Australian Government
Department of Industry, Science, Energy and Resources

National Measurement Institute

Proficiency Test Final Report
AQA 21-20
Cocaine

April 2022

ACKNOWLEDGMENTS

This study was conducted by the National Measurement Institute (NMI). Support funding was provided by the Australian Government Department of Industry, Science, Energy and Resources.

I would like to thank the management and staff of the participating laboratories for supporting the study. It is only through widespread participation that we can provide an effective service to laboratories.

The assistance of the following NMI staff members in the planning, conduct and reporting of the study is acknowledged.

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Accredited for compliance with ISO/IEC 17043

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SUMMARY

AQA 21-20 Cocaine commenced in September 2021. Sample sets, each containing three samples of cocaine hydrochloride, were sent to twenty-six laboratories, with two laboratories requesting two sets of samples to be analysed by different analysts. All participants returned results.

Samples were prepared at the National Measurement Institute (NMI) laboratory in Sydney using seizures of cocaine hydrochloride, approximately 84% base (m/m) supplied by the Australian Federal Police.

The assigned values were the robust averages of participants' results.

Traceability: The consensus of participants' results is not traceable to any external reference, so although expressed in SI units, metrological traceability has not been established.

The outcomes of the study were assessed against the aims as follows:

- *Assess the proficiency of laboratories measuring cocaine in samples typical of a routine seizure.*

Laboratory performance was assessed by *z* score and E_n score.

Of 83 *z* scores, 75 (90%) returned $|z| \leq 2.0$, indicating a satisfactory performance.

Of 83 E_n scores, 74 (89%) returned $|E_n| \leq 1.0$, indicating agreement of the participant's result with the assigned value within their respective expanded uncertainties.

Laboratories **2**, **3**, **4**, **5**, **6**, **7**, **9**, **10**, **11**, **12**, **15**, **16**, **17**, **19**, **22**, **23**, **26**, **27** and **28** returned satisfactory *z* scores and E_n scores for all three samples.

- *Develop a practical application of traceability and measurement uncertainty, and provide participants with information that will assist uncertainty estimates.*

Of 83 numeric results, 80 (96%) were reported with an associated expanded measurement uncertainty. The magnitude of reported uncertainties was within 1.7% to 15% relative.

The metrological traceability of the assigned values has not been established as they were the consensus of participants' results.

- *Test the ability of participants to identify cutting agents commonly found in controlled drug preparation.*

Sample S1 was cut with phenacetin, Sample S2 was cut with phenacetin and quinine, and Sample S3 was cut with sucrose.

Twenty-six participants (93%) reported on the identity of the cutting agents of at least one sample.

Laboratories **1**, **18**, **24**, **25** and **27** correctly identified all cutting agents in all test samples.

- *Produce materials that can be used in method validation and as control samples.*

The test samples of this study are homogeneous and are well characterised. Samples are available for purchase from NMI and can be used for quality control and method validation purposes.

1 INTRODUCTION

1.1 NMI Proficiency Testing Program

The National Measurement Institute (NMI) is responsible for Australia's national measurement infrastructure, providing a range of services including a chemical proficiency testing program.

Proficiency testing (PT) is the 'evaluation of participant performance against pre-established criteria by means of interlaboratory comparison.'[1] NMI PT studies target chemical testing in areas of high public significance such as trade, environment, law enforcement and food safety. NMI offers studies in:

- pesticide residues in fruit and vegetables, soil and water;
- petroleum hydrocarbons in soil and water;
- per- and polyfluoroalkyl substances in water, soil, food and biota;
- inorganic analytes in soil, water, filters, food and pharmaceuticals;
- controlled drug assay, drugs in wipes and clandestine laboratory; and
- allergens in food.

1.2 Study Aims

The aims of the study were to:

- assess the proficiency of laboratories measuring cocaine in samples typical of a routine seizure;
- develop a practical application of traceability and measurement uncertainty, and provide participants with information that will assist uncertainty estimates;
- test the ability of participants to identify cutting agents commonly found in controlled drug preparation; and
- produce materials that can be used in method validation and as control samples.

The choice of test method was left to the participating laboratories.

1.3 Study Conduct

NMI is accredited by the National Association of Testing Authorities, Australia (NATA) to ISO/IEC 17043 as a provider of proficiency testing schemes.[1] This controlled drug PT study is within the scope of NMI's accreditation.

The conduct of NMI proficiency tests is described in the NMI Study Protocol for Proficiency Testing.[2] The statistical methods used are described in the NMI Chemical Proficiency Testing Statistical Manual.[3] These documents have been prepared with reference to ISO/IEC 17043 and The International Harmonized Protocol for Proficiency Testing of Analytical Chemistry Laboratories.[1,4]

2 STUDY INFORMATION

2.1 Study Timetable

The timetable of the study was:

Invitation issued	1 September 2021
Samples dispatched	23 November 2021
Results due	18 March 2022
Interim report issued	21 March 2022

Due to the international circumstances occurring over the course of this study, there were delivery delays to some participants, and so the project timeline has been significantly extended.

2.2 Participation and Laboratory Code

Twenty-six laboratories enrolled to participate in this study. Two laboratories requested two sets of test samples to be analysed by different analysts. Each participant was randomly assigned a confidential laboratory code for this study. All participants returned results.

2.3 Test Material Specification

Three test samples were prepared in October 2021. The starting material was cocaine hydrochloride samples approximately 84% base (m/m) supplied by the Australian Federal Police.

Phenacetin, quinine and sucrose purchased from Sigma Aldrich were used as cutting agents. Sample S1 was cut with phenacetin, Sample S2 was cut with phenacetin and quinine, and Sample S3 was cut with sucrose.

The cocaine was ground and sieved through a 180 µm sieve. The cutting agents were processed similarly. Test samples were then prepared by mixing a known mass of sieved drug material with a known mass of sieved cutting agent in a tumbler overnight. Portions of 150 mg of each of the test samples were then weighed out into labelled glass vials.

Sample S1 was prepared to contain approximately 77% cocaine base (m/m).

Sample S2 was prepared to contain approximately 32% cocaine base (m/m).

Sample S3 was prepared to contain approximately 47% cocaine base (m/m).

2.4 Test Sample Homogeneity

The preparation of homogeneous test samples is an important part of a PT study. Given the small (<150 mg) test portions normally used for controlled substances analysis, the particle size must be sufficiently small and uniformly distributed to ensure minimal influence on analytical precision.

No homogeneity testing was conducted in this PT study. Samples were prepared using the same procedure as previous controlled drug PT studies, which has been demonstrated to produce sufficiently homogeneous samples. Results returned by the participants gave no reason to question the homogeneity of the test samples.

2.5 Sample Dispatch

A set of three test samples, with each sample containing approximately 150 mg of test material, was dispatched to each participant in November 2021. The following items were also packaged with the samples:

- a covering letter with instructions for participants; and
- a form for participants to confirm the receipt of the test samples.

An Excel spreadsheet for the electronic reporting of results was emailed to participants.

2.6 Instructions to Participants

Participants were instructed as follows:

- Analyse each sample for amount of drug by your normal test method. It is recommended to thoroughly mix the content of each vial before taking a test portion for analysis.
- For each sample report % m/m cocaine as base. Report this figure as if reporting to a client.
- For each result report an estimate of the expanded uncertainty as % m/m cocaine as base.
- Report the identity of diluent(s)/adulterant(s) in all three samples if this is within your normal scope of analysis.
- Give brief details of your:
    - Basis of uncertainty estimate (e.g. uncertainty budget method, repeatability precision).
    - Analytical method (e.g. sample treatment, instrument type, calibration method).
    - Reference standard (e.g. source, purity)

  as requested by the results sheet.
- A result spreadsheet has been emailed to you. Please complete this spreadsheet and return by email to jenny.xu@measurement.gov.au.
- Results are to be returned by 1 February 2022.

Due to the international circumstances occurring over the course of this study, there were significant delivery delays to some international participants, and so the results due date was extended to 18 March 2022.

2.7 Interim Report

An interim report was emailed to all participants on 21 March 2022.

3 PARTICIPANT LABORATORY INFORMATION

3.1 Test Methods Reported by Participants

Participants were requested to provide information about their test methods. Responses are presented in Table 1. Some responses may be modified so that the participant cannot be identified.

Table 1 Summary of Participants' Test Methods

Lab. Code	Extraction Solvent	Internal Standard	Calib. Points	Technique	Detector	Column
1	acetonitrile/water (80/20)	none	3	HPLC	DAD	C8
2	Methanol	none	5	HPLC	DAD	Kinetex 2.6 µ XB-C18
3	Methanol	none (External standard)	3	GC	FID	CP sil5CB
4	d-chloroform	1,4-bis(trimethylsilyl) benzene	NA	QNMR	NA	500MHz
5	75:25 ACN:water	Diethylphthalate	3	UPLC	PDA	C18-BEH
6	Methanol		4	HPLC	DAD	Eclipse XDB-C18
7	Methanol	Diazepam	6	GC	FID	J&W DB-5ms (Agilent 128-5512)
8	ACN/MeOH/H2O	Analog cocaine	7	UPLC	MS/MS	C-18 Column
9	methanol	NO	7	HPLC	DAD	zorbax Eclipse XDB-C18 (5 microns, 4.6 mm x150 mm)
10	Methanol	Tetracosane	4	GC	FID	HP5 30m x 0.32 mm x 0.25 µm
11	Acetonitrile	Strychnine	6	GC	FID	HP1-MS
12	acetonitrile/water	none	1	HPLC	DAD	Kromasil
13	Ethanol	Tetracosane	6	GC	FID	HP5
14	Water/ACN	N/A	5	HPLC	UV	Kinetex 5u C18
15	Ethanol	TBA	6	GC	FID	DB5
16	Acetonitrile/water/ trifluoroacetic acid (25/75/0.1)	N/A	3	HPLC	DAD	ODS2 Interpak column
17	Methanol	N/A	6	HPLC	UV	Luna C-18, 0.5% DEA, pH 8.5 : CH3OH, 30:70
18	Ethanol	Propyl Paraben	7	UPLC	DAD	BEH Shield RP18
19	HPLC Methanol	Lidocaine	1	UPLC	DAD	Agilent LiChrospher 60 RP-select B
20	Methanol	N/A	6	UPLC	DAD	Acquity UPLC BEH C18 1.7 µm 2.1 x 100 mm

Lab. Code	Extraction Solvent	Internal Standard	Calib. Points	Technique	Detector	Column
21	Methanol	nil	6	UPLC	DAD	Acquity UPLC BEH C18 1.7 µm 2.1 x 100 mm
22	water/acetonitrile/ n10 sulfuric acid 90:10:1	None	3	HPLC	Diode Array	Shimpack XR-ODS
23	Ethanol	Tribenzylamine	6	GC	FID	HP5
24	ethanol	tribenzylamine	4	GC	FID	HP-1
25	S1 & S2: Chloroform S3: Mobile Phase	S1 & S2: Benzopinacolone S3: -	S1 & S2: 1 S3: 4	S1 & S2: GC S3: HPLC	S1 & S2: FID S3: PDA	S1 & S2: HP-1 S3: C18 ubondapak
26	Acetonitrile/ Methanol (95:5)	Pholcodine 1mg/ml	3	UPLC	PDA	ACQUITY C-18
27	Ethanol	Triphenylaceto-phenone (TPAP)	3	GC	FID	HP1-MS
28	HPLC Methanol	Lidocaine	1	UPLC	DAD	Agilent LiChrospher 60 RP-select B

3.2 Details of Participant Calibration Standard

Participants were requested to provide information about their calibration standard used. Responses are presented in Table 2. Some responses may be modified so that the participant cannot be identified.

Table 2 Participant Calibration Standard

Lab. Code	Reference Standard	Purity (%)
1	lipomed	99.503
2	Lipomed	>98.5
3	Duchefa	>99
4	NMI	99.7
5	NMI	99.8
6		
7	Lipomed	99.3
8	Unikem	100
9	Lipomed	99.9
10	Merck	100
11	NMI	99.8
12	Sigma	99.9
13	Alcaliber	100
14	Johnson Matthey	100.3
15	Lipomed	99.503% +/- 0.026% free base content 88.7%
16	Johnson Matthey (MacFarlan Smith)	100.3
17	NMI	99.8

Lab. Code	Reference Standard	Purity (%)
18	NMI	96
19	Lipomed	99.503 ± 0.026
20	Cerilliant	100 (1.000 mg/mL)
21	Cerilliant	100 (1.000 mg/mL)
22	Sigma Aldrich	99.7
23	Lipomed	99.503
24	Cocaine Fagron	100
25	Macfarlan Smith	100.2
26	NMI	99.8
27	NMI	99.8±0.8
28	Lipomed	99.503 ± 0.026

3.3 Reported Basis of Participants' Measurement Uncertainty Estimates

Participants were requested to provide information about their basis of measurement uncertainty (MU). Responses are presented in Table 3. Some responses may be modified so that the participant cannot be identified.

Table 3 Reported Basis of Uncertainty Estimate

Lab. Code	Approach to Estimating MU	Information Sources for MU Estimation*		Guide Document for Estimating MU
		Precision	Method Bias	
1	Top Down - precision and estimates of the method and laboratory bias	Control samples - RM	Laboratory bias from PT studies	NF V03-110
2	Top Down - precision and estimates of the method and laboratory bias	Control samples - CRM Duplicate analysis	Instrument calibration Homogeneity of sample Masses and volumes Recoveries of SS	Eurachem/CITAC Guide
3	Top Down - precision and estimates of the method and laboratory bias	Control samples - SS Duplicate analysis	Instrument calibration Recoveries of SS	ISO/GUM
4	Top Down - precision and estimates of the method and laboratory bias	Control samples - previously analysed real seizure samples Duplicate analysis	Instrument calibration Matrix effects	Eurachem/CITAC Guide
5	Top Down - reproducibility (standard deviation) from PT studies used directly	Control samples - RM Duplicate analysis	Homogeneity of sample Standard purity	Eurachem/CITAC Guide
6	Top Down - precision and estimates of the method and laboratory bias	Duplicate analysis	Instrument calibration Masses and volumes Recoveries of SS Standard purity	
7	Estimating Measurement Uncertainty by black box with pairs of values	Standard deviation from PT studies only.		ISO/GUM ISO 21748

Lab. Code	Approach to Estimating MU	Information Sources for MU Estimation*		Guide Document for Estimating MU
		Precision	Method Bias	
8	Top Down - precision and estimates of the method and laboratory bias	Control samples - RM		
9	Top Down - precision and estimates of the method and laboratory bias	Control samples - CRM Duplicate analysis	Instrument calibration Homogeneity of sample Masses and volumes Laboratory bias from PT studies Standard purity	Eurachem/CITAC Guide
10	Top Down - precision and estimates of the method and laboratory bias	Control samples - Authentic powders	Instrument calibration Matrix effects Laboratory bias from PT studies Recoveries of SS Standard purity	
11	Bottom Up (ISO/GUM, fish bone/cause and effect diagram)	Control samples - CRM Duplicate analysis	Instrument calibration Masses and volumes Matrix effects	ISO/GUM
12	Standard deviation of replicate analyses multiplied by 2 or 3	Control samples - RM		ISO/GUM
13	Top Down - precision and estimates of the method and laboratory bias	Control samples – In-house RM Duplicate analysis	Matrix effects Standard purity	ISO/GUM
14	Top Down - precision and estimates of the method and laboratory bias	Duplicate analysis	Instrument calibration Homogeneity of sample Masses and volumes Matrix effects Standard purity	Eurachem/CITAC Guide
15	Top Down - precision and estimates of the method and laboratory bias	Standard deviation from PT studies only		
		Control samples - CRM Duplicate analysis	Instrument calibration Homogeneity of sample Masses and volumes Matrix effects Laboratory bias from PT studies Standard purity	
16	Standard deviation of replicate analyses multiplied by 2 or 3	Control samples - SS	Instrument calibration Masses and volumes Recoveries of SS	ISO/GUM
17	Top Down - precision and estimates of the method and laboratory bias	Control samples - RM Duplicate analysis		Eurachem/CITAC Guide
18				
19	Top Down - precision and estimates of the method and laboratory bias	Control samples - SS Duplicate analysis	Laboratory bias from PT studies Recoveries of SS	Eurachem/CITAC Guide

Lab. Code	Approach to Estimating MU	Information Sources for MU Estimation*		Guide Document for Estimating MU
		Precision	Method Bias	
20	Top Down - precision and estimates of the method and laboratory bias	Control samples Duplicate analysis	Instrument calibration Homogeneity of sample Masses and volumes Standard purity	NATA GAG Estimating and Reporting Measurement Uncertainty of Chemical Test Results
21	Top Down - precision and estimates of the method and laboratory bias	Control samples Duplicate analysis	Instrument calibration Homogeneity of sample Masses and volumes Standard purity	NATA GAG Estimating and Reporting Measurement Uncertainty of Chemical Test Results
22	Standard deviation of replicate analyses multiplied by 2 or 3	Control samples - CRM Duplicate analysis	Standard purity	ISO/GUM
23	Top Down - precision and estimates of the method and laboratory bias	Control samples - RM	Standard purity	
24	Top Down - precision and estimates of the method and laboratory bias	Control samples - RM Duplicate analysis		Internal document based on Eurachem/CITAC Guide; ISO:GUM
25				
26	Top Down - precision and estimates of the method and laboratory bias	Control samples - CRM Duplicate analysis	Instrument calibration Homogeneity of sample Masses and volumes	NATA GAG Estimating and Reporting Measurement Uncertainty of Chemical Test Results
27	Bottom Up (ISO/GUM, fish bone/cause and effect diagram)	Control samples - CRM Duplicate analysis	Instrument calibration Homogeneity of sample Masses and volumes Matrix effects Recoveries of SS Standard purity	Eurachem/CITAC Guide
28	Top Down - precision and estimates of the method and laboratory bias	Control samples - SS Duplicate analysis	Laboratory bias from PT studies Recoveries of SS	Eurachem/CITAC Guide

* CRM = Certified Reference Material, RM = Reference Material, SS = Spiked Samples

3.4 Participant Comments

The study coordinator welcomes comments or suggestions from participants as it can provide information which will improve future studies. Comments received in this study are presented in Table 4. Some responses may be modified so that the participant cannot be identified.

Table 4 Participant Comments

Lab. Code	Participant Comments
1	sample 2 contained quinine; quinine prevented us from doing the cocaine assay because this compound was coeluted with cocaine at the chosen wavelengths.
4	Methodology: No reference standard involved
9	Qualitative analysis was carried out by GC-MS
12	Methodology: 1 point, 5 injections
16	Uncertainty: The reported result (in routine case samples) is defined as the average of the individual results multiplied by the uncertainty correction factor and is rounded down to the nearest whole number (unless <1%w/w). The uncertainty correction factor is defined as (mean-2SD)/Mean expressed as a percentage using the relative standard control chart. E.g. a result of 53.8% would give a reported result of 53.8 * 0.9709 = 52.23 therefore rounded down to 52%
22	Uncertainty: UoM determined from 3 x std deviation of multiple injections expanded by professional judgement.
25	Methodology: Different methods used due to interfering substance - phenacetin
27	Methodology: A small amount of dichloromethane was used to dissolve the TPAP prior to the addition of Ethanol

4 PRESENTATION OF RESULTS AND STATISTICAL ANALYSIS

4.1 Results Summary

Participant results are listed in Tables 5 to 7 with the summary statistics: robust average, median, mean, number of numeric results (N), maximum (Max.), minimum (Min.), robust standard deviation (Robust SD) and robust coefficient of variation (Robust CV).

Bar charts of results and performance scores are presented in Figures 2 to 4. An example chart with interpretation guide is shown in Figure 1.

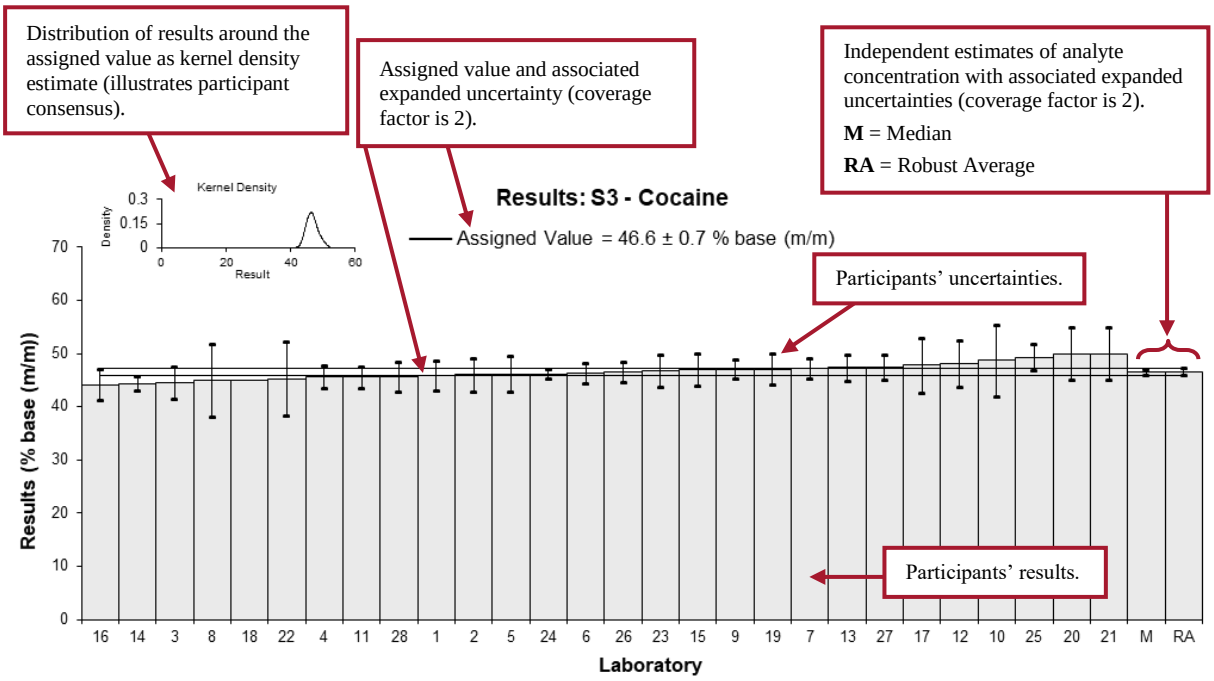


Figure 1 Guide to Presentation of Results

4.2 Assigned Value

The assigned value is defined as the 'value attributed to a particular property of a proficiency test item'.[1] In this study, the property is the % cocaine base (m/m) in the test samples. The assigned values were the robust averages of participants' results and the expanded uncertainties were estimated from the associated robust SDs (Appendix 1).

4.3 Robust Average and Robust Between-Laboratory Coefficient of Variation

The robust averages and associated expanded MUs, and robust CVs (a measure of the variability of participants' results) were calculated using the procedure described in ISO 13528:2015.[5]

4.4 Performance Coefficient of Variation (PCV)

The performance coefficient of variation (PCV) is a measure of the between-laboratory variation that in the judgement of the study coordinator would be expected from participants, given the levels of analytes present. The PCV is set by the study coordinator, and it is not the CV of participants' results. The PCV is based on the mass fraction of the analytes and experience from previous studies, and is also supported by mathematical models such as the Thompson-Horwitz equation.[6] By setting a fixed and realistic value for the PCV, a participant's performance does not depend on other participants' performances, and can be compared from study to study.

4.5 Target Standard Deviation

The target standard deviation (σ) is the product of the assigned value (X) and the PCV, as presented in Equation 1.

$$\sigma = X \times \text{PCV} \qquad \textit{Equation 1}$$

4.6 *z* Score

For each participant result, a *z* score is calculated according to Equation 2.

$$z = \frac{(\chi - X)}{\sigma} \qquad \textit{Equation 2}$$

where:

z is *z* score

χ is a participant's result

X is the assigned value

σ is the target standard deviation from Equation 1

For the absolute value of a *z* score:

- $|z| \leq 2.0$ is satisfactory;
- $2.0 < |z| < 3.0$ is questionable; and
- $|z| \geq 3.0$ is unsatisfactory.

4.7 E_n Score

The E_n score is complementary to the *z* score in assessment of laboratory performance. The E_n score includes uncertainty and is calculated according to Equation 3.

$$E_n = \frac{(\chi - X)}{\sqrt{U_\chi^2 + U_X^2}} \qquad \textit{Equation 3}$$

where:

E_n is E_n score

χ is a participant's result

X is the assigned value

U_χ is the expanded uncertainty of the participant's result

U_X is the expanded uncertainty of the assigned value

For the absolute value of an E_n score:

- $|E_n| \leq 1.0$ is satisfactory; and
- $|E_n| > 1.0$ is unsatisfactory.

4.8 Traceability and Measurement Uncertainty

Laboratories accredited to ISO/IEC 17025 must establish and demonstrate the traceability and measurement uncertainty associated with their test results.[7]

Guidelines for quantifying uncertainty in analytical measurement are described in the Eurachem/CITAC Guide.[8]

5 TABLES AND FIGURES

Table 5

Sample Details

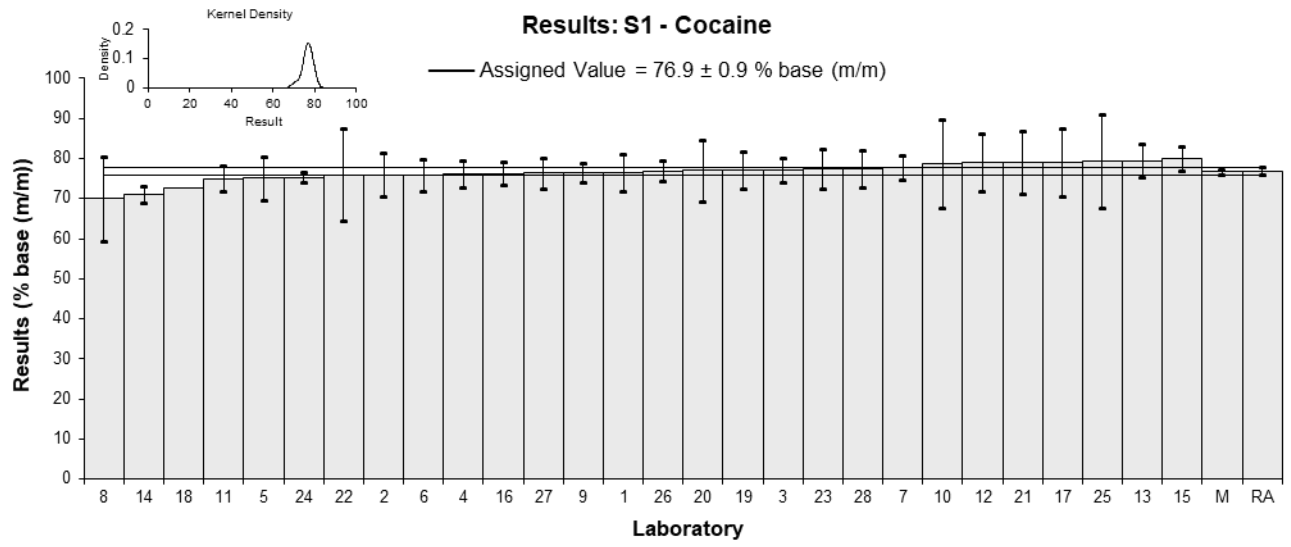
Sample	S1
Analyte	Cocaine
Matrix	Powder
Unit	% base (m/m)

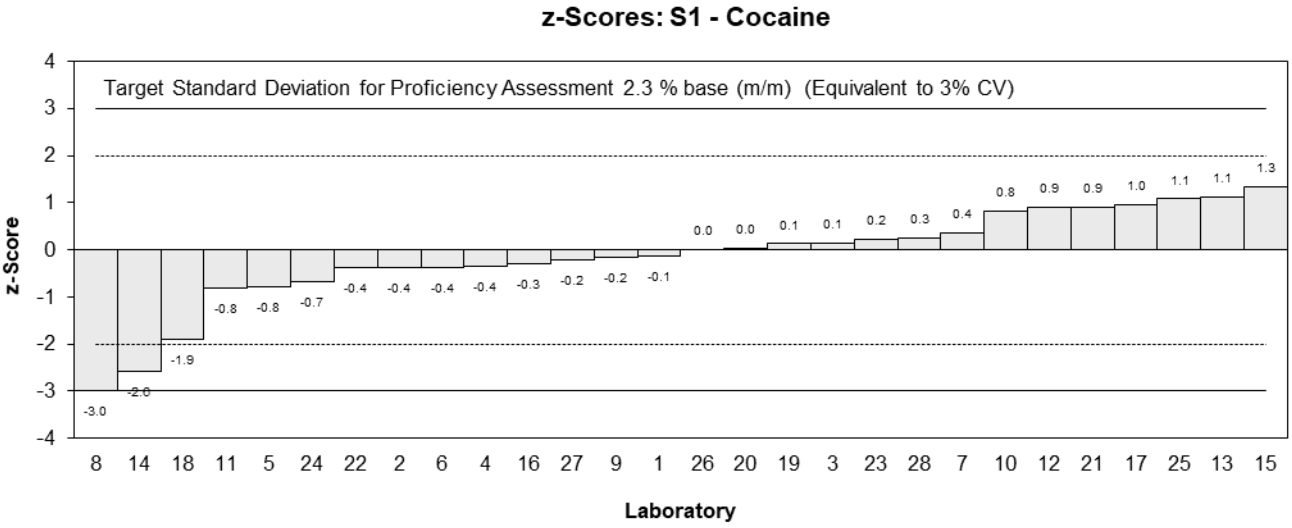
Participant Results

Lab. Code	Result	U	z	E_n
1	76.58	4.59	-0.14	-0.07
2	76	5.3	-0.39	-0.17
3	77.23	3	0.14	0.11
4	76.1	3.4	-0.35	-0.23
5	75.1	5.4	-0.78	-0.33
6	76	4	-0.39	-0.22
7	77.7	3.1	0.35	0.25
8	70	10.5	-2.99	-0.65
9	76.5	2.3	-0.17	-0.16
10	78.8	11	0.82	0.17
11	75	3.3	-0.82	-0.56
12	78.99	7.1	0.91	0.29
13	79.5	4.1	1.13	0.62
14	70.95	2.13	-2.58	-2.57
15	80	3	1.34	0.99
16	76.2	2.91	-0.30	-0.23
17	79.1	8.5	0.95	0.26
18	72.5	NR	-1.91	-4.89
19	77.2	4.7	0.13	0.06
20	77	7.7	0.04	0.01
21	79	7.9	0.91	0.26
22	75.99	11.54	-0.39	-0.08
23	77.4	5	0.22	0.10
24	75.3	1.3	-0.69	-1.01
25	79.4	11.6	1.08	0.21
26	76.9	2.6	0.00	0.00
27	76.4	3.9	-0.22	-0.12
28	77.5	4.7	0.26	0.13

Statistics

Assigned Value	76.9	0.9
Robust Average	76.9	0.9
Median	76.7	0.6
Mean	76.6	0.9
N	28	
Max.	80	
Min.	70	
Robust SD	1.9	
Robust CV	2.5%	


Results: S1 - Cocaine
Kernel Density
Density
Result
Assigned Value = 76.9 ± 0.9 % base (m/m)
Results (% base (m/m))
Laboratory
8 14 18 11 5 24 22 2 6 4 16 27 9 1 26 20 19 3 23 28 7 10 12 21 17 25 13 15 M RA


z-Scores: S1 - Cocaine
Target Standard Deviation for Proficiency Assessment 2.3 % base (m/m) (Equivalent to 3% CV)
z-Score
Laboratory
8 14 18 11 5 24 22 2 6 4 16 27 9 1 26 20 19 3 23 28 7 10 12 21 17 25 13 15
-3.0 -2.6 -1.9 -0.8 -0.8 -0.7 -0.4 -0.4 -0.4 -0.4 -0.3 -0.2 -0.2 -0.1 0.0 0.0 0.1 0.1 0.2 0.3 0.4 0.8 0.9 0.9 1.0 1.1 1.1 1.3

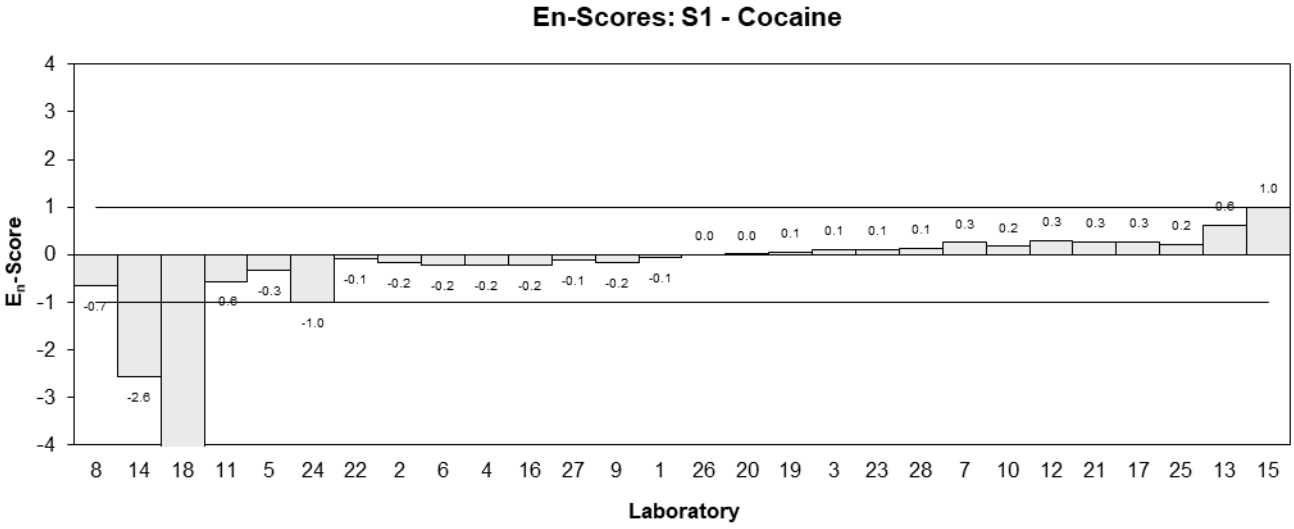

En-Scores: S1 - Cocaine
En-Score
Laboratory
8 14 18 11 5 24 22 2 6 4 16 27 9 1 26 20 19 3 23 28 7 10 12 21 17 25 13 15
-0.7 -2.6 -1.0 0.6 -0.3 -0.1 -0.2 -0.2 -0.2 -0.2 -0.1 -0.2 -0.1 0.0 0.0 0.1 0.1 0.1 0.1 0.3 0.2 0.3 0.3 0.3 0.2 0.6 1.0

Figure 2

Table 6

Sample Details

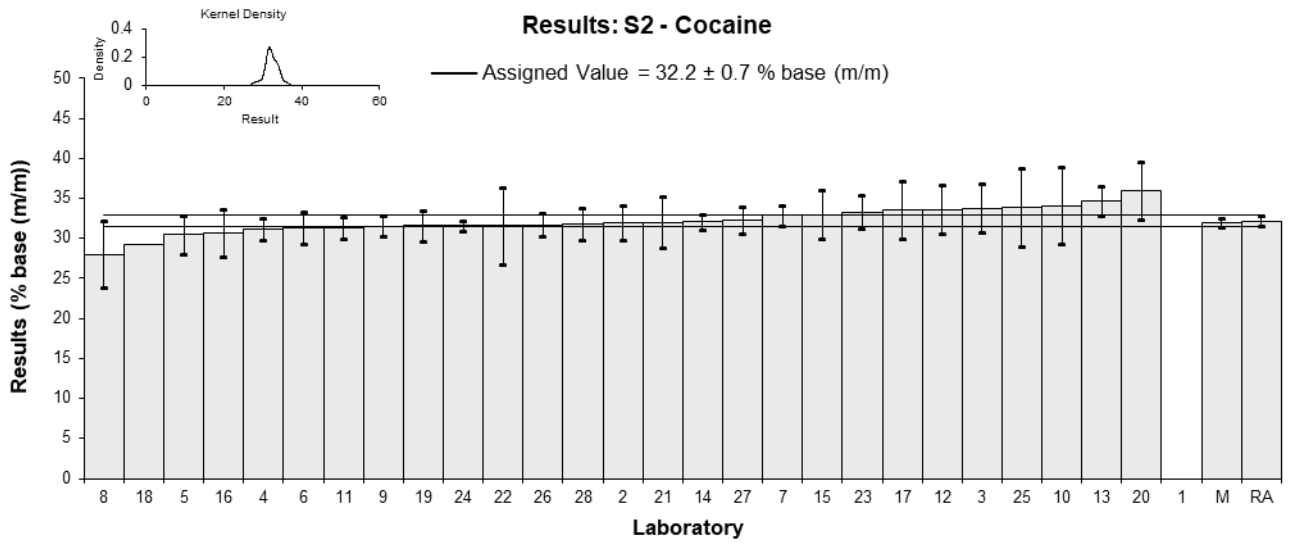
Sample	S2
Analyte	Cocaine
Matrix	Powder
Unit	% base (m/m)

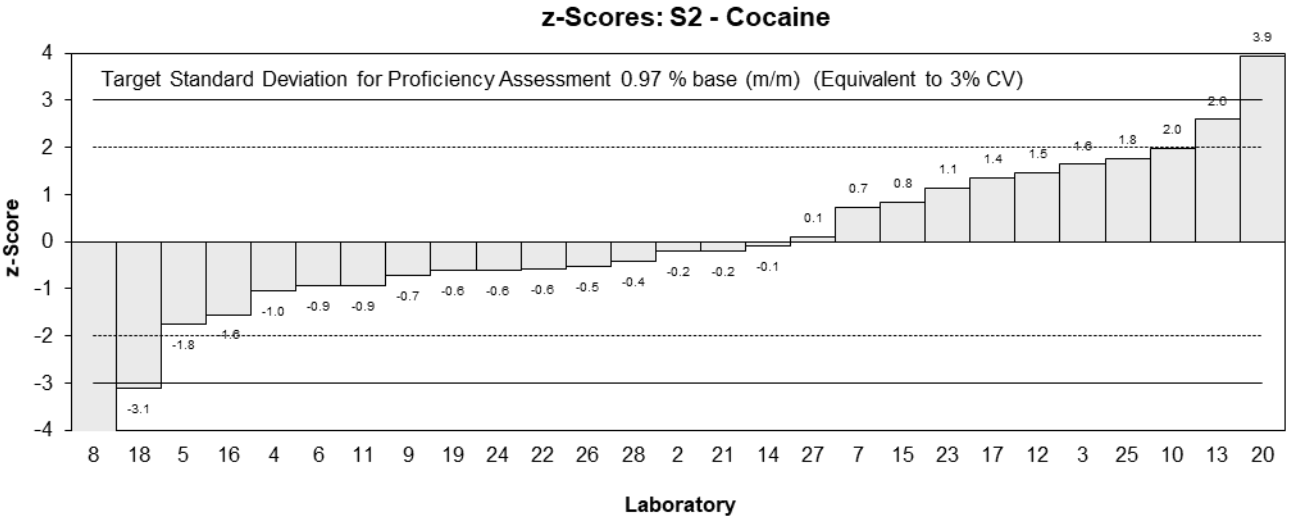
Participant Results

Lab. Code	Result	U	z	E_n
1	NR	NR		
2	32	2.2	-0.21	-0.09
3	33.78	3	1.64	0.51
4	31.2	1.4	-1.04	-0.64
5	30.5	2.4	-1.76	-0.68
6	31.3	2	-0.93	-0.42
7	32.9	1.3	0.72	0.47
8	28	4.2	-4.35	-0.99
9	31.5	1.3	-0.72	-0.47
10	34.1	4.8	1.97	0.39
11	31.3	1.4	-0.93	-0.57
12	33.62	3.0	1.47	0.46
13	34.7	1.8	2.59	1.29
14	32.11	0.96	-0.09	-0.08
15	33	3	0.83	0.26
16	30.7	2.91	-1.55	-0.50
17	33.5	3.6	1.35	0.35
18	29.2	NR	-3.11	-4.29
19	31.6	1.9	-0.62	-0.30
20	36	3.6	3.93	1.04
21	32	3.2	-0.21	-0.06
22	31.63	4.8	-0.59	-0.12
23	33.3	2.1	1.14	0.50
24	31.6	0.6	-0.62	-0.65
25	33.9	4.9	1.76	0.34
26	31.7	1.5	-0.52	-0.30
27	32.3	1.7	0.10	0.05
28	31.8	2.0	-0.41	-0.19

Statistics

Assigned Value	32.2	0.7
Robust Average	32.2	0.7
Median	32.0	0.6
Mean	32.2	0.6
N	27	
Max.	36	
Min.	28	
Robust SD	1.5	
Robust CV	4.6%	


Kernel Density
Density
Result
Results: S2 - Cocaine
Assigned Value = 32.2 ± 0.7 % base (m/m)
Results (% base (m/m))
Laboratory
8 18 5 16 4 6 11 9 19 24 22 26 28 2 21 14 27 7 15 23 17 12 3 25 10 13 20 1 M RA


z-Scores: S2 - Cocaine
Target Standard Deviation for Proficiency Assessment 0.97 % base (m/m) (Equivalent to 3% CV)
z-Score
Laboratory
8 18 5 16 4 6 11 9 19 24 22 26 28 2 21 14 27 7 15 23 17 12 3 25 10 13 20
-3.1 -1.8 -1.6 -1.0 -0.9 -0.9 -0.7 -0.6 -0.6 -0.6 -0.5 -0.4 -0.2 -0.2 -0.1 0.1 0.7 0.8 1.1 1.4 1.5 1.6 1.8 2.0 2.6 3.9

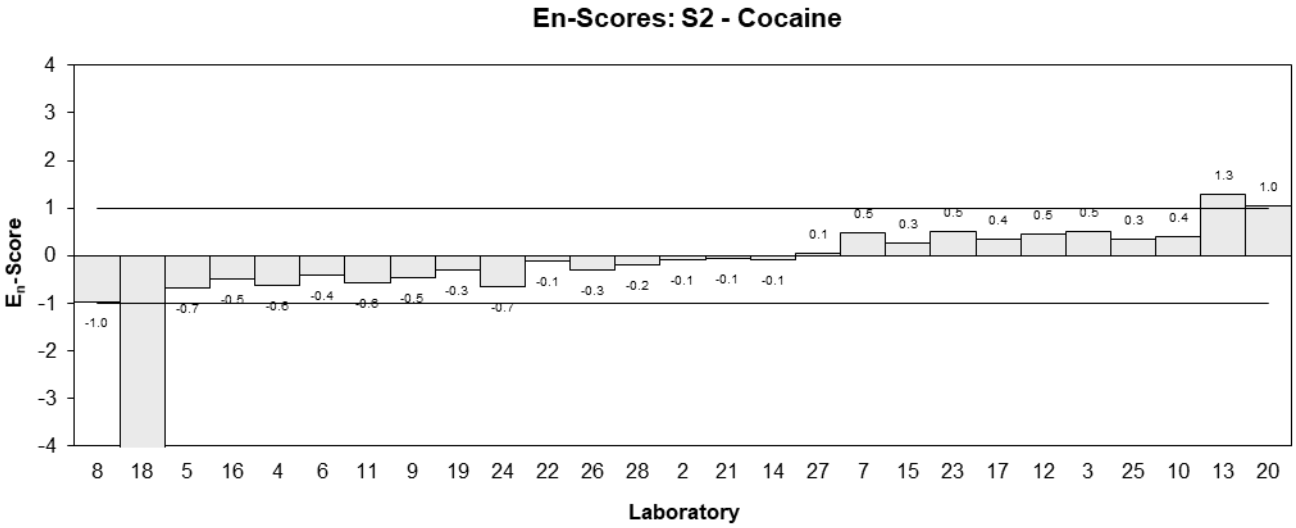

En-Scores: S2 - Cocaine
En-Score
Laboratory
8 18 5 16 4 6 11 9 19 24 22 26 28 2 21 14 27 7 15 23 17 12 3 25 10 13 20
-1.0 -0.7 -0.5 -0.6 -0.4 -0.6 -0.5 -0.3 -0.7 -0.1 -0.3 -0.2 -0.1 -0.1 -0.1 0.1 0.5 0.3 0.5 0.4 0.5 0.5 0.3 0.4 1.3 1.0

Figure 3

Table 7

Sample Details

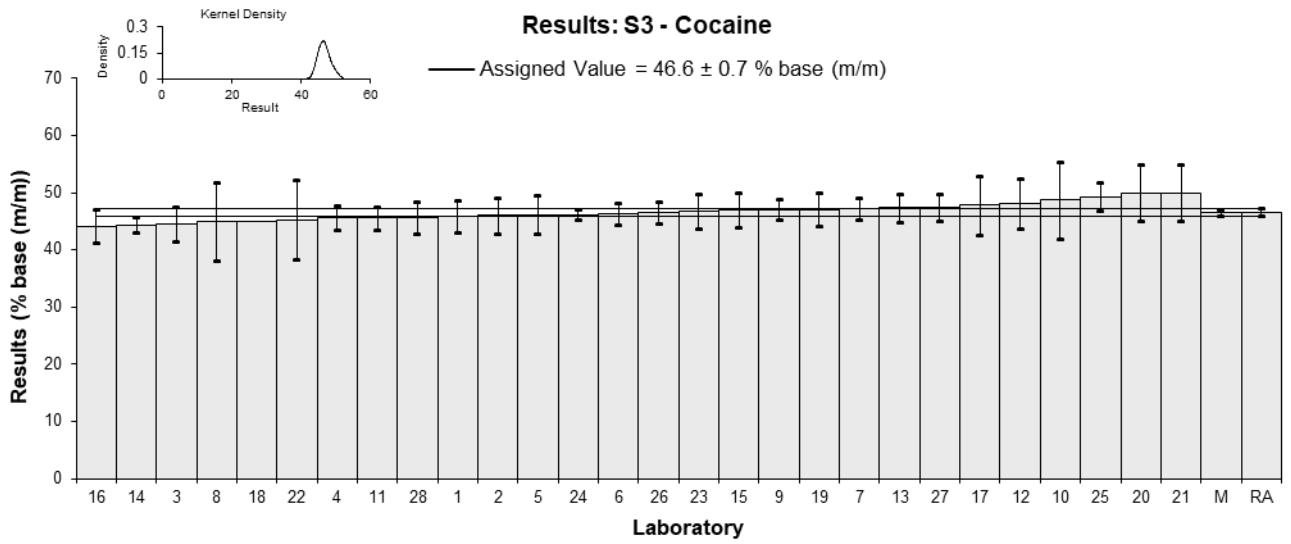
Sample	S3
Analyte	Cocaine
Matrix	Powder
Unit	% base (m/m)

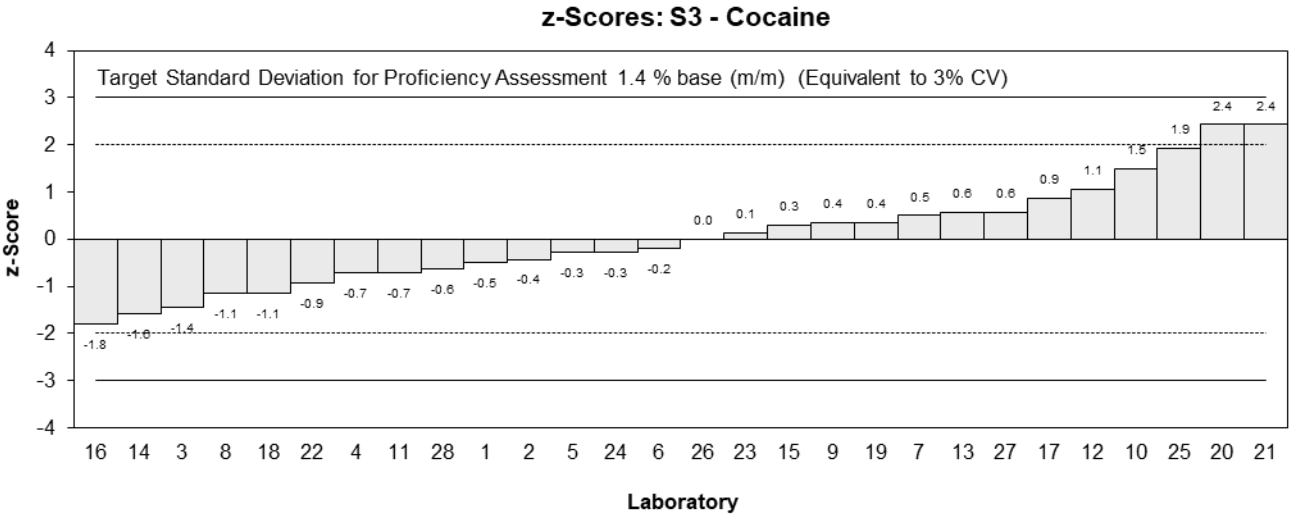
Participant Results

Lab. Code	Result	U	z	E_n
1	45.91	2.75	-0.49	-0.24
2	46	3.2	-0.43	-0.18
3	44.58	3	-1.44	-0.66
4	45.6	2.1	-0.72	-0.45
5	46.2	3.3	-0.29	-0.12
6	46.3	2	-0.21	-0.14
7	47.3	1.9	0.50	0.35
8	45	6.8	-1.14	-0.23
9	47.1	1.7	0.36	0.27
10	48.7	6.8	1.50	0.31
11	45.6	2	-0.72	-0.47
12	48.08	4.3	1.06	0.34
13	47.4	2.5	0.57	0.31
14	44.39	1.33	-1.58	-1.47
15	47	3	0.29	0.13
16	44.1	2.91	-1.79	-0.84
17	47.8	5.2	0.86	0.23
18	45	NR	-1.14	-2.29
19	47.1	2.9	0.36	0.17
20	50	5.0	2.43	0.67
21	50	5	2.43	0.67
22	45.3	6.88	-0.93	-0.19
23	46.8	3	0.14	0.06
24	46.2	0.8	-0.29	-0.38
25	49.3	2.4	1.93	1.08
26	46.6	1.9	0.00	0.00
27	47.4	2.4	0.57	0.32
28	45.7	2.8	-0.64	-0.31

Statistics

Assigned Value	46.6	0.7
Robust Average	46.6	0.7
Median	46.5	0.6
Mean	46.7	0.6
N	28	
Max.	50	
Min.	44.1	
Robust SD	1.6	
Robust CV	3.4%	


Results: S3 - Cocaine
Kernel Density
Density
Result
Assigned Value = 46.6 ± 0.7 % base (m/m)
Results (% base (m/m))
16 14 3 8 18 22 4 11 28 1 2 5 24 6 26 23 15 9 19 7 13 27 17 12 10 25 20 21 M RA
Laboratory


z-Scores: S3 - Cocaine
Target Standard Deviation for Proficiency Assessment 1.4 % base (m/m) (Equivalent to 3% CV)
z-Score
16 14 3 8 18 22 4 11 28 1 2 5 24 6 26 23 15 9 19 7 13 27 17 12 10 25 20 21
Laboratory

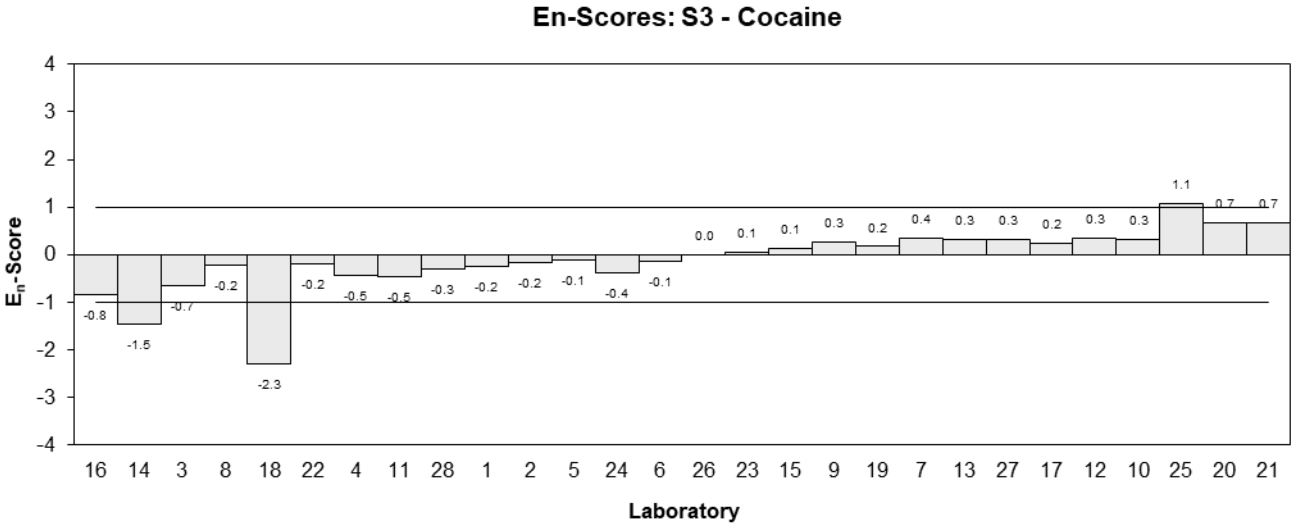

En-Scores: S3 - Cocaine
En-Score
16 14 3 8 18 22 4 11 28 1 2 5 24 6 26 23 15 9 19 7 13 27 17 12 10 25 20 21
Laboratory

Figure 4

Table 8 Participants' Identification of Cutting Agents*

Lab. Code	Cutting Agents		
	S1	S2	S3
Preparation	Phenacetin	Phenacetin and Quinine	Sucrose
1	phenacetin	quinine, phenacetin	sucrose
2	phenacetin	phenacetin, quinine	n/a
3			
4	phenacetin	phenacetin, quinine	
5		phenacetin	
6	Phenacetin	Phenacetin & Quinidine	
7	phenacetin	phenacetin	
8	Phenacetin	Phenacetin	none
9	phenacetin	phenacetin and quinine	
10	Phenacetin	Phenacetin, Quinine	
11	Phenacetin	Phenacetin	Sucrose
12	phenacetin	phenacetin	sucrose
13		Phenacetin	
14	Phenacetin	Phenacetin	Sucrose
15	Phenacetin	Phenacetin	-
16	Phenacetin	Phenacetin	N/A
17			
18	Phenacetin : 10.1 %	Phenacetin : 52.3 % Quinine	Sucrose : 42.5 %
19	Phenacetin	Phenacetin, Quinine	-
20	Phenacetin	Phenacetin	Sucrose
21	phenacetin	phenacetin	sucrose
22	Phenacetin	Phenacetin	none detected (inert to GCMS method)
23	phenacetin (9.7%)	phenacetin (54.3%), quinine (not quantified)	/
24	phenacetin	phenacetin, probably quinine	sucrose
25	phenacetin	phenacetin, quinine	sucrose
26	phenacetin	phenacetin	
27	Phenacetin	Phenacetin, Quinine	Sucrose
28	Phenacetin	Phenacetin, Quinine	-

* Some responses may have been modified so that the participant cannot be identified.

6 DISCUSSION OF RESULTS

6.1 Assigned Value

The assigned values were the robust averages of the results reported by participants. The robust averages and associated expanded uncertainties were calculated using the procedure described in ISO 13528:2015.[5] Results less than 50% and greater than 150% of the robust average were removed before calculation of the assigned value, if applicable.[3,4] The calculation of the expanded uncertainty for robust averages is presented in Appendix 1, with Sample S1 as an example.

Traceability: The consensus of participants' results is not traceable to any external reference, so although expressed in SI units, metrological traceability has not been established.

6.2 Measurement Uncertainty Reported by Participants

Participants were asked to report an estimate of the expanded MU associated with their results and the basis of this uncertainty estimate (Section 3.3). Some participants reported using the NATA GAG Estimating and Reporting MU as their guide; NATA no longer publishes this document.[9]

It is a requirement of ISO/IEC 17025 that laboratories have procedures to estimate the uncertainty of chemical measurements and to report this uncertainty in specific circumstances, including when the client's instruction so requires.[7] From July 2012 this is also a requirement of the ANAB-ASCLD/LAB international accreditation program.

Of 83 numeric results, 80 (96%) were reported with an associated expanded MU. Laboratory **18** did not report any uncertainties; this laboratory reported that they were not accredited.

The magnitude of reported uncertainties was within the range 1.7% to 15% relative. Of the 80 expanded MUs, 61 (76%) were between 3% and 10% relative to the result. Laboratories reporting uncertainties smaller than 3% or larger than 10% relative may wish to consider whether these estimates are realistic or fit for purpose.

Uncertainties associated with results returning a satisfactory *z* score but an unsatisfactory E_n score may have been underestimated.

In some cases the results were reported with an inappropriate number of significant figures. Including too many significant figures may inaccurately reflect the precision of measurements. The recommended format is to write the uncertainty to no more than two significant figures, and then to write the result with the corresponding number of decimal places. For example, instead of 76.58 ± 4.59%, it is better to report this as 76.6 ± 4.6%.[8]

6.3 *z* Score

A target SD equivalent to 3% PCV was used to calculate *z* scores. The CVs predicted by the Thompson-Horwitz equation,[6] target SDs (as PCV), and between-laboratory CVs (as robust CV) obtained in this study are presented in Table 9.

Table 9 Comparison of Thompson-Horwitz CVs, Target SDs and Between-Laboratory CVs

Sample	Analyte	Assigned value (% base (m/m))	Thompson-Horwitz CV (%)	Target SD (as PCV) (%)	Between-Laboratory CV (%)
S1	Cocaine	76.9	1.1	3	2.5
S2	Cocaine	32.2	1.8	3	4.6
S3	Cocaine	46.6	1.5	3	3.4

Of 83 results for which *z* scores were calculated, 75 (90%) returned a *z* score of $|z| \leq 2.0$, indicating a satisfactory performance.

Twenty-one participants: **2**, **3**, **4**, **5**, **6**, **7**, **9**, **10**, **11**, **12**, **15**, **16**, **17**, **19**, **22**, **23**, **24**, **25**, **26**, **27** and **28** returned satisfactory *z* scores for all three samples. Laboratory **1** reported numeric results for Samples S1 and S3 only, and both returned satisfactory *z* scores.

Six participants returned at least one questionable or unsatisfactory *z* score.

The dispersal of participants' *z* scores is presented graphically by laboratory in Figure 5.

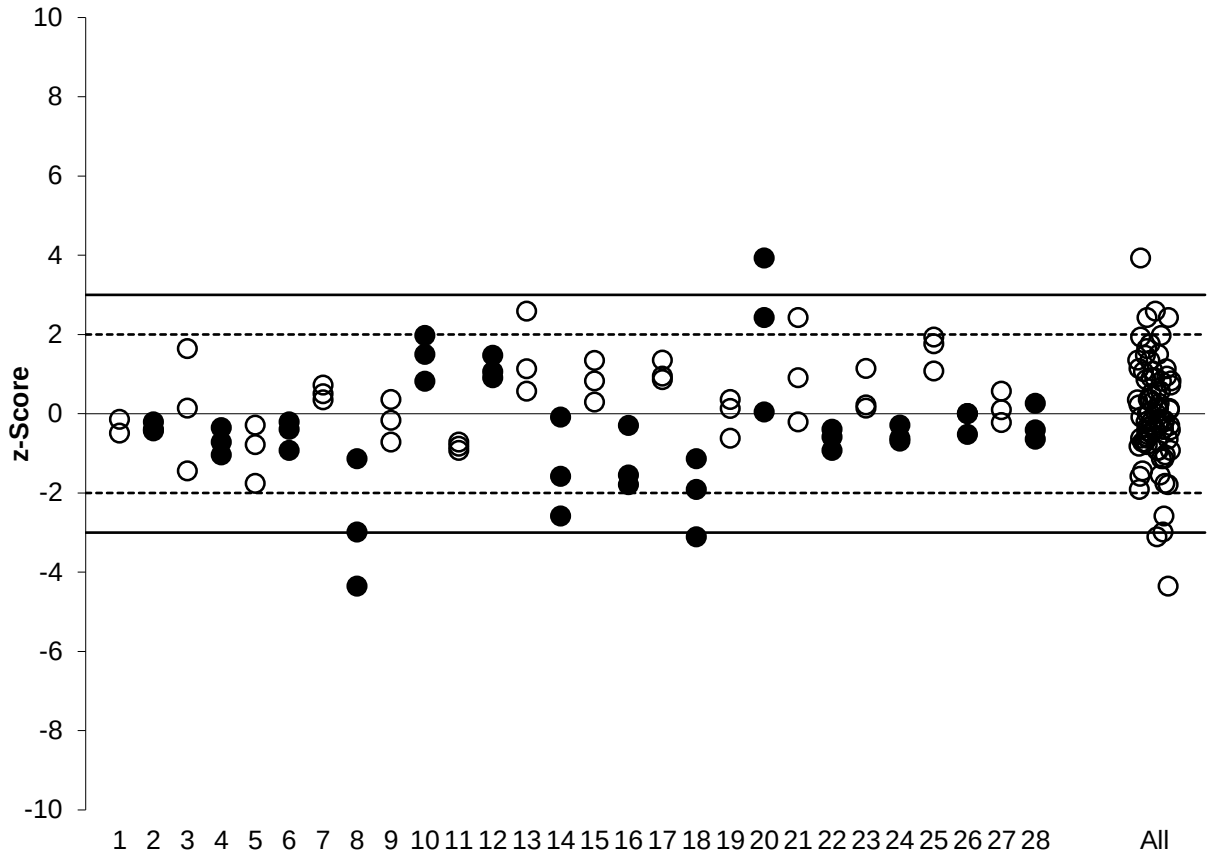


Figure 5 *z* Score Dispersal by Laboratory

6.4 E_n Score

If a participant did not report an expanded uncertainty with a result, an expanded uncertainty of zero (0) was used to calculate their E_n score.

Of 83 results for which E_n scores were calculated, 74 (89%) returned a satisfactory E_n score of $|E_n| \leq 1.0$, indicating agreement of the participant's result with the assigned value within their respective expanded uncertainties.

Twenty-one participants: **2**, **3**, **4**, **5**, **6**, **7**, **8**, **9**, **10**, **11**, **12**, **15**, **16**, **17**, **19**, **21**, **22**, **23**, **26**, **27** and **28** returned satisfactory E_n scores for all three samples. Laboratory **1** reported numeric results for Samples S1 and S3 only, and both returned satisfactory E_n scores.

Laboratory **18** returned unsatisfactory E_n scores for all three samples.

The dispersal of participants' E_n scores is presented graphically by laboratory in Figure 6.

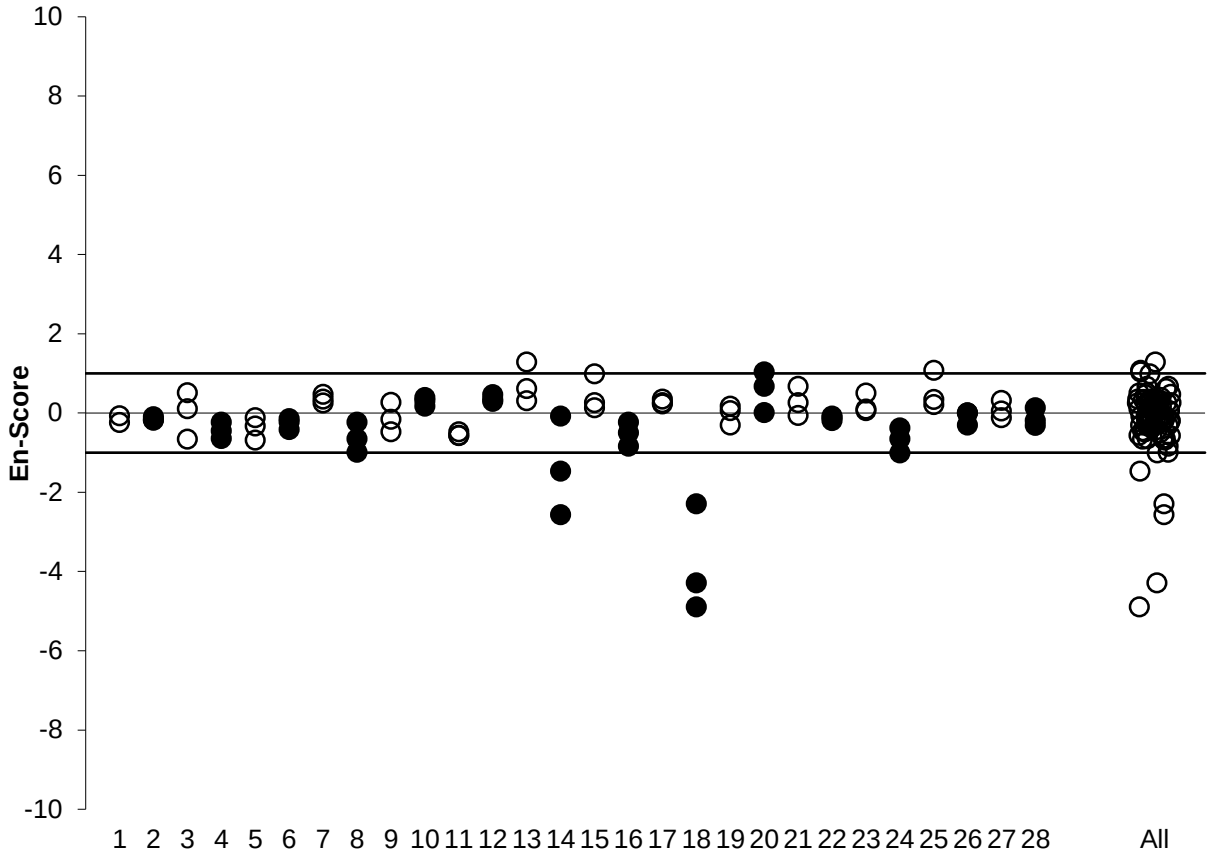


Figure 6 E_n score Dispersal by Laboratory

6.5 Identification of Cutting Agent

The test samples were prepared using seizures of cocaine hydrochloride approximately 84% base (m/m), supplied by the Australian Federal Police. The study coordinator added phenacetin to Sample S1, phenacetin and quinine to Sample S2, and sucrose to Sample S3.

Twenty-six participants (93%) reported on the identity of the cutting agents in at least one sample (Table 8).

Laboratories **1**, **18**, **24**, **25** and **27** correctly identified all cutting agents in the samples.

Phenacetin was added to both Samples S1 and S2, in different proportions (approximately 9% (w/w) in Sample S1 and 53% (w/w) in Sample S2). Most participants reporting on the cutting agents identified phenacetin in both samples, except for Laboratories **5** and **13** who reported phenacetin in Sample S2 only.

A small amount of quinine was added also to Sample S2 (approximately 9% (w/w)). Twelve participants correctly identified the presence of quinine in this sample. One participant reported quinidine.

For Sample S3, 10 participants correctly identified that sucrose was used as the cutting agent.

6.6 Participants' Analytical Methods

Participants were requested to analyse the samples using their normal test methods and to report a single result for each sample as they would normally report to a client. Results reported in this way reflect the true variability of results reported to laboratory clients. The method descriptions provided by participants are presented in Table 1.

A summary of accreditation status, methods and reference standards is presented in Table 10.

Table 10 Summary of Participants' Analyses

		Lab. Code
Accreditation	Yes to ISO/IEC 17025	2, 3, 4, 5, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 19, 20, 21, 22, 23, 24, 25, 27, 28
	Not Accredited / Not Reported	1, 6, 18, 26
Average Sample Mass Used per Analysis (mg)	5 – 20	1, 4, 5, 6, 7, 12, 13, 14, 15, 16, 17, 23
	21 – 30	2, 19, 20, 21, 26, 27, 28
	31 – 50	3, 8, 9, 10, 11, 18, 22, 24
	51 – 150	25
Conversion to Base?	Yes	1, 3, 4, 5, 7, 9, 10, 12, 13, 14, 15, 16, 23, 25, 26
	No	2, 6, 11, 17, 18, 19, 20, 21, 22, 24, 27, 28
	Not Reported	8
Instrument Used for Quantification	HPLC-DAD	1, 2, 6, 9, 12, 16, 22, 25 (S3)
	HPLC-UV/Vis	14, 17
	UPLC-DAD	5, 18, 19, 20, 21, 26, 28
	UPLC-MS/MS	8
	GC-FID	3, 7, 10, 11, 13, 15, 23, 24, 25 (S1 and S2), 27
	QNMR	4
Solvent	Acetonitrile/Water(/Other)	1, 5, 8, 12, 14, 16, 22
	Methanol	2, 3, 6, 7, 9, 10, 17, 19, 20, 21, 28
	Ethanol	13, 15, 18, 23, 24, 27
	Other / Not reported	4, 11, 25, 26
Sources of Calibration Standard	NMI Australia	4, 5, 11, 17, 18, 26, 27
	Lipomed	1, 2, 7, 9, 15, 19, 23, 28
	Merck / Sigma Aldrich	10, 12, 22
	Johnson Matthey / MacFarlan Smith	14, 16, 25
	Cerilliant	20, 21
	Other	3, 8, 13, 24
	Not Reported	6

Laboratory **25** reported using two different methodologies, due to the interference from the phenacetin cutting agent in Samples S1 and S2.

Laboratory **1** did not report a result for Sample S2, commenting that the quinine cutting agent interfered with their sample analysis. This participant should modify or develop cocaine quantitation methods that are not affected by interferences including quinine, as such cutting agents may be a component in routine cocaine samples.[10]

Plots of the *z* score versus various methodology parameters are presented in Figures 7 to 11. No significant trends were observed.

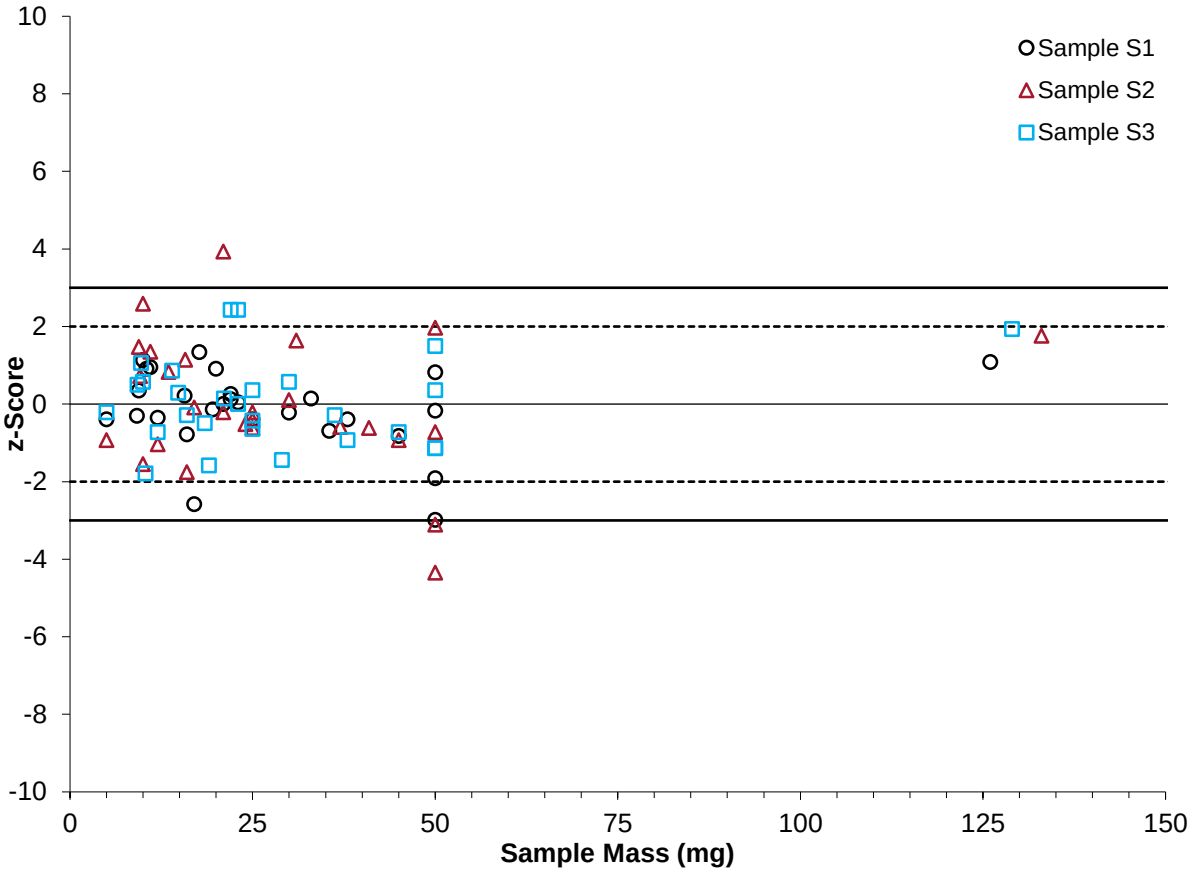


Figure 7 *z* Score vs Sample Mass Used For Analysis

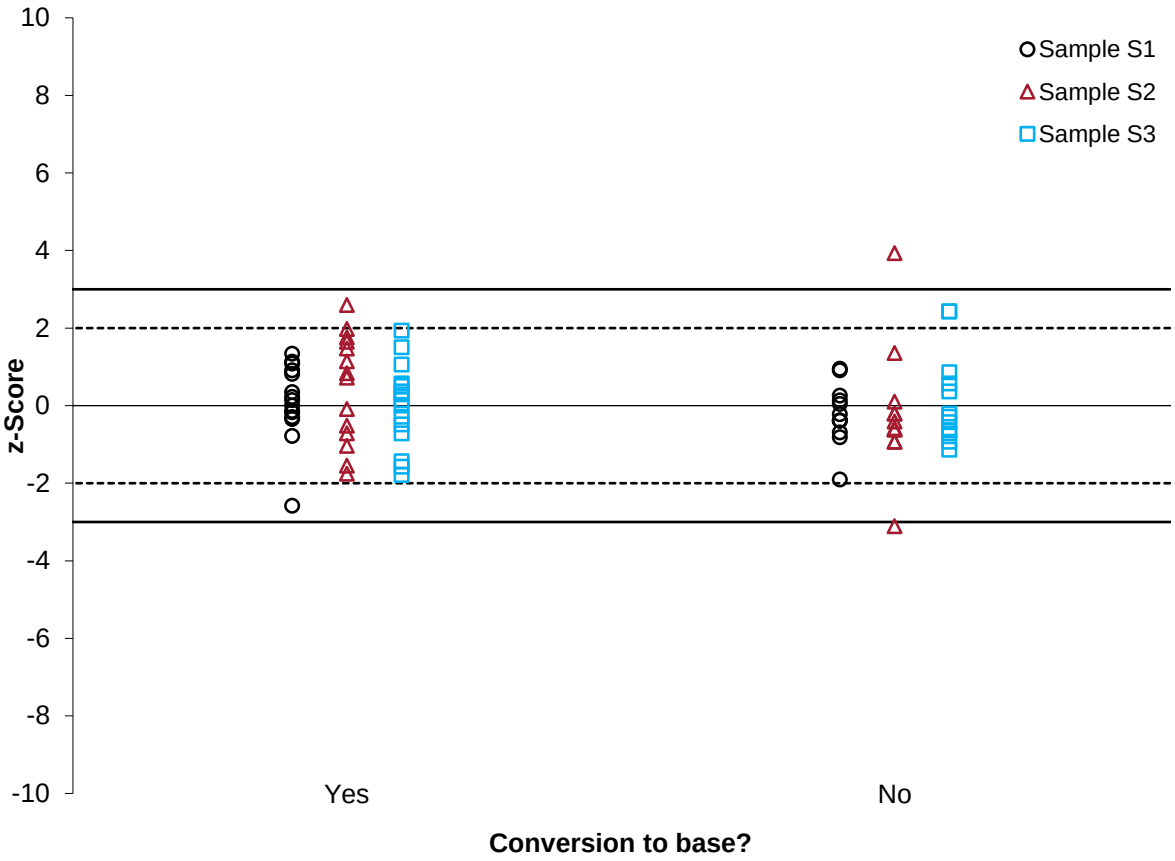


Figure 8 *z* Score vs Sample Processing

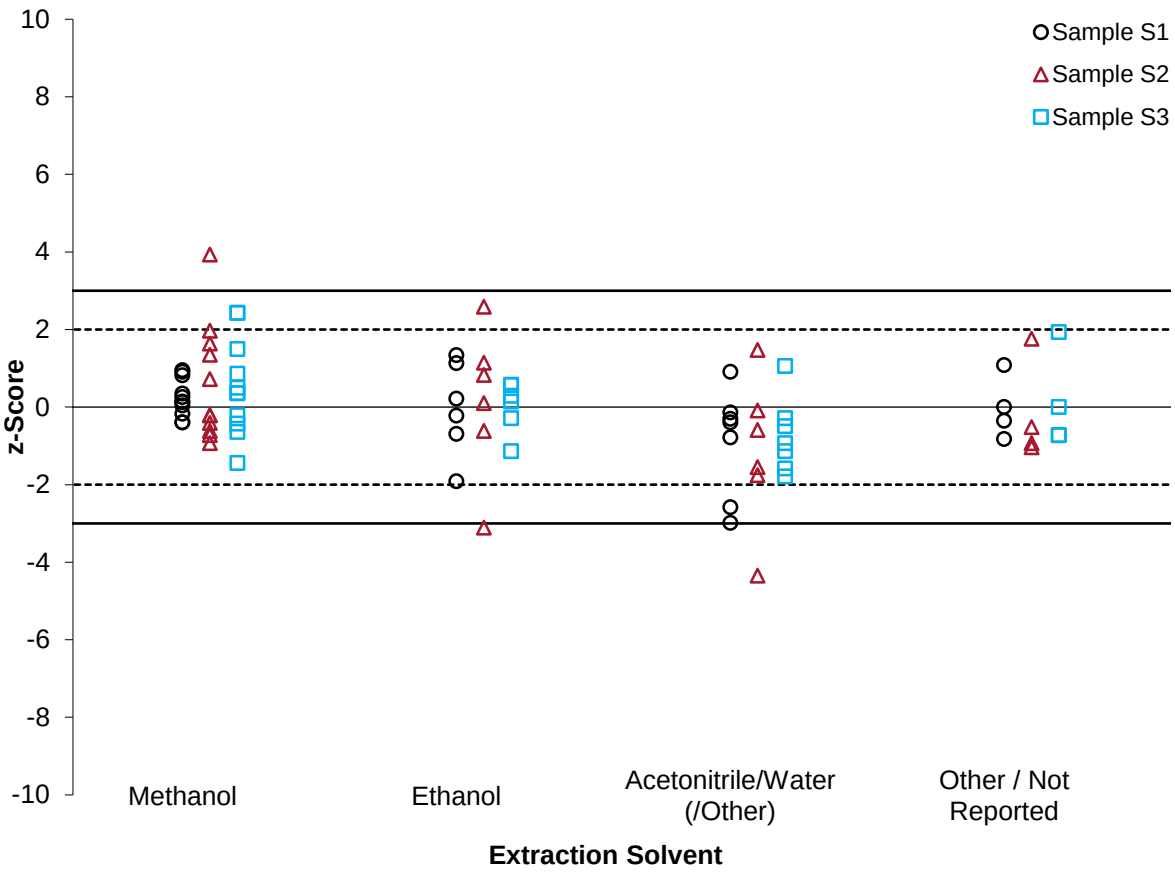


Figure 9 *z* Score vs Extraction Solvent

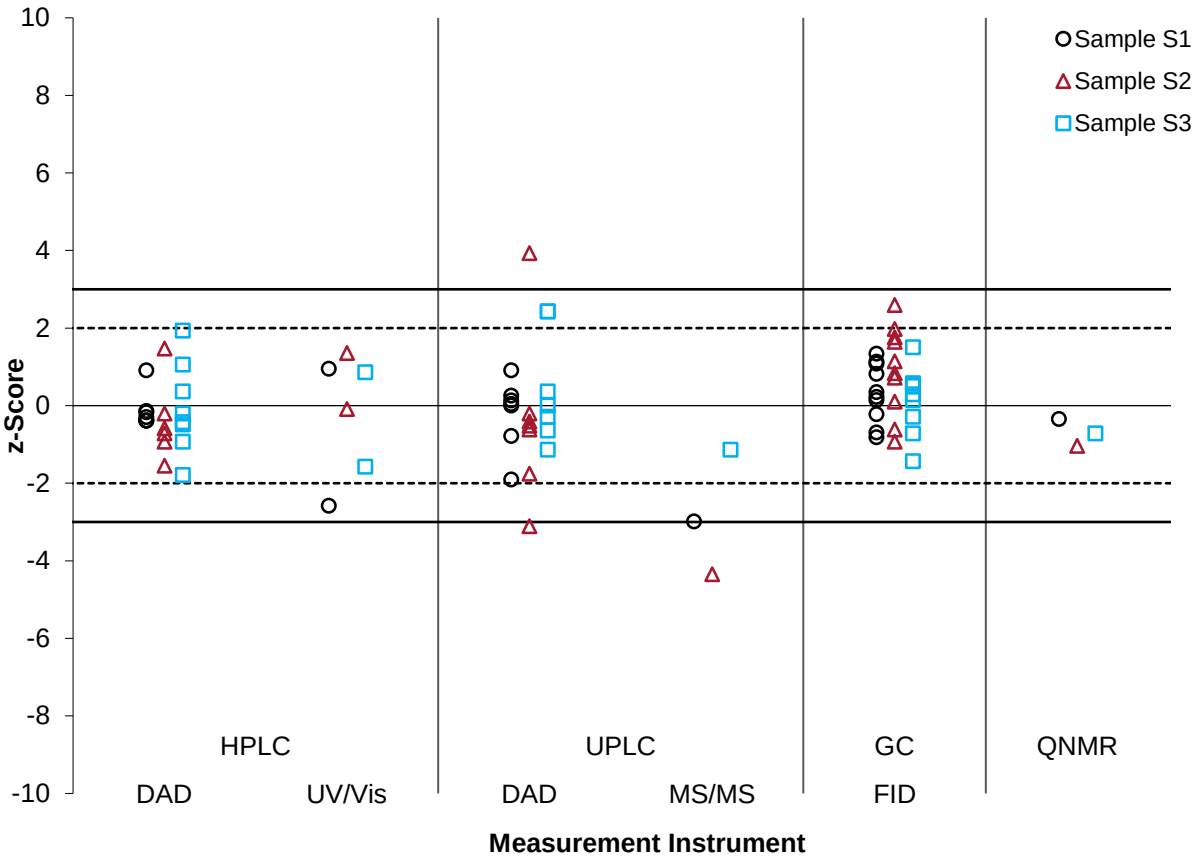


Figure 10 *z* Score vs Measurement Instrument

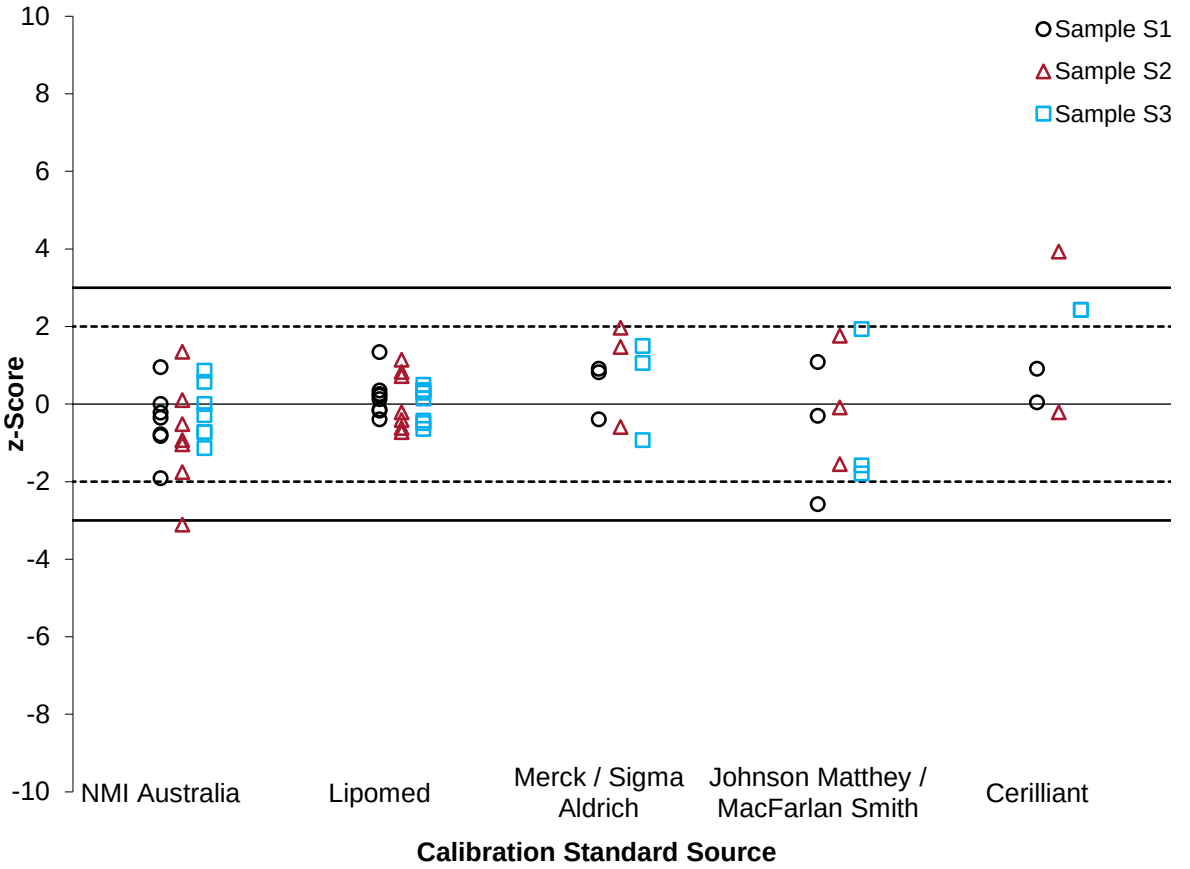


Figure 11 *z* Score vs Source of Calibration Standard

6.7 Comparison of Results and Date of Analysis

As there were delays with sample delivery to some participants, the samples were analysed by participants over the course of approximately 3 months. No trend was found between when the samples were analysed and the results obtained (Figure 12).

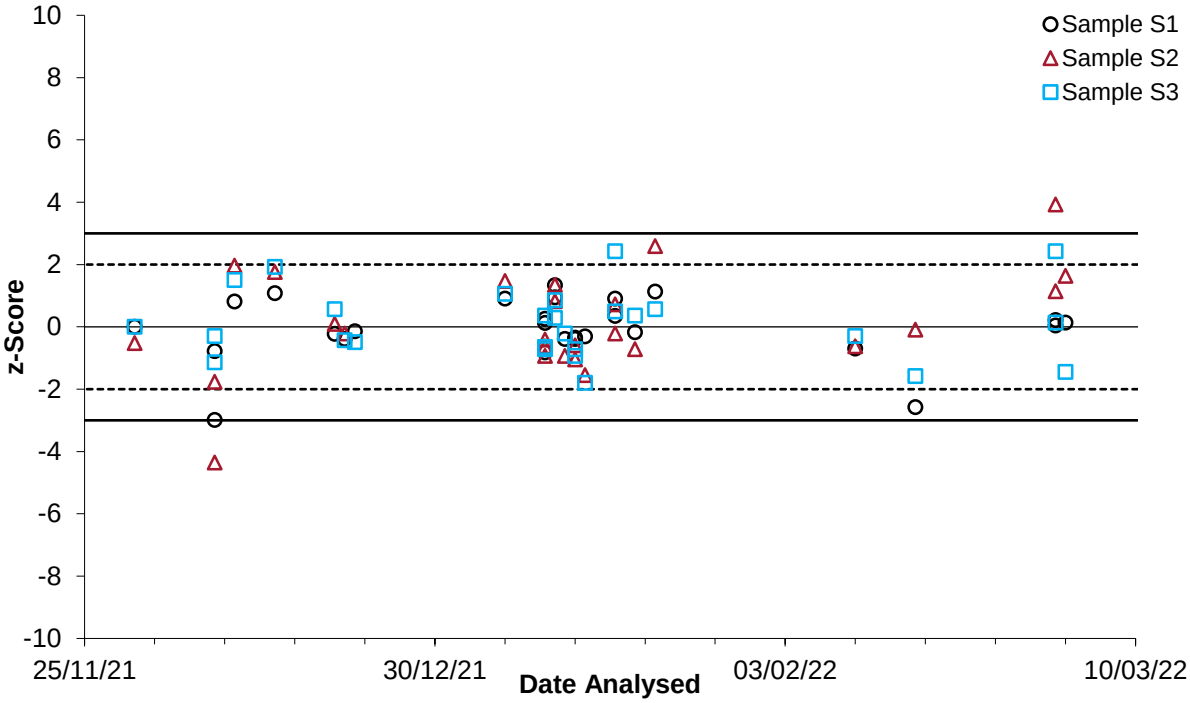


Figure 12 *z* Score vs Sample Analysis Date

6.8 Comparison with Previous Cocaine PT Studies

To enable direct comparison with previous Cocaine PT studies, the target SD used to calculate *z* scores has been kept constant at 3% PCV.

A summary of the satisfactory performance, presented as a percentage of the total number of scores, obtained by participants from 2012 to 2021 (last 10 studies) are presented in Figure 13. The average proportion of satisfactory *z* scores and E_n scores over this period is 80% and 83% respectively. While each PT study has a different group of participants, taken as a group, the performance over this period has been improving.

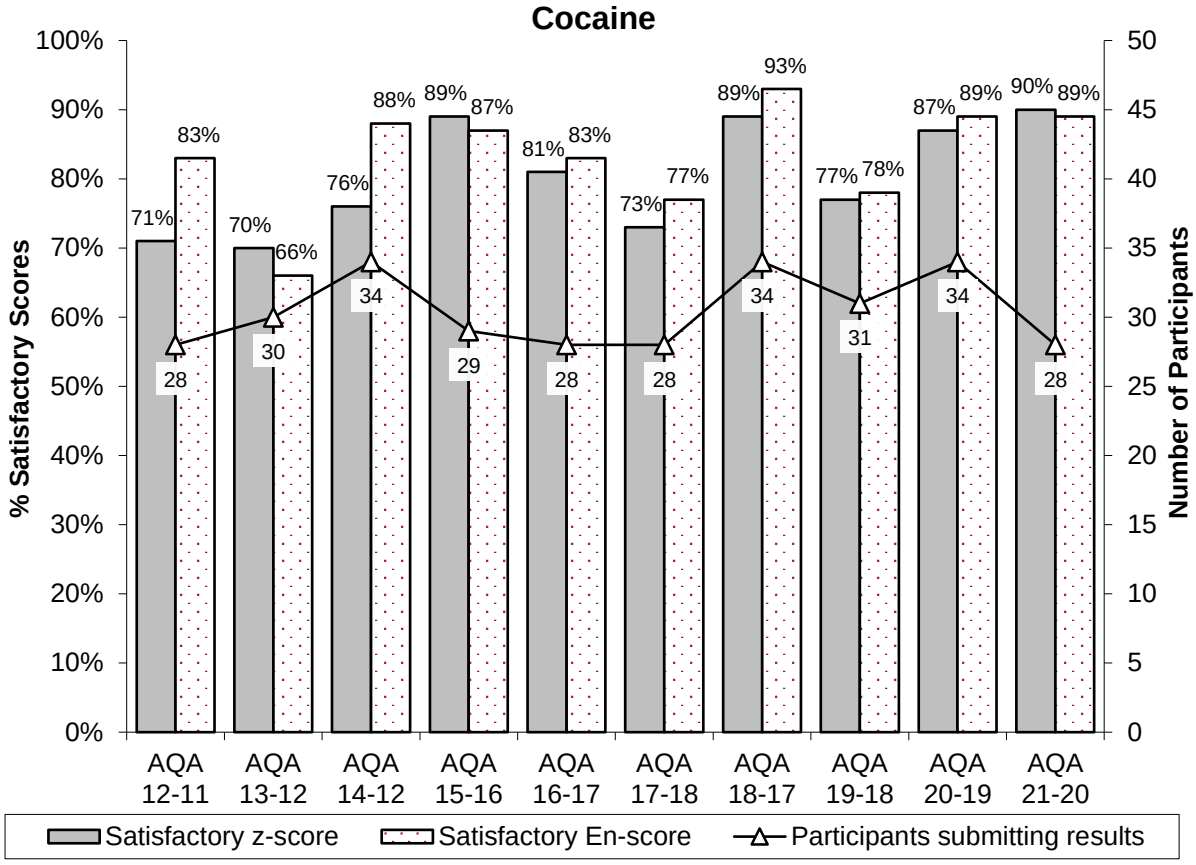


Figure 13 Summary of Participants' Performance in Cocaine PT Studies

A number of participants have consistently participated in NMI Cocaine PT studies, and individual performance history reports are emailed to each participant at the end of the study. The consideration of *z* scores for an analyte over time provides much more useful information than a single *z* score. Over time, laboratories should expect at least 95% of their scores to lie within the range $|z| \leq 2.0$. Scores in the range $2.0 < |z| < 3.0$ can occasionally occur, however, these should be interpreted in conjunction with the other scores obtained by that laboratory. For example, a trend of *z* scores on one side of the zero line is an indication of method or laboratory bias.

A summary of individual laboratory's performances over the last ten NMI Cocaine PT studies is presented in Figures 14 and 15 for Australian and international laboratories respectively. One Australian and two international laboratories have achieved satisfactory *z* scores across all samples in all cocaine PT studies participated in over this period.

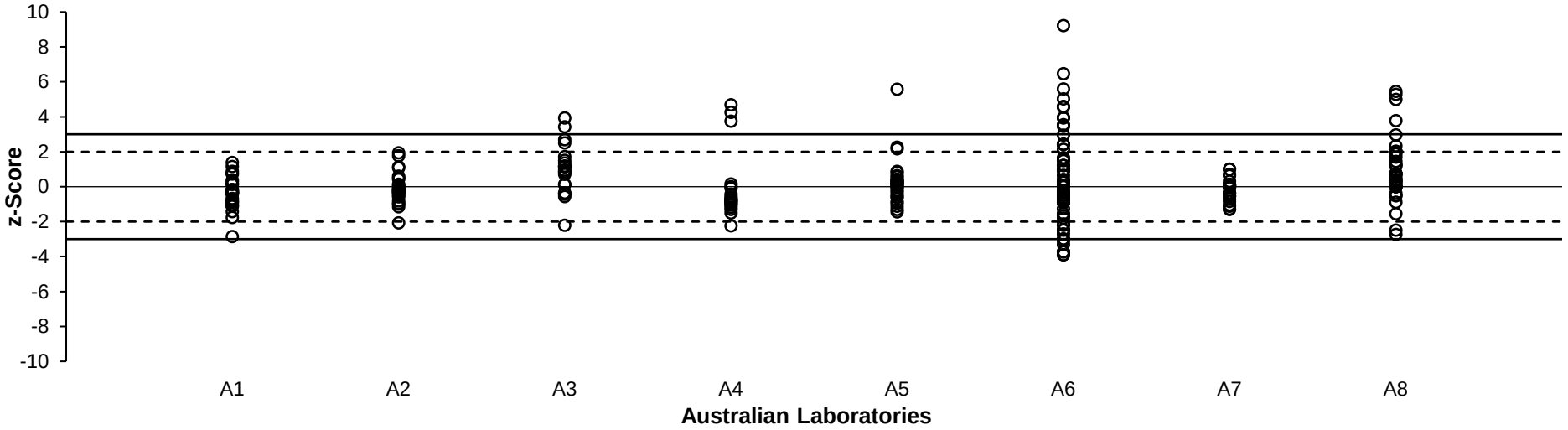


Figure 14 Summary of Australian Participants' *z* Scores in NMI Cocaine PT Studies

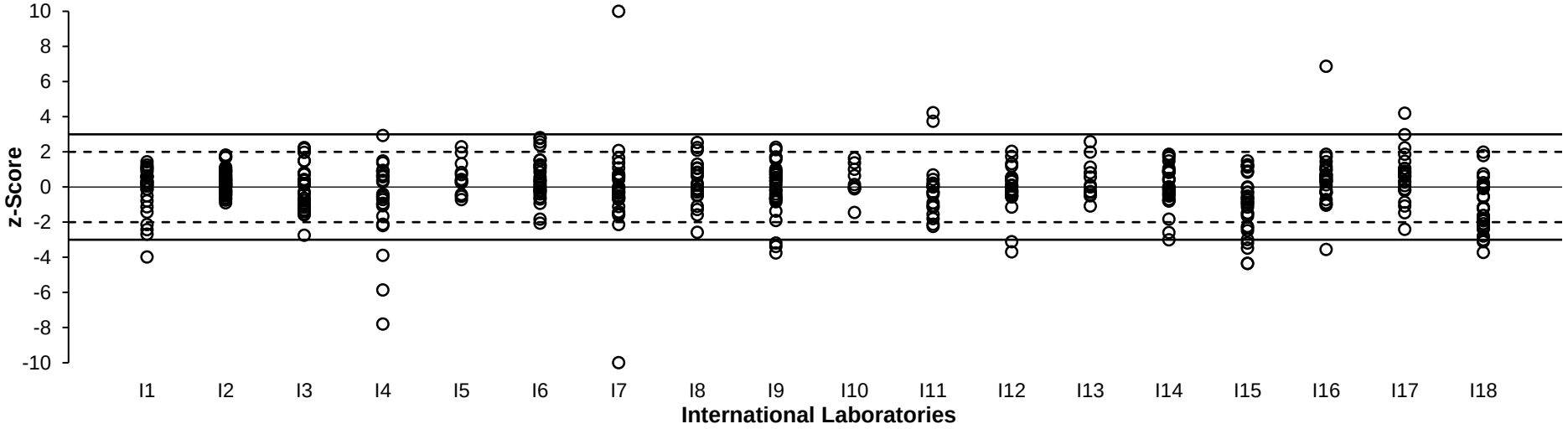


Figure 15 Summary of International Participants' *z* Scores in NMI Cocaine PT Studies

A comparison of all results from Australian and international laboratories in NMI Cocaine PT studies over the last ten years is presented in Figure 16. Overall both groups have performed very similarly, achieving 80% and 81% satisfactory *z* scores over this period for Australian laboratories and international laboratories respectively.

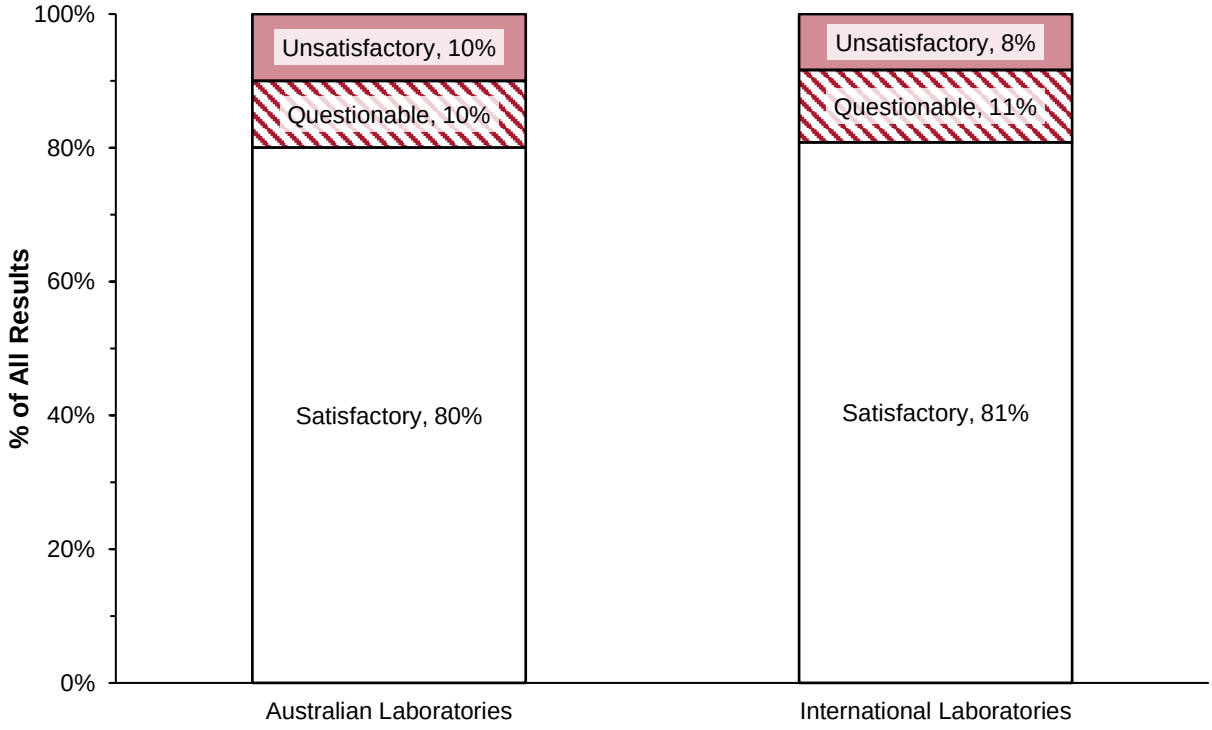


Figure 16 Comparison of Australian and International Laboratories in NMI Cocaine PT Studies

7 REFERENCES

[1] ISO/IEC 17043:2010, *Conformity assessment – General requirements for proficiency testing*.

[2] NMI, 2021, *Study Protocol for Proficiency Testing*, viewed April 2022, <https://www.industry.gov.au/sites/default/files/2020-10/cpt_study_protocol.pdf>

[3] NMI, 2022, *Chemical Proficiency Testing Statistical Manual*, viewed April 2022, <https://www.industry.gov.au/sites/default/files/2019-07/cpt_statistical_manual.pdf>

[4] Thompson, M., Ellison, S.L.R. and Wood, R., 2006, 'The International Harmonized Protocol for the Proficiency Testing of Analytical Chemistry Laboratories', *Pure Appl. Chem.*, vol. 78, pp. 145-196.

[5] ISO 13528:2015, *Statistical methods for use in proficiency testing by interlaboratory comparison*.

[6] Thompson, M., 2000, 'Recent trends in inter-laboratory precision at ppb and sub-ppb concentrations in relation to fitness for purpose criteria in proficiency testing', *Analyst*, vol. 125, pp. 385-386.

[7] ISO/IEC 17025:2017, *General requirements for the competence of testing and calibration laboratories*.

[8] Eurachem/CITAC Guide CG 4, QUAM:2012.P1, *Quantifying Uncertainty in Analytical Measurement*, 3rd Edition, viewed April 2022, <http://eurachem.org/images/stories/guides/pdf/quam2012_P1.pdf>

[9] NATA, 2020, *Update to Measurement Uncertainty resources*, viewed April 2022, <https://nata.com.au/news/update-to-measurement-uncertainty-resources/>

[10] UNODC, 2005, *ST/NAR/35 Methods for Impurity Profiling of Heroin and Cocaine*.

APPENDIX 1 – ROBUST AVERAGE AND ASSOCIATED UNCERTAINTY, *z* SCORE AND E_n SCORE CALCULATIONS

A1.1 Robust Average and Associated Uncertainty

Robust averages were calculated using the procedure described in ISO 13528:2015.[5] The associated uncertainties were estimated as according to Equation 4.

$$u_{rob\ av} = 1.25 \times \frac{S_{rob\ av}}{\sqrt{p}} \qquad \textit{Equation 4}$$

where:

$u_{rob\ av}$ is the standard uncertainty of the robust average

$S_{rob\ av}$ is the standard deviation of the robust average

p is the number of results

The expanded uncertainty ($U_{rob\ average}$) is the standard uncertainty multiplied by a coverage factor of 2 at approximately 95% confidence level.

A worked example for Sample S1 is set out below in Table 11.

Table 11 Uncertainty of Sample S1 Robust Average

No. results (p)	28
Robust average	76.88% base (m/m)
$S_{rob\ average}$	1.94% base (m/m)
$u_{rob\ average}$	0.46% base (m/m)
k	2
$U_{rob\ average}$	0.92% base (m/m)

Therefore, the robust average of Sample S1 is 76.9 ± 0.9% base (m/m).

A1.2 *z* Score and E_n Score Calculations

For each participant's result, a *z* score and E_n score are calculated according to Equations 2 and 3 respectively (Section 4).

A worked example is set out below in Table 12.

Table 12 *z* Score and E_n Score Calculation for Sample S1 Result Reported by Laboratory 1

Participant Result (% base (m/m))	Assigned Value (% base (m/m))	Target SD	*z* Score	E_n Score
76.58 ± 4.59	76.9 ± 0.9	3% as PCV, or: 0.03 × 76.9 = 2.3% base (m/m)	*z* Score = $\frac{76.58-76.9}{2.3}$ = -0.14	E_n Score = $\frac{76.58-76.9}{\sqrt{4.59^2+0.9^2}}$ = -0.07

APPENDIX 2 – ACRONYMS AND ABBREVIATIONS

ANAB	ANSI (American National Standards Institute) National Accreditation Board
ASCLD/LAB	American Society of Crime Laboratory Directors/Laboratory Accreditation Board
CITAC	Cooperation on International Traceability in Analytical Chemistry
CRM	Certified Reference Material
CV	Coefficient of Variation
DAD	Diode Array Detector
FID	Flame Ionisation Detector
GAG	General Accreditation Guidance (NATA)
GC	Gas Chromatography
GUM	Guide to the expression of Uncertainty in Measurement
HPLC	High Performance Liquid Chromatography
IEC	International Electrotechnical Commission
ISO	International Organization for Standardization
M	Median
Max.	Maximum value in a set of results
Min.	Minimum value in a set of results
MS	Mass Spectrometry
MS/MS	Tandem Mass Spectrometry
MU	Measurement Uncertainty
N	Number of numeric results
NATA	National Association of Testing Authorities, Australia
NMI	National Measurement Institute, Australia
NR	Not Reported
PCV	Performance Coefficient of Variation
PDA	Photodiode Array
PT	Proficiency Test
QNMR	Quantitative Nuclear Magnetic Resonance
RA	Robust Average
RM	Reference Material
SD	Standard Deviation
SI	International System of Units
SS	Spiked Samples
U	Expanded Uncertainty
UNODC	United Nations Office on Drugs and Crime
UPLC	Ultra Performance Liquid Chromatography
UV/Vis	Ultraviolet/Visible detector

END OF REPORT