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# Measurement Uncertainty

# Introduction

As an ISO 17025 compliant, NATA-accredited facility, the Public Health Laboratory is required to inform customers of our ability to provide Measurement Uncertainty (MU) estimations with results. MU will be provided when relevant to the validity of a result, when the MU may affect compliance to a specification limit and when requested by customers.

MU is only applicable to quantitative results, so qualitative tests, such as Presence/Absence for *Salmonella* and *Listeria*, do not require MU. Similarly, quantitative results less than the detection limit (e.g. *E. coli* < 1 cfu/100 mL) do not require a statement of MU. Sample types with regulatory specification limits include Heterotrophic Colony Count (HCC) in swimming pools (HCC < 100 cfu/mL), cooling towers (HCC < 100 000 cfu/mL) and enumeration of *L. monocytogenes* (< 100 cfu/g).

This notice is provided to help you decide if you want this service. References are provided at the end for more information, but the following provides a 'plain English' account (where possible!).

#### What is Measurement Uncertainty?

All measurements have an inherent uncertainty due to small unavoidable variations in the sample, the media used in testing, the person doing the analysis and all other conditions at the time of analysis. These variations persist despite efforts to standardise measurement conditions, and cannot be eliminated entirely. As a result, repeated tests done on the same sample will give a set of results, each slightly different, grouped around the 'true' value.

Measurement Uncertainty is the estimate of the range around a measured value in which the 'true' value occurs for a given level of confidence (the Confidence Interval).

#### What is a Confidence Interval?

The Confidence Interval (CI) is the range likely to contain the measurement and the recommended reporting CI is 95 per cent. For example, if a test for HCC is reported as 85 cfu/mL [66, 110] at the 95% CI, this means that saying that the 'true' colony count in the sample is between 66 cfu/mL and 110 cfu/mL, will be true 95 per cent of the time. To put it another way, there is only a 1 in 20 (five per cent) chance of the 'true' value being outside this range. Higher CI (e.g. 99 per cent) can be calculated but have a wider range of possible values.

### How is MU Reported?

Measurement Uncertainty may be reported in several ways. For example, a result of 100 cfu/mL has been obtained for a water test. The laboratory MU is log 0.11 for this test (95% Cl), the result may be reported as:

- Base-10 logarithm e.g. 2 log ± 0.11 log at the 95% Cl
- Standard Deviation e.g. 2 log [1.89, 2.11] at the 95% CI
- Natural Values e.g. 100 cfu/mL [78,129] at the 95% CI
- Percentage e.g. 100 cfu/mL [100-22%, 100+ 29%] at the 95% CI

#### How is MU Reported?

Graphically, the result and CI may be shown like this:

129	-	Т		
100	-	Ý		
78	-			

### **Do You Need MU Estimation?**

In most cases the MU estimate will not significantly affect your results as the possible dispersion of the 'true' value is not great. However, if your test is required for compliance to a specification then MU may be important if the result is near to the specification.

For example, pools must have a HCC of < 100 cfu/mL. If the laboratory MU value is log 0.11 for HCC, the corresponding CI for the results is [78, 129], meaning that the 'true' value will fall between between 78 and 129 cfu/ml. This allows the **possibility** that a failing result of 110 cfu/mL may have had a 'true' value below 100 cfu/mL, **OR** that a passing result of 95 cfu/mL may have had a 'true' value above 100 cfu/mL. A result within the MU of the specification could be regarded as marginal and re-testing may be advisable.

The diagram below illustrates the possible effect of MU. It shows four test results, A-D, for a test with a specification limit whose value is shown by the horizontal line. Result A is a clear fail while result D is a clear pass, as the results, **and CI**, are above and below the limit respectively.

Result B is a fail but is debatable as the limit is within the uncertainty for the test. Similarly, result C is a pass but the uncertainty of the test means the 'true' value could be above the limit and the result could be contested. These results may be judged inconclusive, and may require retesting to obtain a definitive result.



#### MU and the Public Health Laboratory

The Public Health Laboratory has calculated MU for all NATA-accredited, quantitative water and food methods. Non-quantitative results, such as Presence/Absence tests for *Salmonella* and *Listeria*, do not require MU. Also, quantitative results less than the detection limit (e.g. *E. coli* <1 cfu/mL) do not require a statement of MU.

Annex A shows the calculated Expanded Uncertainty (U) for each method – U is a log value which is applied to the logarithm of the test result to generate the CI, as shown below.

Example:	A sample HCC result is	85 cfu/mL (=1.93 log), U = 0.11 log at 95% Cl
	Result with uncertainty is:	1.93 log cfu/mL <u>[1.93 – 0.11, 1.93 + 0.11]</u>
	Therefore, the result is	1.93 log cfu/mL [1.82, 2.04]
	In natural values, the result is:	85 cfu/mL [66, 110]

Customers requiring MU will receive a separate report in addition to our standard report. The additional report will attract an administrative fee of \$20 per report.

The following reply slip should be returned only if you wish to have MU calculated and reported on your results. You can request MU at any time by emailing the laboratory (<a href="mailto:publichealth.lab@health.tas.gov.au">publichealth.tas.gov.au</a>) or by noting it under 'Comments' on your sample request forms.

#### **References:**

- ISO, 2005. General requirements for the competence of testing and calibration laboratories. ISO/TS 17025. International Organisation for Standardization.
- ISO, 2006. Microbiology of food and animal feeding stuffs Guidelines for the estimation of measurement uncertainty for quantitative determinations. ISO/TS 19036. International Organisation for Standardization.
- ISO, 2009. ISO/TS 19036 Amendment 1: Measurement uncertainty for low counts.
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#### **Request for Measurement Uncertainty**

I request that MU be calculated for: All Work / Specified Work.

I acknowledge that an administration fee of \$20 will be charged for a separate MU report.

I prefer MU reported as: Standard Deviation (logarithm values) / Range (natural values).

Customer: .....

## Annex A

## PHL MU for NATA-accredited Methods

Measurement Uncertainty calculated for				
Water methods used in the Public Health Laboratory				
Method	Standard	Expanded Uncertainty, U	n	
Legionella – Spread Plate	AS/NZS 3896:2017	0.264	64	
HCC - YEA (35°C)	AS/NZS4276.3.1:2007	0.100	19	
HCC - YEA (21°C)	AS/NZS4276.3.1:2007	0.109	17	
HCC (Biocides) - R2A (35°C)	AS/NZS4276.3.2:2003	0.112	27	
HCC (Biocides) - R2A (21°C)	AS/NZS4276.3.2:2003	0.116	21	
HCC - HPC for Quanti-tray	PHL 11.3 (Non-AS method)	0.127	31	
Coliforms - MF	AS/NZS 4276.5:2007	0.155	28	
Thermotolerant Coliforms - MF	AS/NZS 4276.7:2007	0.131	20	
E. coli - MF	AS/NZS 4276.7:2007	0.131	20	
Enterococci - MF	AS/NZS 4276.9:2007	0.070	23	
Pseudomonas aeruginosa - MF	AS/NZS 4276.13:2008	0.101	13	
Coliforms - Colilert	AS 4276.21:2005	0.164	31	
E. coli - Colilert	AS 4276.21:2005	0.198	24	
Enterococci - Enterolert	PHL 8.4 (Non-AS method)	0.149	26	
Coliforms - MPN	AS/NZS 4276.6:2007	0.510	15	
Thermotolerant Coliforms - MPN	AS/NZS 4276.6:2007	0.397	16	
E. coli - MPN	AS/NZS 4276.6:2007	0.397	16	

MF: Membrane Filtration MPN: Most Probable Number

Measurement Uncertainty calculated for					
Food methods used in the Public Health Laboratory 01/07/2022					
Method	Standard	Expanded Uncertainty, U	n		
SPC - Pour Plate	AS5013.1:2004	0.181	48		
Yeasts - Spread Plate	AS5013.29:2009	0.369	19		
Moulds - Spread Plate	AS5013.29:2009	0.499	25		
B. cereus - Spread Plate	AS5013.2:2007	0.362	30		
Coliforms - MPN	AS5013.3:2004	0.766	39		
Coliforms - Spread Plate	AS5013.4:2004	0.369	34		
E. coli - MPN	AS 5013.15:2004	0.727	31		
E.coli - Spread Plate	AS5013.19.1:2012	0.288	43		
C. perfringens - Spread	AS5013.16:2006	0.402	11		
S. aureus - Spread Plate (BPA)	AS5013.12.1:2004	0.311	22		
S. aureus - Spread Plate (RPF)	AS5013.12.2:2004	0.219	17		
S. aureus - MPN	AS5013.12.3:2004	0.558	13		
L. monocytogenes - Spread Plate	AS5013.24.2:2009	0.346	25		
V. parahaemolyticus - MPN	AS5013.18:2010	0.627	18		

MPN: Most probable Number