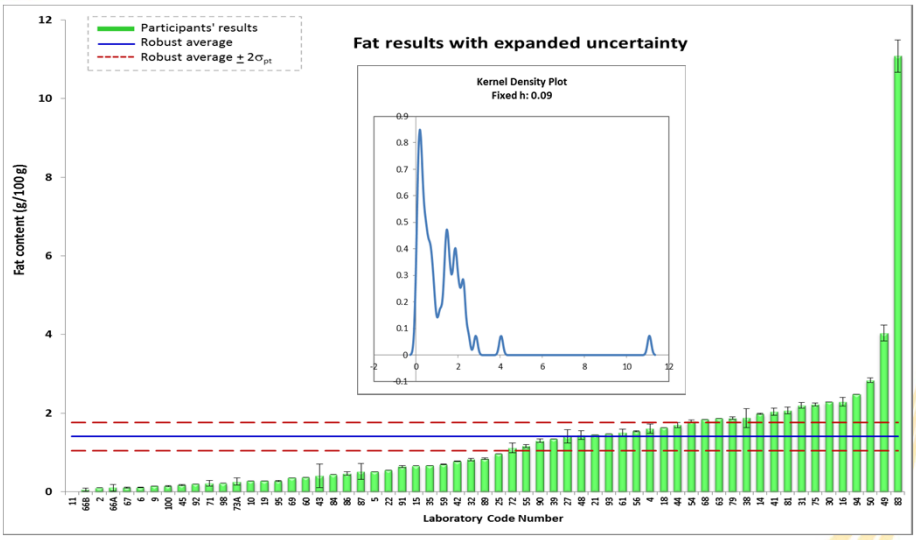

Figure 8: Distribution of total fat results (ascending order) in defatted soybean flour with expanded uncertainty

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Table 6: Laboratory performance on total fat analysis (information only)

Laboratory Number	Total fat (g/100g)	MU (g/100g)	z score	Zeta score	Fat weight (g)	Hydrolysis (Y/N)	Extraction Solvent	Extraction Time (hours)	Method Reference
11	0.00	-	Not evaluate due to high variation of results		4.0000	N	Petroleum Ether	16-18 hr	AOAC (2016) 922.06
12	< LOD	-			2	Yes, Acid digestion	Petroleum Ether	2	AOAC (2016) 984.15
14	1.98	0.02			2	Y	Diethyl Ether and Petroleum Ether	3	AOAC 922.06
15	0.66	-			1	Y	Petroleum Ether	1	Based on ISO 1443:1973
16	2.29	0.11			2	Y	Diethyl Ether + Petroleum Ether	-	SNI 01-2891-1992 Food & Beverage
18	1.64	-			2.0	Y	Petroleum Benzine	6	SNI 01-2891-1992
19	0.28	-			1	N	Petroleum Benzene	1	AOAC 2003.05, AN305, FOSS, 2005
21	1.45	-			0.5 to 1.0	Y	Diethyl Ether Petroleum Ether	2	AOAC 932.06 (2016)
22	0.55	-			-	-	-	-	-
23	< LOR	-			1.90	N	Petroleum Ether	1	AOCS Am5-04
25	0.97	-			7.1745	N	Hexane	8	Laboratory Handbook of Methods of Food Analysis, 3rd Ed, R. Lees
27	1.41	0.17			2	Y	Diethyl Ether	2	SNI 2354-3:2017

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Discussion: Total Lipid determination: 66 Lab

Test material Defatted soybean flour:

- Contains low level of total lipid - less than 2 g/100g*.
- For lipid determination of plant origin, e.g., cereals, legumes seeds: acid hydrolysis prior to direct extraction with non-polar solvent, to break the lipid-carbohydrate bond and to hydrolyse protein, is required for sample treatment.
- In this study, 22 out of 55 of participating laboratories (40 %) did not include the acid hydrolysis in the process for lipid analysis.
  - Most of them reported very low levels of total lipid content, ranged from 0 to less than 0.5 g/100 g.
  - Those who included acid treatment prior to solvent extraction reported higher level of total lipid, 1.49 ± 0.67 g)/100 g (mean±SD) but with wide variation (%CV = 45)
- Due to the residual fat in the DFF test material is quite low, resulting in too wide variation of fat values reported by each laboratory. Lab performance on lipid analysis in defatted soybean flour could not be evaluated.

Ref: Thai Food composition database 2018 <http://www.inmu.mahidol.ac.th/thaifcd/home.php>

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Results, Data distribution and Evaluation:

"Total Dietary fibre"

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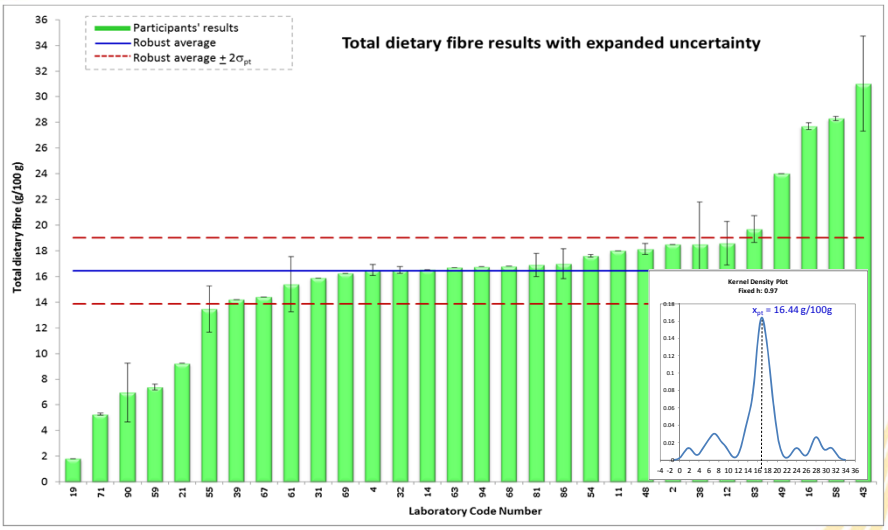

Figure 9: Distribution of dietary fibre results (ascending order) in defatted soybean flour with expanded uncertainty

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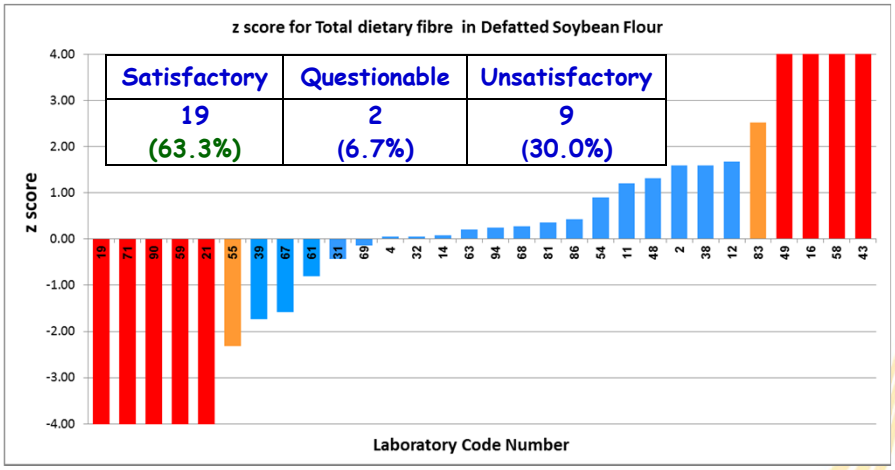
Figure 10: Laboratory performance on **dietary fibre analysis**



Unsatisfactory results (| z | ≥ 3): Lab # 19, 71, 90, 59, 21, 49, 16, 58, 43
Questionable results (2 < | z | < 3): Lab # 55, 83

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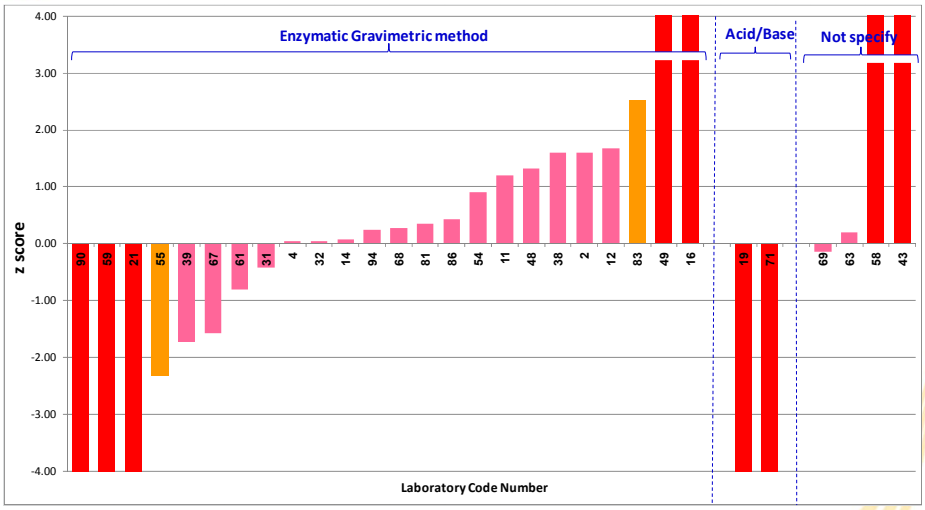

Figure 11. Plot of ordered z score for dietary fibre in defatted soybean flour, categorised in groups according to analytical methods/parameters used

Discussion: Dietary fibre determination: 30 Lab

- Using the defatted soybean flour as the test material, the step for fat removal is omitted
- The AOAC enzymatic gravimetric methods was mainly used for determination of total DF in 0.5 to 1 g of defatted soybean flour (except one good performance lab used 0.3 g).
- 19 laboratories (63%) were identified as good performance laboratories; 4 and 5 laboratories (30%) reported extremely high and low levels, respectively and 2 of them reported questionable levels.
- Activities and purity of enzymes alpha-amylase, protease and amyloglucosidase which involve in removing starch and protein in the sample are the critical factors in the determination of total DF.
  The activities of these enzymes must be checked for each of new lot or at maximal interval of 6 months. Applying the degraded enzymes could affect the efficiencies in removing other components from the sample, resulting in extreme high values of the final residue of the DF.
- Three laboratories used the AOAC methods for crude fibre or ISO method for neutral detergent dietary fibre determination; as expected they reports extremely low levels of DF
- It was found in the previous PT programmes, some laboratories did not corrected the amount of protein and ash from the residue; they reported extreme high values of DF.

The reference value of DF content in defatted soybean flour, obtained from 19 good performance laboratories , is ***16.80 ± 1.26 g/100 g (Mean±SD, N=19, %CV=7.5)***

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Results, Data distribution and Evaluation:

"Ash"

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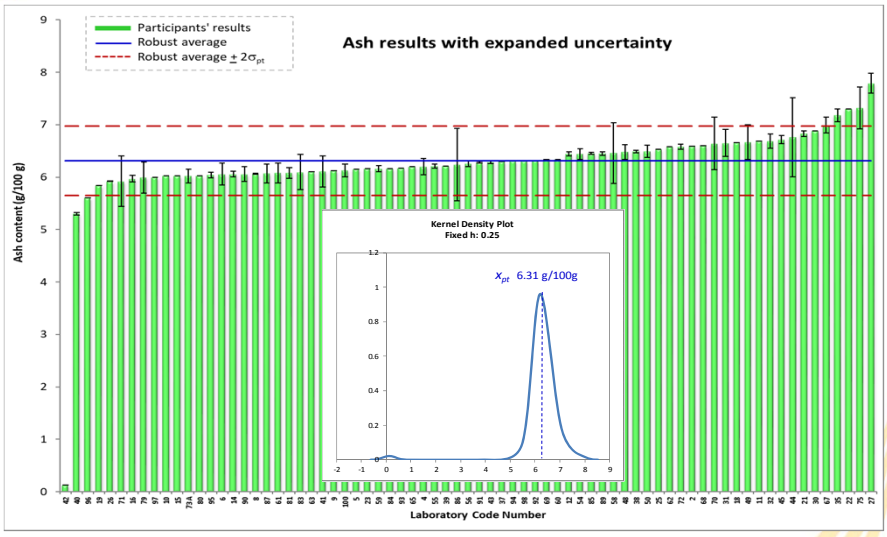

Figure 12: Distribution of ash results (ascending order) in defatted soybean flour with expanded uncertainty

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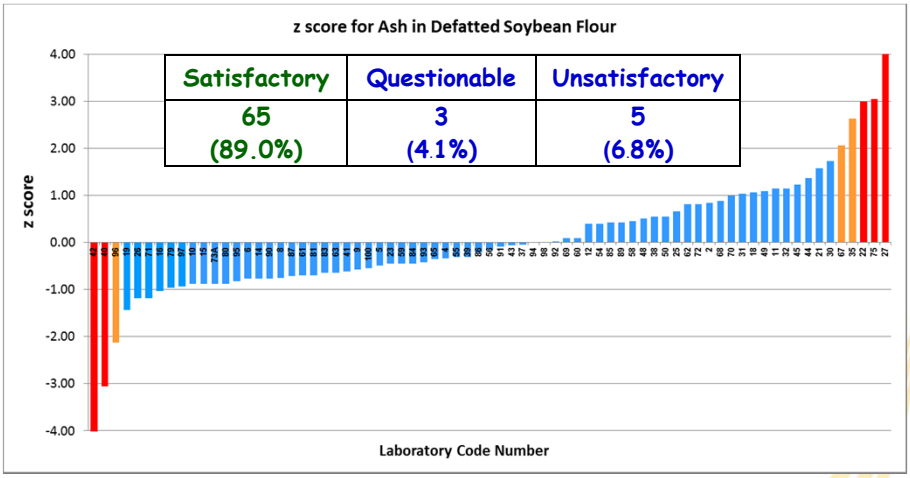
Figure 13: Laboratory performance on **ash analysis**



Unsatisfactory results ($|z| \geq 3$): Lab # 42, 40, 22, 75, 27

Questionable results ($2 < |z| < 3$): Lab # 96, 67, 35

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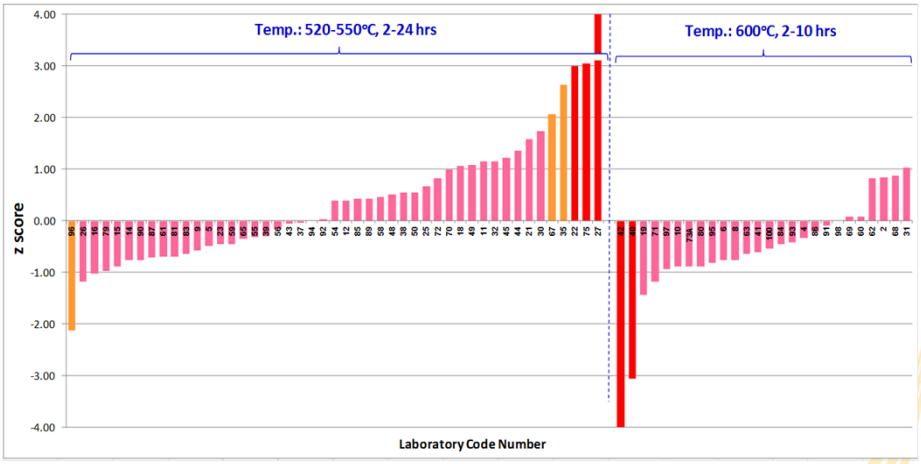

Figure 14. Plot of ordered z score for ash in defatted soybean flour, grouping by ashing temperatures

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Discussion: Ash determination: 73 Lab

- The majority of the participants (47 lab, 64%) used 520-550°C for 2 to 24 h for ashing.
  Others applied higher temperature of 600°C for 2-10 h
- About 56% of total participants (41 out of 73 lab) included *charring* the sample over a hotplate or a Bunsen burner before incineration in a muffle furnace.
- "Charring the sample over a hotplate, initially at low temperature to avoid losing ash with flame, then increase the temperature gradually until smoking ceases before incineration in a muffle furnace. If the sample is not completely white, moist ash with a few drops of water or diluted acid. Then evaporate on water bath and repeat heating in the muffle furnace for 30 – 60 min until constant weight is obtained.
  - This step is recommended as it could reduce the period for getting completely white ash under the high temperature in the muffle furnace.
  - It could prevent fluffing of ash during opening the furnace

Based on z-scores, most laboratories (65 out of 73, 89%), were identified as good performance on ash analysis. Two lab each reported extreme high values and extreme low values and three reported questionable values; they must review each step of ash determination and do corrective action.

The reference value of ash content in the defatted soybean flour, obtained from 65 good performance laboratories , is 6.32 ± 0.32 g/100 g (mean±SD, N=65, %CV=5.0)

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Results, Data distribution and Evaluation:

"Calcium"

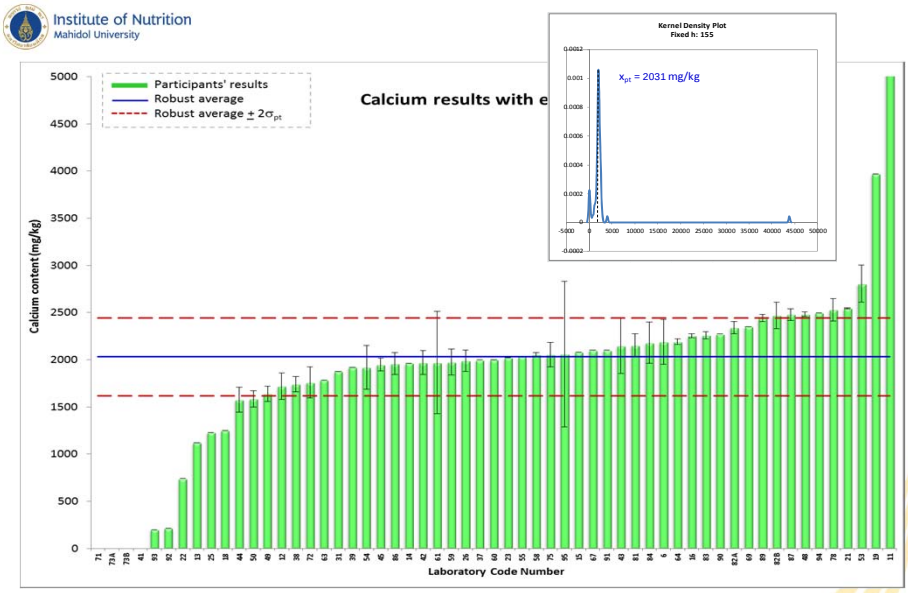

Figure 15: Distribution of calcium results (ascending order) in defatted soybean flour with expanded uncertainty

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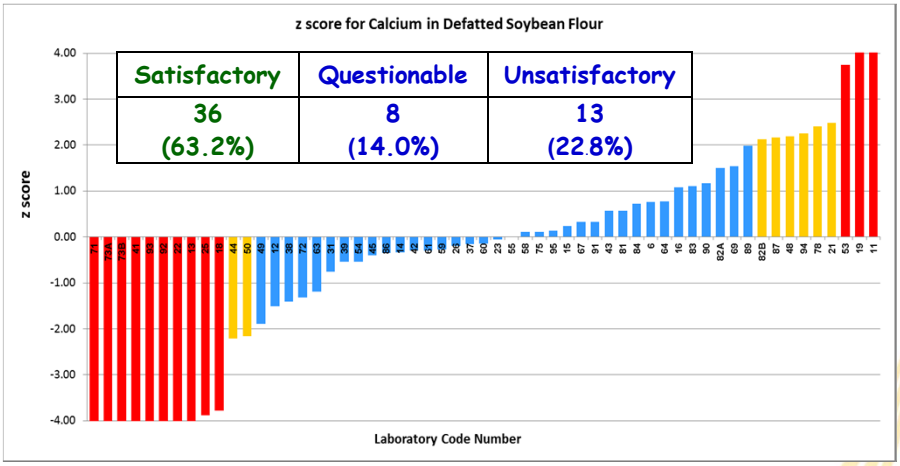
Figure 16: Laboratory performance on **calcium analysis**



Unsatisfactory results (| z | ≥ 3): Lab # 71, 73A, 73B, 41, 93, 92, 22, 13, 25, 18, 53, 19, 11

Questionable results (2 < | z | < 3): Lab # 44, 50, 82B, 87, 48, 94, 78, 21

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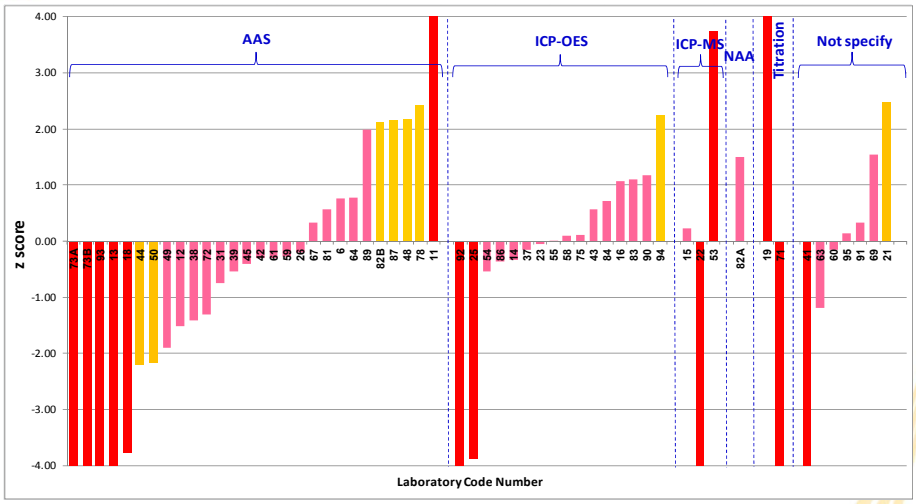

Figure 17. Plot of ordered z score for calcium in defatted soybean flour, categorised in groups according to analytical methods/parameters used

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Results, Data distribution and Evaluation:

"Magnesium"

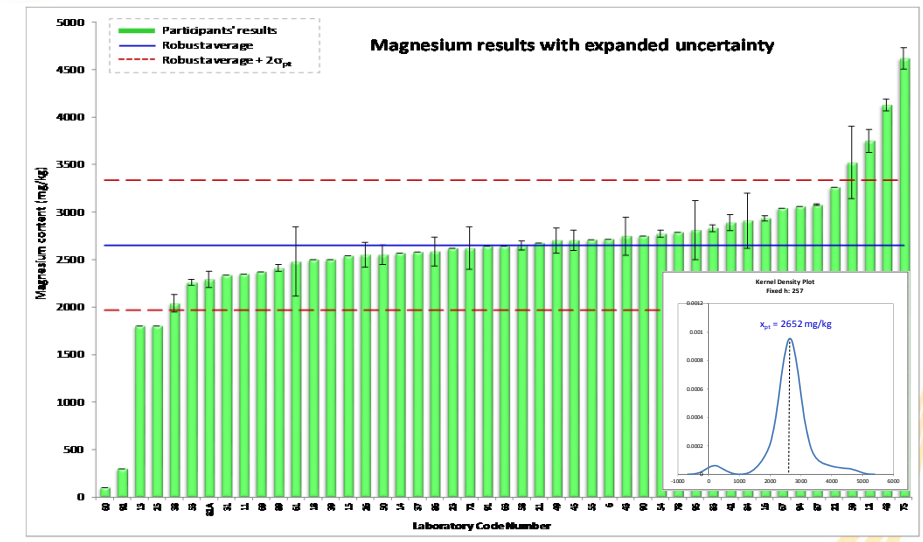
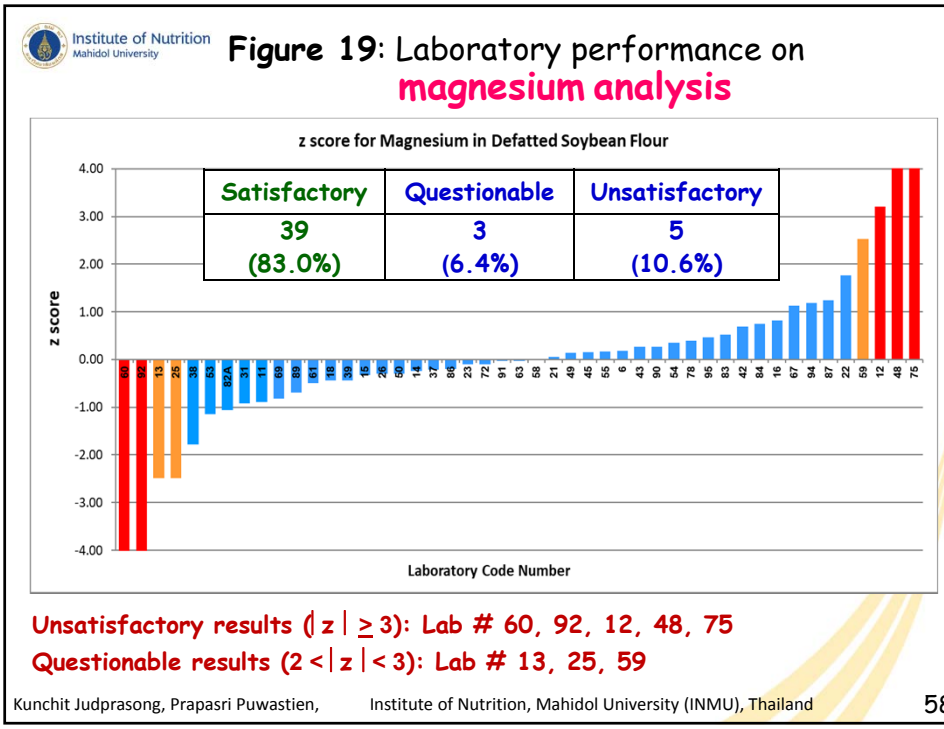


Figure 18: Distribution of magnesium results (ascending order) in defatted soybean flour with expanded uncertainty



Unsatisfactory results ($|z| \geq 3$): Lab # 60, 92, 12, 48, 75

Questionable results ($2 < |z| < 3$): Lab # 13, 25, 59

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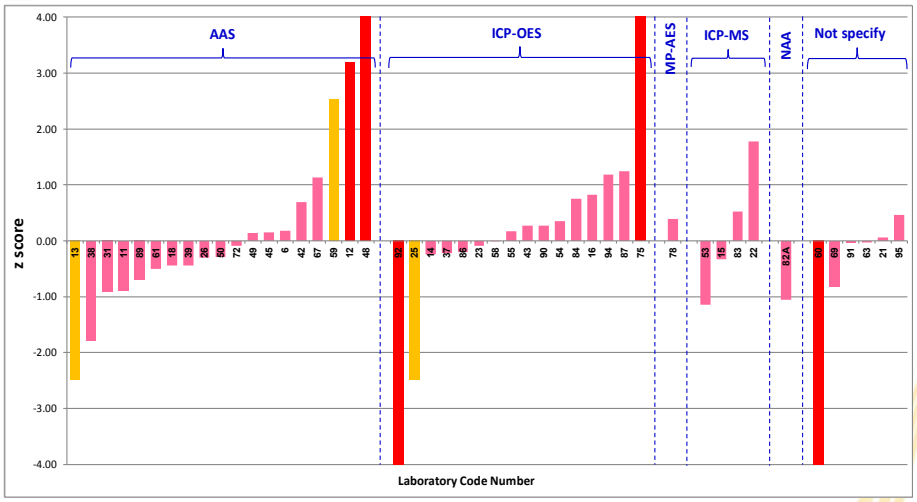

Figure 20. Plot of ordered z score for magnesium in defatted soybean flour, categorised in groups according to analytical methods/parameters used

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Results, Data distribution and Evaluation:

"Phosphorus"

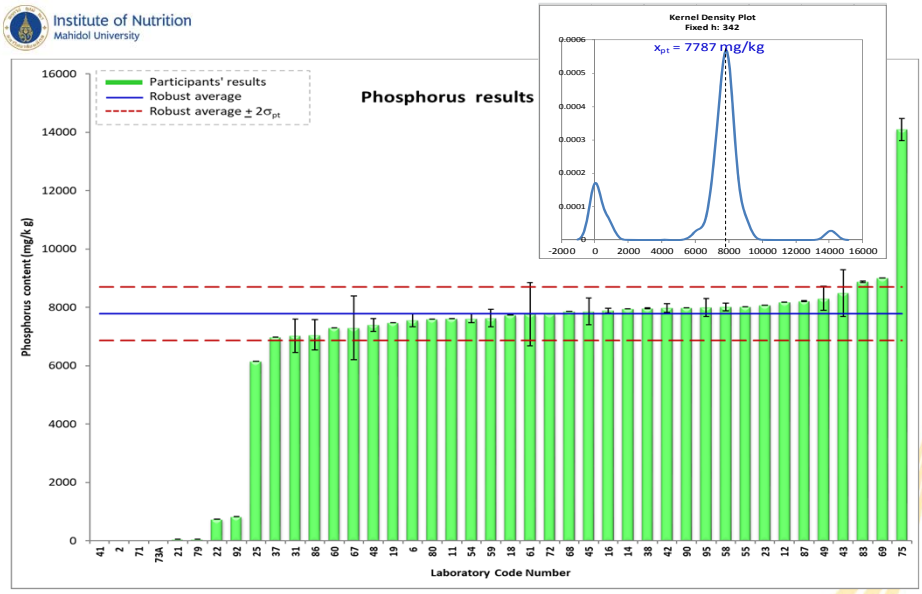

Figure 21: Distribution of phosphorus results (ascending order) in defatted soybean flour with expanded uncertainty

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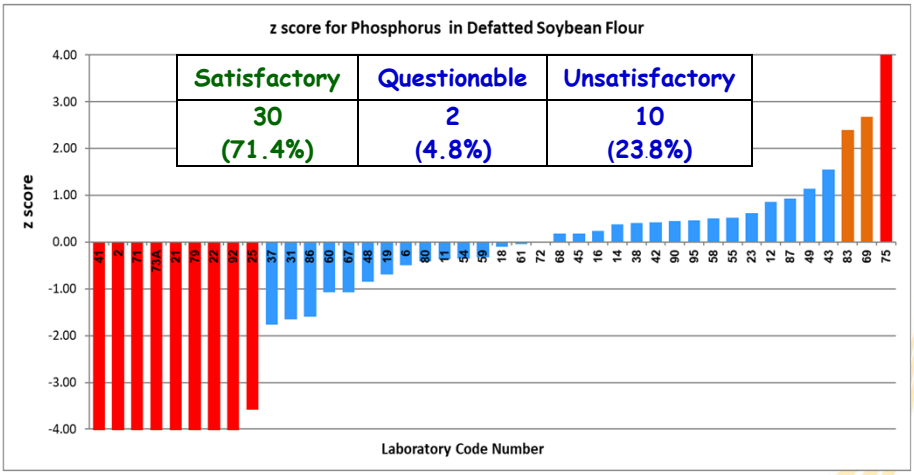
Figure 22: Laboratory performance on **phosphorus analysis**



Unsatisfactory results (| z | ≥ 3): Lab # 41, 2, 71, 73A, 21, 79, 22, 92, 25, 75

Questionable results (2 < | z | < 3): Lab # 83, 69

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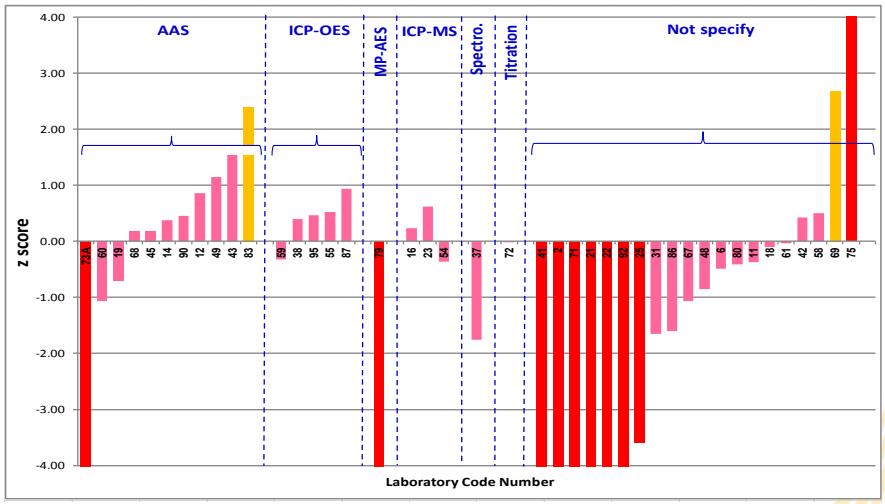

Figure 23. Plot of ordered z score for phosphorus in defatted soybean flour, categorised in groups according to analytical methods/parameters used

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Results, Data distribution and Evaluation:

"Sodium"

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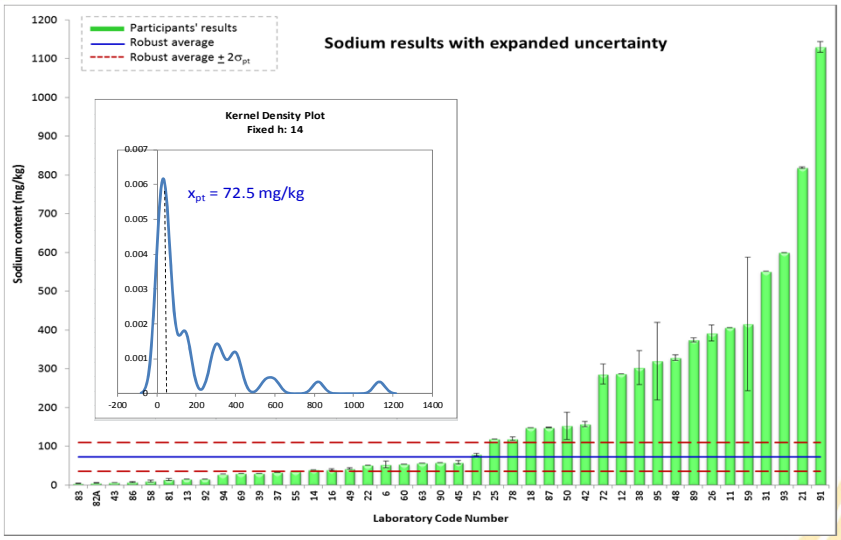

Figure 24: Distribution of sodium results (ascending order) in defatted soybean flour with expanded uncertainty

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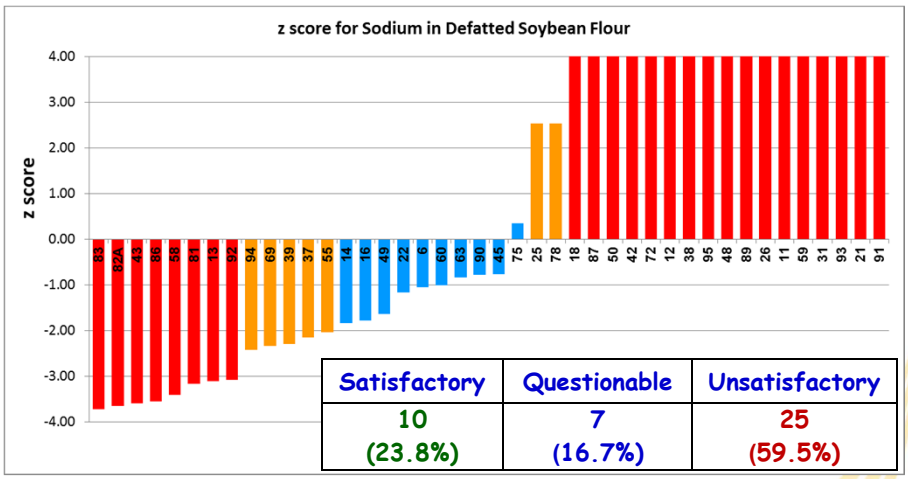
Figure 25: Laboratory performance on **sodium analysis**



Satisfactory	Questionable	Unsatisfactory
10 (23.8%)	7 (16.7%)	25 (59.5%)

Unsatisfactory results ($|z| \geq 3$): Lab # 49, 30, 25, 27, 75
Questionable results ($2 < |z| < 3$): Lab # 54, 11, 94, 50

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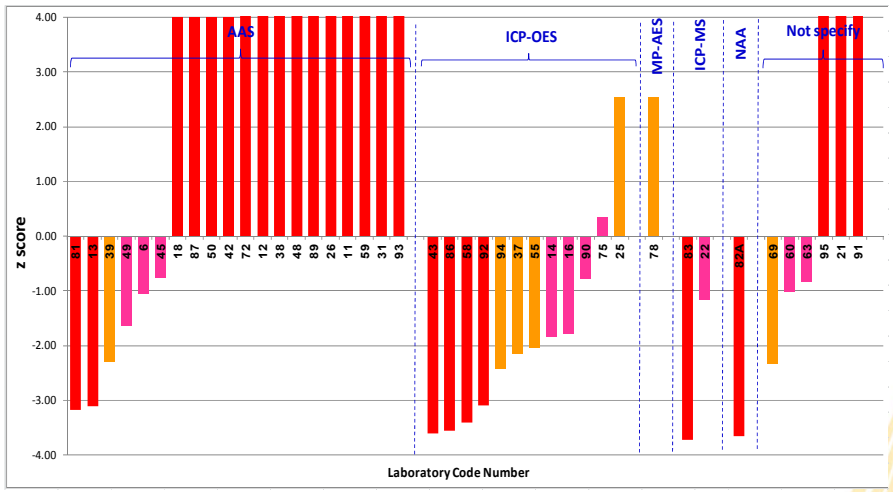

Figure 26. Plot of ordered z score for sodium in defatted soybean flour, categorised in groups according to analytical methods/parameters used

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Results, Data distribution and Evaluation:

"Potassium"

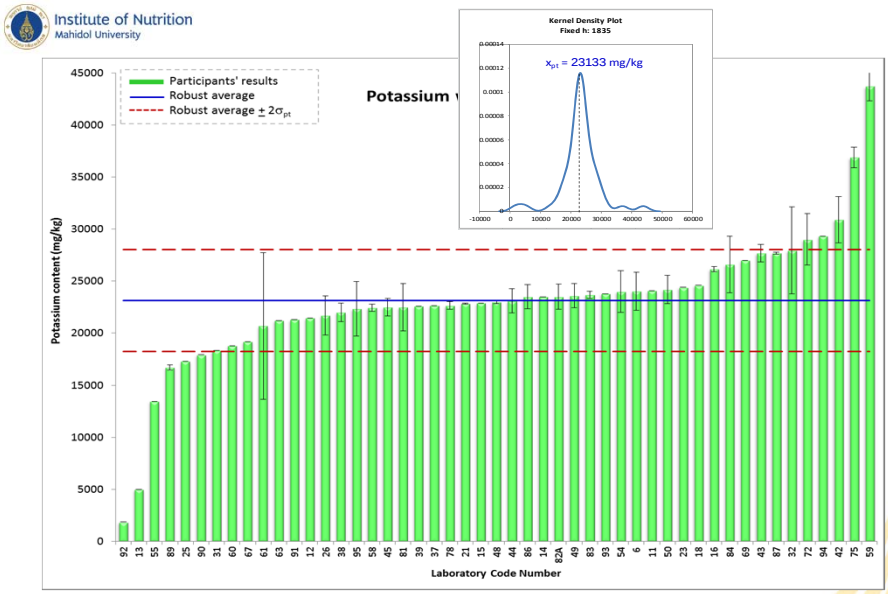

Figure 27: Distribution of potassium results (ascending order) in defatted soybean flour with expanded uncertainty

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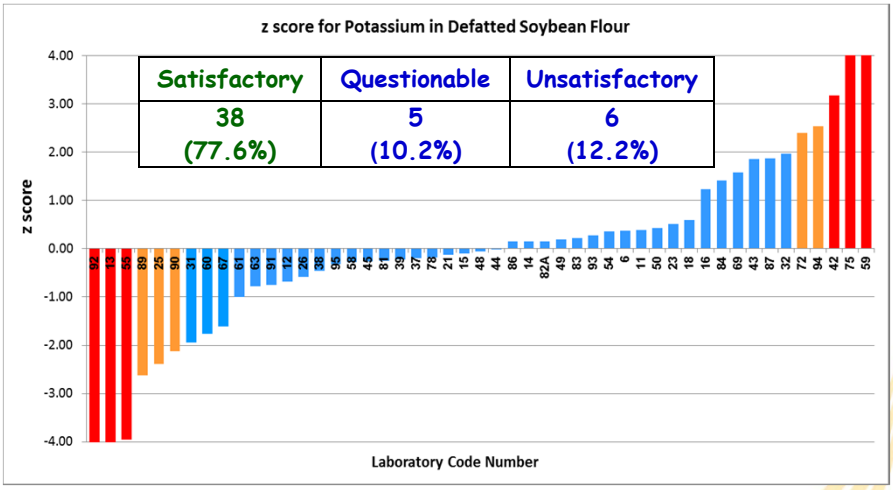
Figure 28: Laboratory performance on **potassium analysis**



Unsatisfactory results (| z | ≥ 3): Lab # 92, 13, 55,42, 75, 59

Questionable results (2 < | z | < 3): Lab # 89, 25, 90, 72, 94

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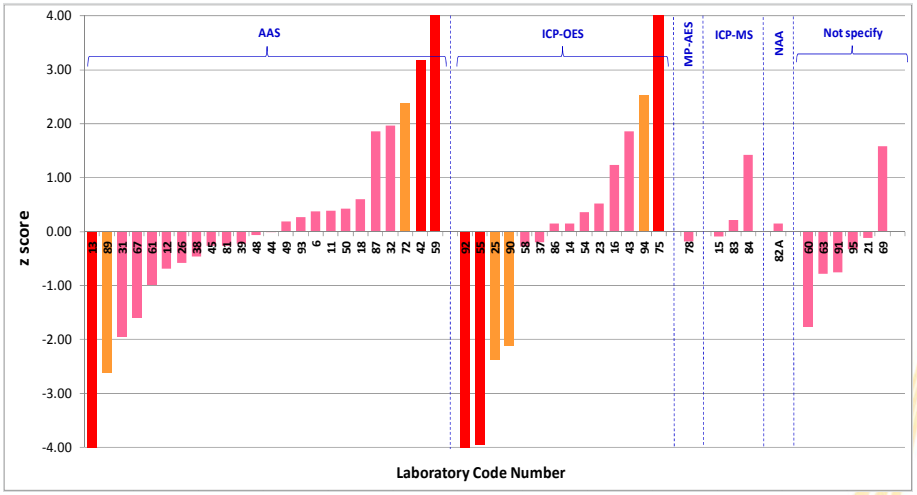

Figure 29. Plot of ordered z score for potassium in defatted soybean flour, categorised in groups according to analytical methods/parameters used

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Results, Data distribution and Evaluation:

"Iron"

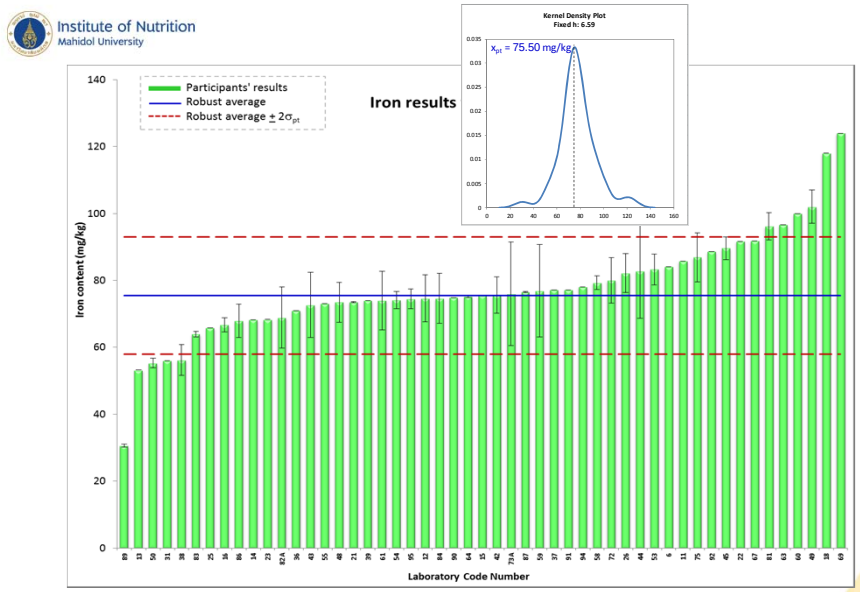

Figure 30: Distribution of iron results (ascending order) in defatted soybean flour with expanded uncertainty

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Table 7: Laboratory performance on **iron analysis**

Laboratory Number	Iron (mg/kg)	MU (mg/kg)	Based on NIMT[1]		Based on median ± NIQR[2]		Sample weight (g)	Digestion Technique	Digestion Medium	Instrument	Wavelength (nm or mass)	Recovery Correction (Y/N)	Method Reference
			z score	Zeta score	z score	Zeta score							
[1] Assigned value obtained from reference values (Isotope Dilution Mass Spectrometry, IDMS, as x_{pt} ± SD_p from Horwitz' s equation) = 75.1 ± 6.4 mg/kg (CV 8.5%); [2] Assigned value obtained from median (Med) ± normalised IQR (nIQR) = 75.50 ± 8.78 mg/kg (CV 11.6%, n= 51)													
Acceptance criteria =			|z score| ≤ 2.00	|ζ score| ≤ 2.00	|z score| ≤ 2.00	|ζ score| ≤ 2.00							
6	84.04	-	1.39	-	0.97	-	2.0000	Acid	HCl:HNO_3:H_2O	AAS	-	Y	AOAC (2016, 20th Ed, 928.08, 985.35 (50.1.14)
11	85.78	-	1.66	-	1.17	-	2.0000	Dry Ashing	HCl:H_2O	AAS	Fe 248.3	Y	AOAC (2016), 975.03, 985.35
12	74.60	7.00	-0.08	-0.12	-0.10	-0.24	0.5	Closed vessel	HNO_3	Fllame AAS	-	N	AOAC (2016), 985.35
13	53.20	-	-3.41	-	-2.54	-	0.5	Microwave	HNO_3 10 mL + HCl 2 mL	Analytikal Jena ContrAA 800 D	-	N	Internal Method
14	68.28	-	-1.06	-	-0.82	-	0.5	Ashing	50% HNO_3: 50% HCl	ICP Horiba Jobin Yvon	Fe 259.94	Y	AOAC 975.03, 984.27
15	75.50	-	0.06	-	0.00	-	0.5	Ultrawave Digestion	5% HNO_3 + 0.5% HCl	ICP-MS (7900 Agilent)	Fe 56	N	Based on USFDA 4.7 version 1.1
16	66.70	2.05	-1.31	-3.16	-1.00	-4.76	0.5	Hot plate	HNO_3+H_2O_2	ICP-OES Optima 7000 DV Perkin Elmer	Fe 238.204	N	In-house Method
18	118.00	-	6.69	-	4.84	-	2.0	Dry Ashing	HNO_3	AAS, Varian	Various	N	AOAC 968.08
21	73.50	0.19	-0.25	-0.65	-0.23	-1.30	0.1	Microwave	180°C	Mar Xpress (CEM)	-	Y	AOAC 2011.14 (2016)
22	91.70	-	2.59	-	1.85	-	0.2 to 0.3	Microwave	HNO_3	ICP-MS Perkin Elmer	-	-	AOAC 2015.06
23	68.30	-	-1.06	-	-0.82	-	1.00	Dry Ashing	-	ICP-OES	238	-	AOAC 985.01
25	65.80	0.07	-1.45	-3.80	-1.10	-6.30	5.0205 / 5.0206	Wet Digestion	HNO_3-HCl	ICP-OES	Fe 238.204	-	USEPA Method 3050B

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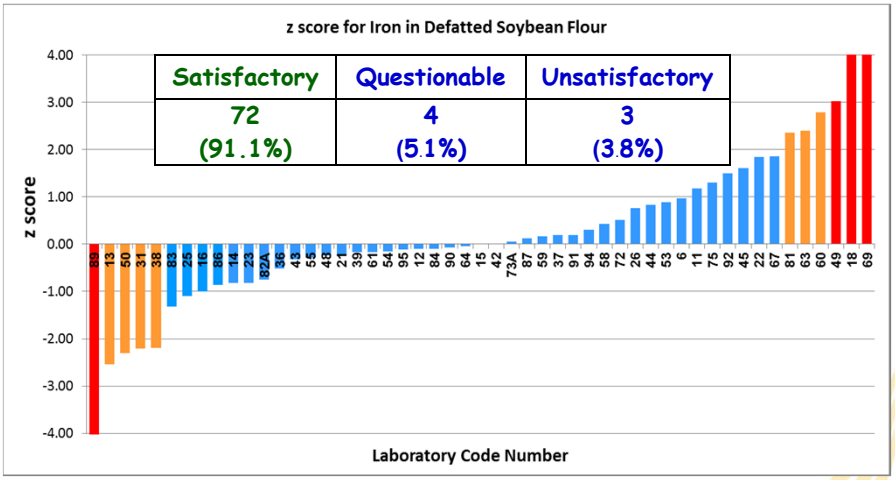
Figure 31: Laboratory performance on **iron analysis**



Unsatisfactory results (| z | ≥ 3): Lab # 49, 30, 25, 27, 75
Questionable results (2 < | z | < 3): Lab # 54, 11, 94, 50

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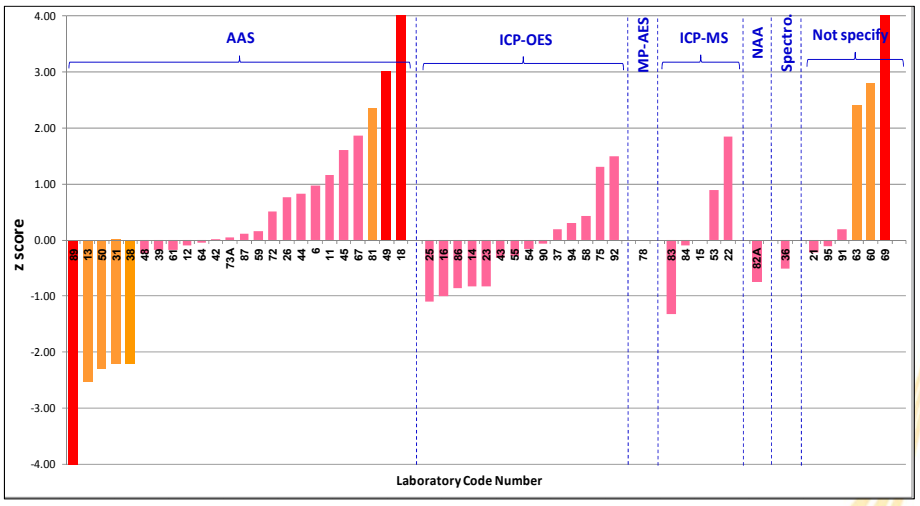

Figure 32. Plot of ordered z score for iron in defatted soybean flour, categorised in groups according to analytical methods/parameters used

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Results, Data distribution and Evaluation:

"Zinc"

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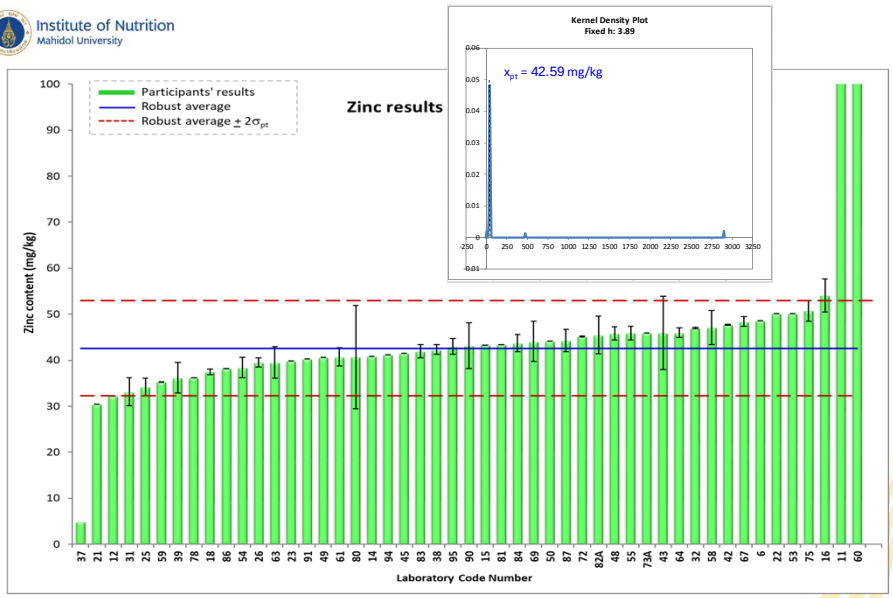

Figure 33: Distribution of zinc results (ascending order) in defatted soybean flour with expanded uncertainty

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Table 8: Laboratory performance on zinc analysis

Lab Number	Zinc (mg/kg)	MU (mg/kg)	Based on NIMT		Based on x* ± s*		Sample weight (g)	Digestion Technique	Digestion Medium	Instrument	Wavelength (nm or mass)	Recovery Correction (Y/N)	Method Reference
			z score	Zeta score	z score	Zeta score							
[1]Assigned value obtained from reference values (Isotope Dilution ICP-MS as x_{pt} ± SD_0 from Horwitz' s equation) = 43.1 ± 3.9 mg/kg (CV 9.0%) with u_{xpt} = 0.6 mg/kg; [2]Assigned value obtained from robust average (x*) ± robust SD (s*) = 42.59 ± 5.18 mg/kg (CV 12.2%, n= 46) with u_{xpt} = 0.95 mg/kg													
Acceptance criteria =			|z score| ≤ 2.00	|ζ score| ≤ 2.00	|z score| ≤ 2.00	|ζ score| ≤ 2.00							
6	**48.61**	-	1.42	-	1.16	-	2.0000	Acid	HCl:HNO_3:H_2O	AAS	-	Y	AOAC (2016, 20th Ed, 928.08, 985.35 (50.1.14)
11	**477.57**	-	**112.16**	-	**83.97**	-	2.0000	Dry Ashing	HCl:H_2O	AAS	-	Y	AOAC (2016), 975.03, 985.35
12	**32.20**	3.00	**-2.81**	**-7.20**	**-2.01**	**-5.85**	0.5	Closed vessel	HNO_3	Flame AAS	-	N	AOAC (2016), 985.35
14	**40.90**	-	-0.57	-	-0.33	-	0.5	Ashing	50% HNO_3, 50% HCl	ICP Horiba Jobin Yvon	Zn 213.856	Y	AOAC 975.03, 984.27
15	**43.30**	-	0.05	-	0.14	-	0.5	Ultrawave Digestion	5% HNO_3 + 0.5% HCl	ICP-MS (7900 Agilent)	-	N	Based on USFDA 4.7 version 1.1
16	**54.10**	5.41	**2.84**	**4.06**	**2.22**	**4.01**	0.5	Hot plate	HNO_3+H_2O_2	ICP-OES Optima 7000 DV Perkin Elmer	-	N	In-house Method
18	**37.50**	-	-1.45	-	-0.98	-	2.0	Dry Ashing	HNO_3	AAS, Varian	Various	N	AOAC 968.08
21	**30.50**	-	**-3.25**	-	**-2.33**	-	0.1	Microwave	180°C	Mar Xpress (CEM)	-	Y	AOAC 2011.14 (2016)
22	**50.10**	-	1.81	-	1.45	-	0.2 to 0.3	Microwave	HNO_3	ICP-MS Perkin Elmer	-	-	AOAC 2015.06
23	**39.80**	-	-0.85	-	-0.54	-	1.00	Dry Ashing	-	ICP-OES	589, 766, 422, 285, 238	-	AOAC 985.01
25	**34.20**	0.07	**-2.30**	**-43.87**	-1.62	**-8.83**	5.0205 / 5.0206	Wet Digestion	HNO_3-HCl	ICP-OES	-	-	USEPA Method 3050B
26	**39.50**	3.43	-0.93	**-2.08**	-0.60	-1.58	4.0	Dry ashing	Water & HCl (1+1)	AAS Shimadzu AA-7000	-	N	AOAC No. 975.03

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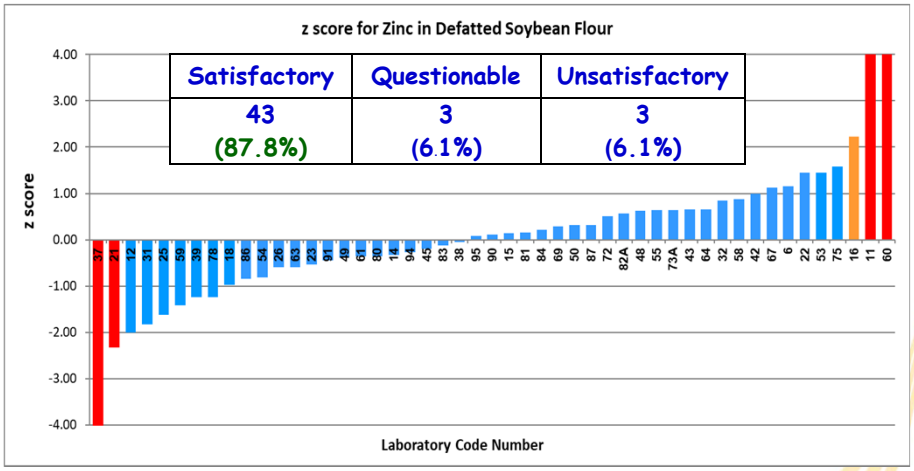
Figure 34: Laboratory performance on zinc analysis based on x* & s*



Unsatisfactory results (| z | ≥ 3): Lab # 37, 21, 11, 60

Questionable results (2 < | z | < 3): Lab # 12, 16

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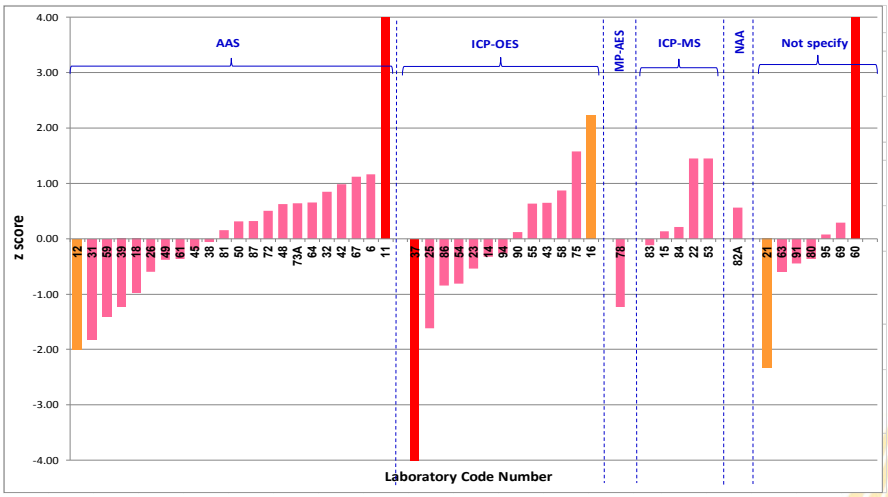

Figure 35. Plot of ordered z score for zinc in defatted soybean flour, categorised in groups according to analytical methods/parameters used

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Results, Data distribution and Evaluation:

"Copper"

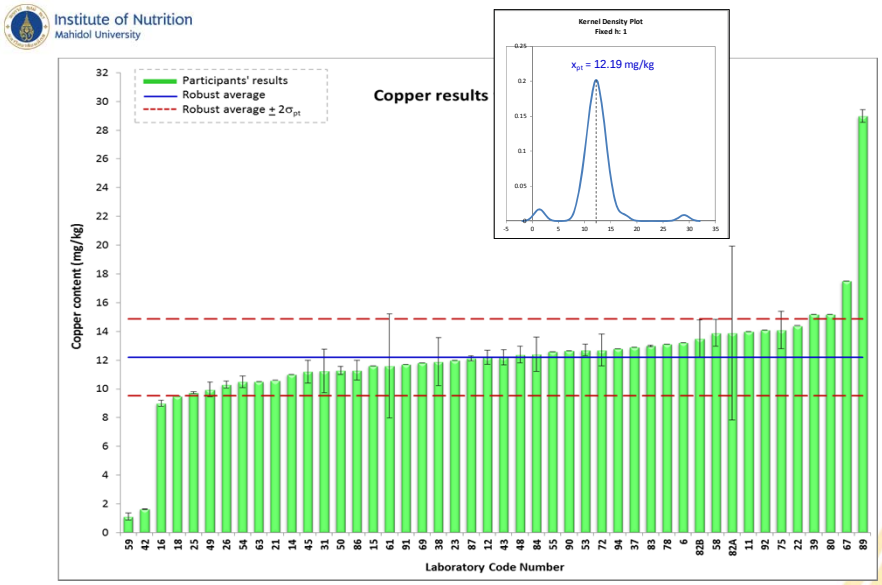

Figure 36: Distribution of copper results (ascending order) in defatted soybean flour with expanded uncertainty

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Table 9: Laboratory performance on copper analysis

Lab Number	Copper (mg/kg)	MU (mg/kg)	Based on NIMT		Based on x* ± SDp		Sample weight (g)	Digestion Technique	Digestion Medium	Instrument	Wave-length (nm or mass)	Recovery Correction (Y/N)	Method Reference
			z score	Zeta score	z score	Zeta score							
[1]Assigned value obtained from reference values (Isotope Dilution ICP-MS as x_{pt} ± SD_p from Horwitz' s equation) = 12.5 ± 1.3 mg/kg (CV 10.7%) with u_{xpt} = 0.2 mg/kg; [2]Assigned value obtained from robust average (x*) ± SDp from Horwitz' s equation = 12.19 ± 1.34 mg/kg (CV 11.0%, n= 46) with u_{xpt} = 0.25 mg/kg													
Acceptance criteria =			|z score| ≤ 2.00	|ζ score| ≤ 2.00	|z score| ≤ 2.00	|ζ score| ≤ 2.00							
6	13.21	-	0.53	-	0.76	-	2.0000	Acid	HCl:HNO3:H2O	AAS	-	Y	AOAC (2016, 20th Ed, 928.08, 985.35 (50.1.14)
11	13.98	-	1.11	-	1.34	-	2.0000	Dry Ashing	HCl:H2O	AAS	-	Y	AOAC (2016), 975.03, 985.35
12	12.20	0.50	-0.22	-0.94	0.01	0.03	0.5	Closed vessel	HNO3	Flame AAS	-	N	AOAC (2016),
14	11.01	-	-1.11	-	-0.88	-	0.5	Ashing	50% HNO3, 50% HCl	ICP Horiba Jobin Yvon	Cu 224.70	Y	AOAC 975.03, 984.27
15	11.60	-	-0.67	-	-0.44	-	0.5	Ultrawave Digestion	5% HNO3 + 0.5% HCl	ICP-MS (7900 Agilent)	-	N	Based on USFDA 4.7 version 1.1
16	9.01	0.21	-2.61	-15.45	-2.37	-11.73	0.5	Hot plate	HNO3+H2O2	ICP-OES Optima 7000 DV Perkin Elmer	-	N	In-house Method
18	9.49	-	-2.25	-	-2.01	-	2.0	Dry Ashing	HNO3	AAS, Varian	Various	N	AOAC 968.08

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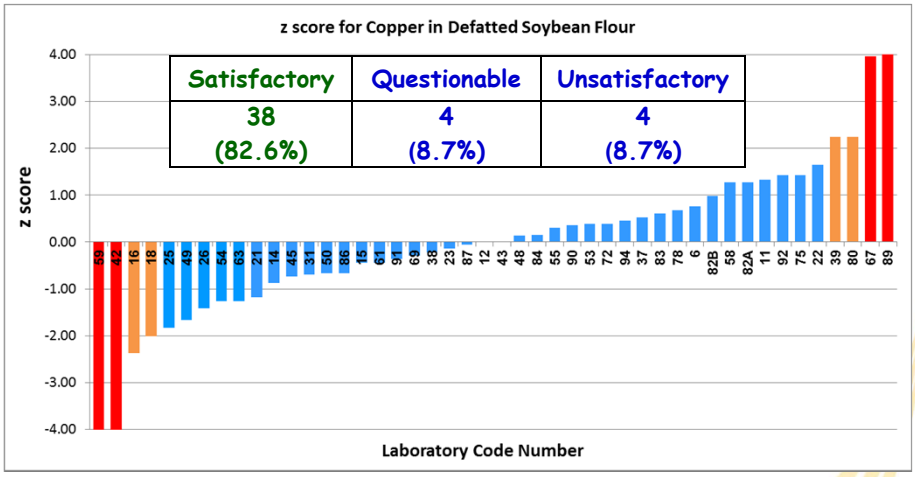
Figure 37: Laboratory performance on **copper analysis**



Unsatisfactory results ($|z| \geq 3$): Lab # 59, 42, 67, 89
Questionable results ($2 < |z| < 3$): Lab # 16, 16,39, 80

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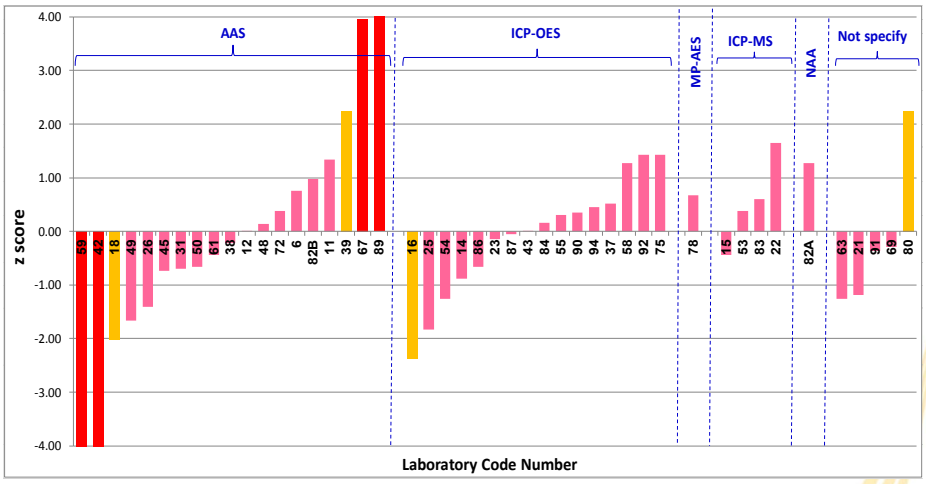

Figure 38. Plot of ordered z score for copper in defatted soybean flour, categorised in groups according to analytical methods/parameters used

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NOTES FOR MINERALS DETERMINATION:

- If ash will be used for mineral analysis, temperature of the furnace should not exceed 450°C.
- Too high a temperature may cause the volatilisation of certain elements particularly, Na, K, P, Fe, S, & Cl. It may also cause the mineral matter to fuse and melt.
- In this PT: the ashing temperture higher than 450°C, 520-550°C and 600°C, were used.
- More than 50% of the Lab prepared the sample for mineral analysis by dry ashing
- Laboratory performance on mineral analysis: effects of sample preparation, dry ashing VS wet digestion, were evaluated

laboratory performance on mineral analysis: effects of sample preparation (dry ashing vs wet digestion)

Lab performance on Mineral analysis	Sample preparation	
	Dry ashing	Wet digestion
CALCIUM (Ca)		
Total No. of submitted data (N=47)	28 (60%)	19
% Lab with Good performance	**57**	**63**
% Lab with Extreme high/low values	28	26
% Questionable high/low values	14	11
POTASSIUM (K)		
Total No. of submitted data (N=45)	25 (56%)	20
% Lab with Good performance	**76**	**75**
% Lab with Extreme high/low values	12	15
% Questionable high/low values	12	10
IRON (Fe)		
Total No. of submitted data (N=48)	26 (54%)	22
% Lab with Good performance	**65**	**82**
% Lab with Extreme high/low values	16	9
% Questionable high/low values	19	9

Data evaluation: a trial

For Calcium and Iron: sample prepared by wet digestion shows higher percentage of laboratory with good performance compared to those prepared by dry ashing.

However, no effect of the methods used for sample preparation on the laboratory performance of Patsssium determination.

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Table 10: Summary of laboratory performance on analyses of nutrients in defatted soybean flour

Parameters	Total participants	Evaluation results (number of laboratory, percentage in bracket)		
		Satisfactory	Questionable	Unsatisfactory
Moisture (g/100g)	79	72 (91.1%)	4 (5.1%)	3 (3.8%)
Total nitrogen (g/100g)	66	57 (86.4%)	5 (7.6%)	4 (6.1%)
Fat (g/100g)	40	Not evaluate due to high variation of results (0.0-4.0 g/100g)		
Ash (g/100g)	73	65 (89.0%)	3 (4.1%)	5 (6.8%)
Total dietary fibre (g/100g)	30	19 (63.3%)	2 (6.7%)	9 (30.0%)
Calcium (mg/kg)	57	36 (63.2%)	8 (14.0%)	13 (22.8%)
Magnesium (mg/kg)	47	39 (83.0%)	3 (6.4%)	5 (10.6%)
Phosphorus (mg/kg)	42	30 (71.4%)	2 (4.8%)	10 (23.8%)
Sodium (mg/kg)	42	10 (23.8%)	7 (16.7%)	25 (59.5%)
Potassium (mg/kg)	49	38 (77.6%)	5 (10.2%)	6 (12.2%)
Iron (mg/kg)	51	40 (78.4%)	7 (13.7%)	4 (7.8%)
Zinc (mg/kg)	49	43 (87.8%)	3 (6.1%)	3 (6.1%)
Copper (mg/kg)	46	38 (82.6%)	4 (8.7%)	4 (8.7%)

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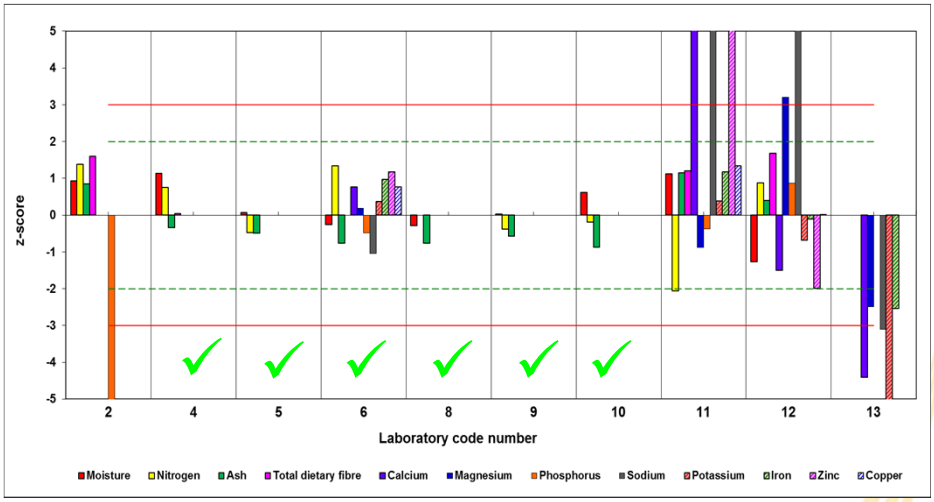

Figure 39: Bar-chart of z-scores for APFAN PT-2 in which the participants determined all nutrients using defatted soybean flour as test material: **laboratory code number 2-13.**

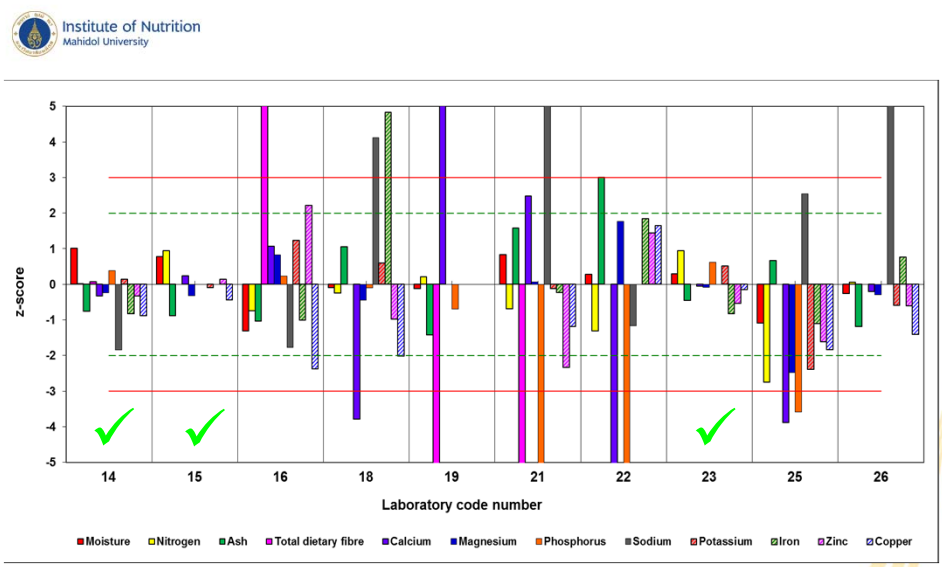

Figure 40: Bar-chart of z-scores for APFAN PT-2 in which the participants determined all nutrients using defatted soybean flour as test material: **laboratory code number 14-26**.

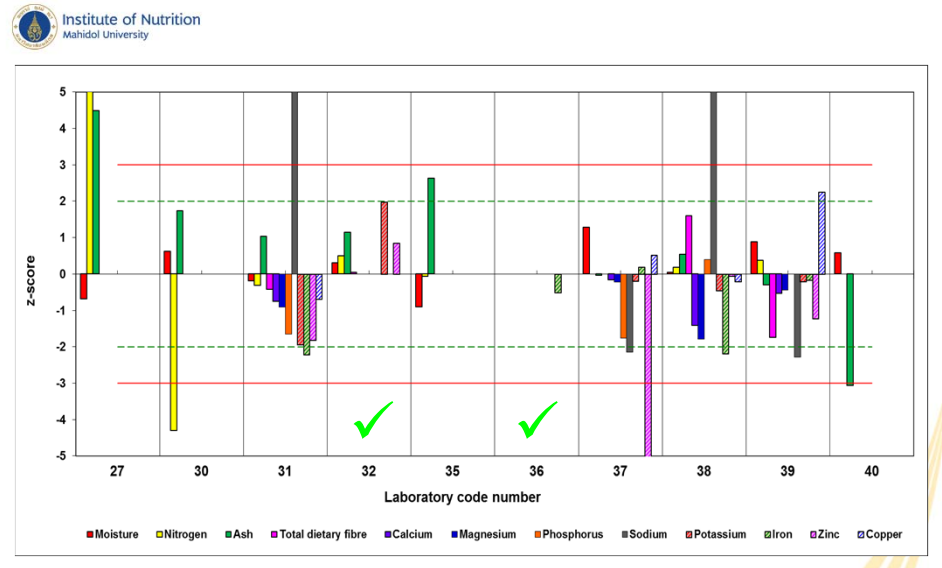

Figure 41: Bar-chart of z-scores for APFAN PT-2 in which the participants determined all nutrients using defatted soybean flour as test material: **laboratory code number 27-40**.

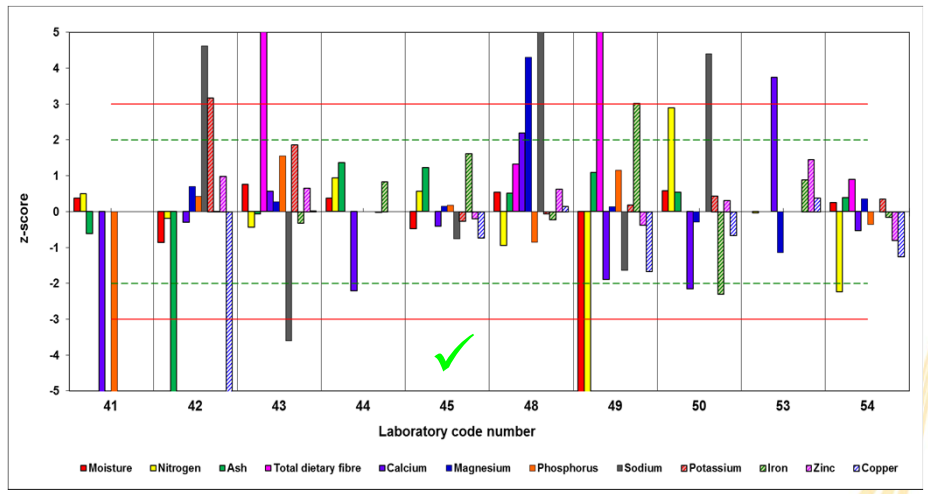

Figure 42: Bar-chart of z-scores for APFAN PT-2 in which the participants determined all nutrients using defatted soybean flour as test material: **laboratory code number 41-54**.

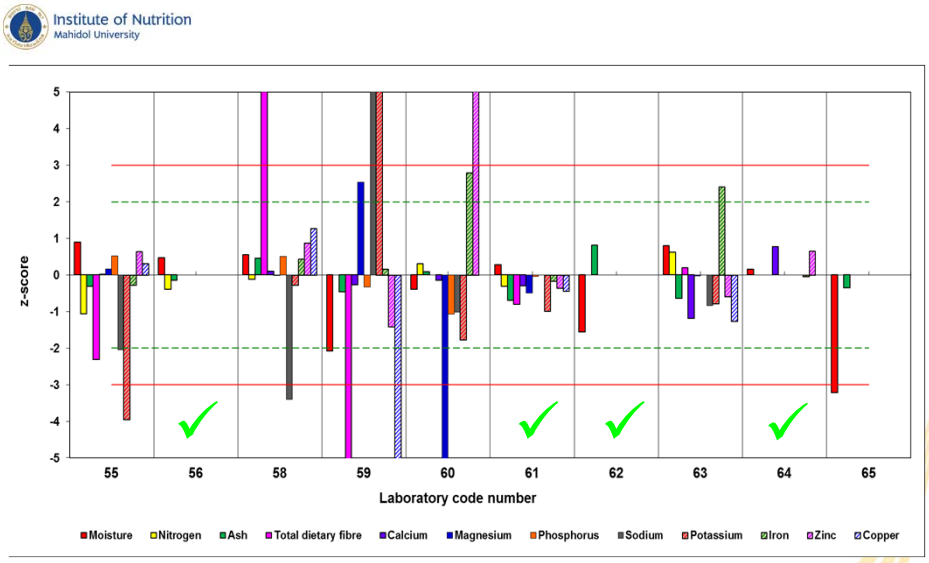

Figure 43: Bar-chart of z-scores for APFAN PT-2 in which the participants determined all nutrients using defatted soybean flour as test material: **laboratory code number 55-65**.

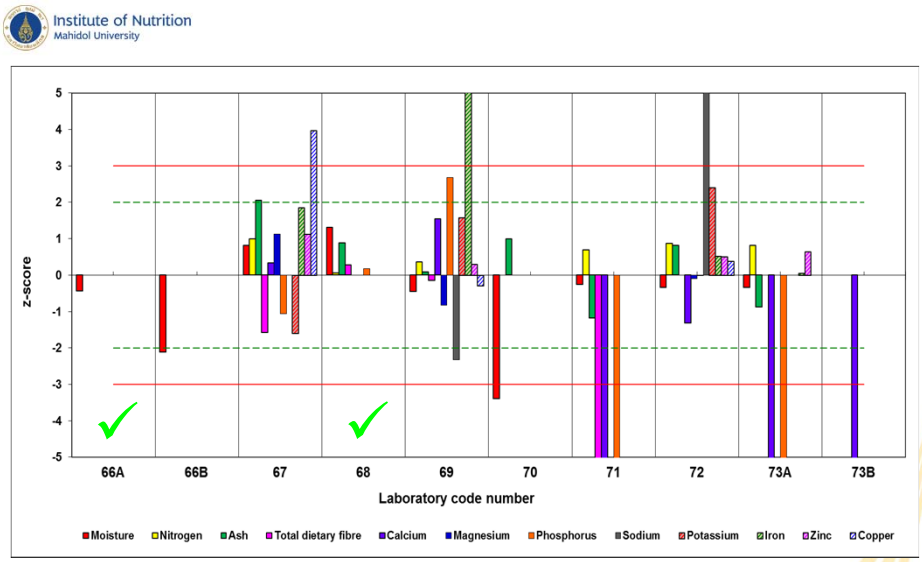

Figure 44: Bar-chart of z-scores for APFAN PT-2 in which the participants determined all nutrients using defatted soybean flour as test material: **laboratory code number 66A-73B**.

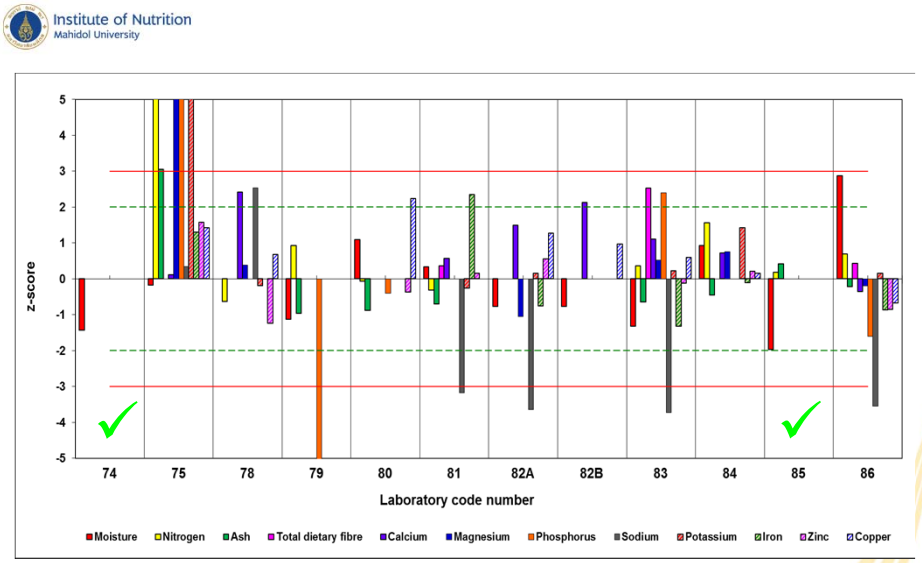

Figure 45: Bar-chart of z-scores for APFAN PT-2 in which the participants determined all nutrients using defatted soybean flour as test material: **laboratory code number 74-86**.

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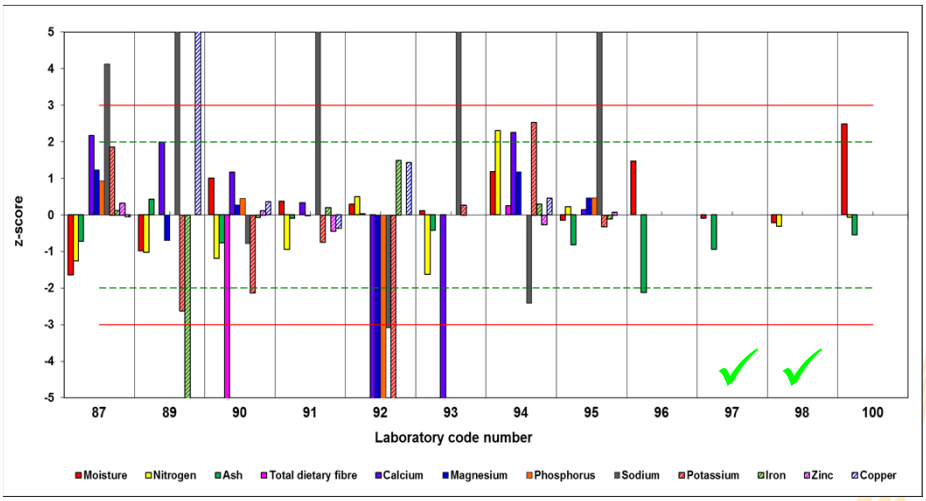

Figure 46: Bar-chart of z-scores for APFAN PT-2 in which the participants determined all nutrients using defatted soybean flour as test material: **laboratory code number 74-86**.

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Table 11. Summary of reference values of nutrients in defatted soybean flour

Parameters	Number of laboratories (n)	Reference values	
		Mean ± SD	%CV
Moisture (g/100g)	70	7.27 ± 0.57	7.8
Total nitrogen (g/100g)	56	7.88 ± 0.12	1.5
Ash (g/100g)	66	6.32 ± 0.32	5.0
Total dietary fibre (g/100g)	19	16.80 ± 1.26	7.5
Calcium (mg/kg)	36	2039 ± 184	9.0
Magnesium (mg/kg)	39	2652 ± 246	9.3
Phosphorus (mg/kg)	30	7758 ± 377	4.9
Sodium (mg/kg)	10	53.2 ± 11.7	21.9
Potassium (mg/kg)	38	23270 ± 2252	9.7
Iron (mg/kg)	40	76.71 ± 7.02	9.2
Zinc (mg/kg)	42	42.77 ± 4.42	10.3
Copper (mg/kg)	38	12.19 ± 1.23	10.1

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Conclusion: laboratory performance of APFAN-PT-2 participants

- ✓ Good performance for moisture, total nitrogen, ash, magnesium, zinc, and copper are achieved (|z score| ≤ 2, satisfactory results) by **>80% of all participants**.
- ✓ Satisfactory performance for analyses of total dietary fibre, calcium, phosphorus, potassium, and iron by participating laboratories are shown in **60-80% of all participants**.
- ✓ **Sodium analysis was classified as problematic nutrients**, only about 24% of all participants submitted values which were identified as satisfactory results.

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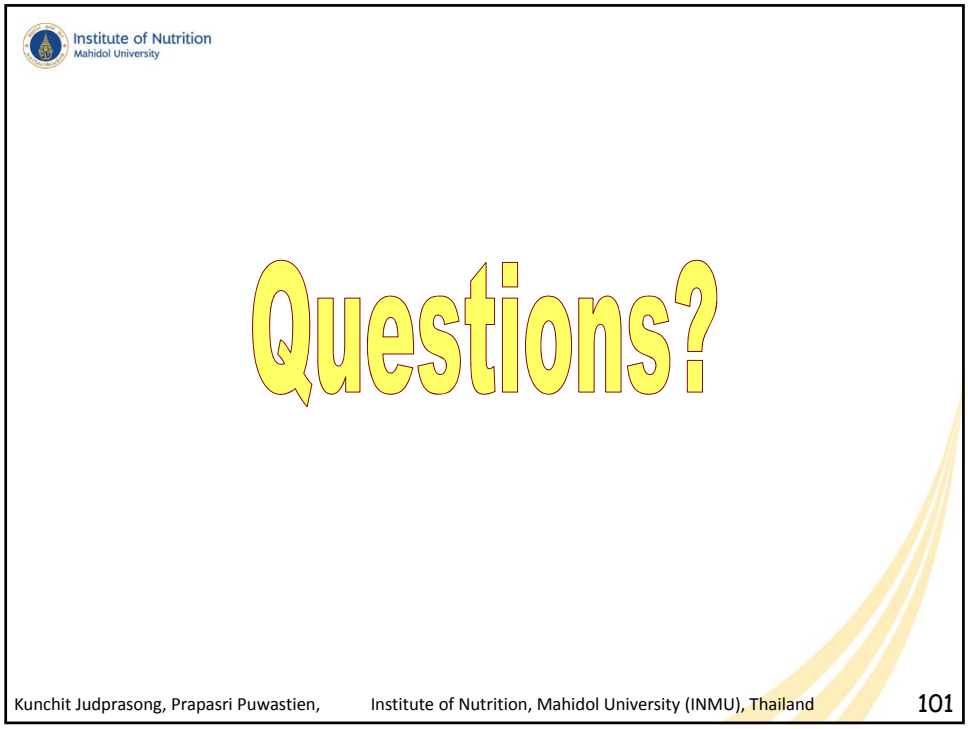
General comments: PT programme on nutrient analysis

- Test material for PT programme must be appropriately selected to support the target measurands.

  The national FCDB can be used as a guideline for selecting appropriate test material and target nutrients. Thai FCDB: https://inmu2.mahidol.ac.th/thaifcd/home.php
- Information on methods of nutrient analyses is one of the crucial information to interpret the results and to identify possible causes of extreme high or low values. The amount of sample used and all critical steps in each nutrient analysis should be included in the form for collection of methods used.
  PT participants should collaborate in answering the questionnaire.
- Unit of expression for nutrients in foods and in feeds which have been developed as the international guidelines should followed; as it is one of the common causes of mistake/error for unsatisfied performance
- The reported data (the figure, unit of expression, decimal places, etc.) must be carefully checked before submitting (share a case study)
- Technical Meeting after a PT programme at national level to discuss some issues, i.e., possible causes involving in discrepancies of submitted results, methods of analysis, critical control steps, etc., is recommended.
- After technical meeting, analysis of the problematic nutrients could be repeated
- Problematic nutrients should be included as target measurands in the next PT round.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Questions?
Kunchit Judprasong, Prapasri Puwastien, Institute of Nutrition, Mahidol University (INMU), Thailand
101