

**REPORT NO. 503**

**ALGAE ROUND 9**

**PROFICIENCY TESTING PROGRAM**

**MARCH 2006**

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## CONTENTS

	Page(s)
1. Foreword	1
2. Statistical Design of the Program	1
3. Features of the Program	1
4. Format of Appendices	2
5. Outlier Results	2 - 3
6. PTA and Technical Advisers' Comments	3 - 8
7. References	8
TABLE A Valid Benchmark Taxa Present In Samples A, B & C	4
TABLE B Extreme Results – Identification	8
TABLE C Extreme Results – Enumeration	8

### APPENDIX A

Appendix Format	A(i) – A(ii)
<u>All Results</u>	
Identification	A1 – A12
Enumeration	A13 – A16
<u>Z-Scores and Summary Statistics</u>	
Organism 1 – Sample Pair B & C	A17 – A19
Organism 1 – Sample A	A20 – A21
Organism 2 – Sample Pair B & C	A22 – A24
Organism 2 – Sample A	A25 – A26
Organism 3 – Sample Pair B & C	A27 – A29
Organism 3 – Sample A	A30 – A31
Organism 4 – Sample Pair B & C	A32 – A34
Organism 4 – Sample A	A35 – A36

### APPENDIX B

Sample Preparation and Distribution	B1
Homogeneity and Stability Testing	B1 – B2

### APPENDIX C

Instructions to Participants and Results Sheets	C1 – C6
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### APPENDIX D

TABLE D Z-Score Calculation Parameters	D1
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## 1. **FOREWORD**

This report summarises the results of a proficiency testing program covering the identification and enumeration of selected algae.

NATA's Proficiency Testing Group conducted the exercise in October 2005 as part of its laboratory accreditation activities. Note that from 1 January 2006 the delivery of proficiency testing services was transferred from NATA to a new, wholly owned subsidiary called Proficiency Testing Australia (PTA).

The main aim of the program was to assess laboratories' ability to competently perform the tests examined.

## 2. **STATISTICAL DESIGN OF THE PROGRAM**

Each participating laboratory was provided with three samples of water, each containing a range of algal and cyanobacterial genera, labelled Sample A, Sample B and Sample C. In all samples, certain genera were clearly dominant in cell number.

Robust statistical procedures were used to generate the z-scores and summary statistics for each sample – number of results, median, normalised interquartile range, minimum, maximum and range.

## 3. **FEATURES OF THE PROGRAM**

- (a) A total of 24 laboratories received samples. 22 laboratories submitted results by the due date. Lab code 24 reported 2 sets of results (24A & 24B).
- (b) Participants were supplied with three samples, preserved in Lugol's solution, of approximately 20mL in dark glass bottles. Samples B & C were duplicates. Sample A contained the same algal composition, but differed predominantly in the cell concentration of the cyanobacterium *Arthrospira*.
- (c) Prior to sample distribution, 8 randomly selected samples of each matrix (B/C & A) were analysed for homogeneity. Based on the results of this testing it was concluded that the samples were sufficiently homogenous (see Appendix B), and those results identified as outliers in this report cannot be attributed to significant sample variability.
- (d) The results for each test as reported by participants are presented in Appendix A, together with summary statistics, calculated z-scores and graphical presentations of the data.
- (e) Participating laboratories were requested to perform the tests according to Instructions to Participants, and to record their results on the accompanying Results Sheets, all of which were distributed to laboratories with the test samples.

Copies of the Instructions to Participants and Results sheets are given in Appendix C of this report.

- (f) Each laboratory was randomly allocated a unique code number for the program to ensure confidentiality of results. Reference to each laboratory in this report is by its code number.

#### 4. **FORMAT OF APPENDICES**

##### **Appendix A**

**Identification:** The dominant genera identified as present by each laboratory are tabulated.

**Enumeration:** For sample pair B & C and sample A, the following is given for each of the four genera enumerated.

- (i) The results of the enumeration (in cells/mL) as reported by participating laboratories, and the corresponding transformed ( $\log_{10}$ ) values.
- (ii) The calculated “between-laboratories z-score” and “within-laboratories z-score” for each result for sample pair B & C and robust z-scores for each result for sample A, with outliers identified.
- (iii) A list of summary statistics.
- (iv) A bar-chart of the ordered between-laboratories z-scores (sample pair B & C).
- (v) A bar chart of the ordered within-laboratories z-score (sample pair B & C).
- (vi) A bar-chart of the ordered robust z-scores (sample A).
- (vii) A Youden Diagram (sample pair B & C only).

##### **Appendix B**

- (i) Sample Preparation and Distribution.
- (ii) Homogeneity Testing.

##### **Appendix C**

- (i) Instructions to Participants.
- (ii) Results Sheets.

##### **Appendix D**

- (i) Z-Score Calculation Parameters (Table D).

#### 5. **OUTLIER RESULTS**

##### **Identification**

Any genera reported other than the listed benchmark genera are considered false (extreme) results and are marked in Appendix A by the symbol ‡. Participants who identified less than the required number of algal genera in each group are also considered extreme results, and these are marked by the symbol †.

### Enumeration

In order to achieve the program's aim of assessing laboratories' testing performance, a robust statistical approach, which uses z-scores to assess participants' performance, has been utilised. The z-score is a measure of how far the results are from the consensus value - a normalised value which gives a "score" to each result relative to the other results in the group. Therefore a z-score close to zero means that the result agrees well with those from other laboratories. An outlier is any result which has an absolute z-score value greater than three and is marked in Appendix A by the symbol §. Any results deemed 'mis-identifications' are marked by \* and were not included in the analysis. These are counted as extreme results.

## 6. PTA AND TECHNICAL ADVISERS' COMMENTS

### General Comments/Overall Performance

The test samples used in Round 9 were prepared from naturally occurring material that contained several algal genera from different classification groups (refer Table A). These samples were considered representative of those that would normally be encountered by an analyst in routine work. Participants were asked to identify and enumerate genera and species from various nominated groups that were commonly present or dominant in the test samples. These included green algae, filamentous green alga, motile algae and cyanobacteria. This required a knowledge of the major algal groups and their morphological characteristics and an ability to make a judgement of the relative abundance (by cell number) of all algae in the sample. The test samples were not controlled in terms of the total number of genera that were present and for some classification groups, the number of valid identifications exceeded the number required in the instructions to participants.

As in previous rounds, participants were invited to choose their own method for enumeration, rather than adhere strictly to a prescribed method. Individual judgements could be made on suitable magnification, type of counting chamber, the proportion of chamber to be counted, the number of cells or filaments to count and the appropriate methods to estimate cells in colonies or trichomes.

### Benchmark and Consensus Results

Benchmark results were used for the identification component of the proficiency test and were determined by Technical Advisers at the time of sample preparation. These may include alternative identifications that have close taxonomic affinity to the benchmark result.

The "instructions to participants" requested identification of algae or cyanobacteria that were present in each sample, fitting the following criteria:

1. Two (2) genera of green algae that represent the order Chlorococcales; one of the Family Oocystaceae and the other of the Family Dictyosphaeraceae.
2. The dominant green filamentous algal genus.
3. Two (2) dominant genera of motile algae representing the order Volvocales.
4. Seven (7) genera of cyanobacteria representing the orders Nostocales and Oscillatoriales.

The benchmark identifications are listed in Table A.

For each sample, an identification and an estimate of cell abundance (reported as cells/mL) was required for:

1. The abundant green alga representing the Order Chlorococcales; Family Oocystaceae
2. The dominant (by cell number) filamentous green algal genus.

3. The dominant (by cell number) unicellular motile genus representing the Order Volvocales.
4. The dominant (by cell number) cyanobacterium representing the Order Oscillatoriales.

For the purposes of testing enumeration proficiency, the consensus value was derived from the median result of all participants that are deemed to have enumerated the same nominated organism, irrespective of benchmark identification.

A total of 6 participants correctly identified genera in the three samples, based on the benchmark results, while 16 of the 23 participants did not report any outliers in the enumeration of the 4 requested genera in each sample, based upon variability about the consensus median result.

**TABLE A. VALID BENCHMARK TAXA PRESENT IN SAMPLES A, B & C**

<b>Green Algae (Chlorococcales: Oocystaceae, Dictyosphaeraceae)</b>	<b>Filamentous Green Alga</b>	<b>Motile Algae (Volvocales)</b>	<b>Cyanobacteria (Nostocales, Oscillatoriales) (any 7 of the following)</b>
<i>Oocystis</i>	<i>Mougeotia</i>	<i>Volvox</i>	<i>Anabaena</i>
<i>Botryococcus</i>		<i>Chlamydomonas</i>	<i>Anabaenopsis</i>
			<i>Cylindrospermopsis</i>
			<i>Aphanizomenon</i>
			<i>Cylindrospermum</i>
			<i>Planktolyngbya</i>
			<i>Arthrospira</i>
			<i>Trichodesmium</i>
			<i>Oscillatoria</i>
			<i>Limnothrix</i>

### Identification

The most common green algae genera (Chlorococcales:Oocystaceae, Dictyosphaeraceae) present in all three samples were *Oocystis* and *Botryococcus* and 18 participants correctly reported both genera for all three samples. Lab codes 6, 23, 24A & 24B incorrectly reported *Dictyosphaerium* in all three samples, while lab code 22 incorrectly reported *Lobocystis* in all three samples. All three test samples contained another colonial green alga in significant abundance, which may have been mis-identified by some participants as *Dictyosphaerium* and *Lobocystis*. The classification of this alga was referred to a recognised expert in phytoplankton taxonomy, and tentatively identified as *Rhombocystis* (Family Oocystaceae).

The dominant filamentous green alga was *Mougeotia* and 21 out of 23 participants reported correctly for all three samples. Lab code 6 incorrectly reported *Hormidium* for each of the three samples (A, B & C), while Lab code 11 incorrectly reported *Chlorhormidium* in all three samples. *Hormidium* and *Chlorhormidium* are regarded as synonyms and can be distinguished by the presence of parietal chloroplasts in contrast to axial chloroplasts of *Mougeotia*. Lab code 13 identified the filamentous green alga as either *Mougeotia* or *Oedogonium*, while lab code 20 nominated either *Mougeotia* or *Debarya*. While *Debarya* has an axial chloroplast and resembles species of *Mougeotia*, *Oedogonium* has a parietal network and is characterised by ringlike scars on the cell wall adjacent the cross-walls. Neither of these participants' results were deemed to be extreme.

The most common motile algal genera (Volvocales) in the three samples were *Volvox* and *Chlamydomonas* and 10 out of 23 participants reported correctly for the three samples. Lab codes 1, 6, 8, 15, 21, 24A & 24B incorrectly reported *Gonium* in all three samples. Lab codes 4, 7 & 19 incorrectly reported *Pandorina* in all three samples. Lab code 1 incorrectly reported *Stenophora* for each sample and lab code 22 incorrectly reported *Lobomonas* in each sample. Lab code 9 only reported *Chlamydomonas* and lab code 13 did not report any genus in this category. Mis-identification of *Volvox* by several participants can be attributed to the almost complete disaggregation of colonies soon after sample dispatch to single cells or small groups of cells in a flat plane. At the time of sample preparation, large fragments of *Volvox* colonies were still present in all samples.

The accepted results for the cyanobacterial genus (Nostocales, Oscillatoriales) in the three samples were *Anabaena*, *Anabaenopsis*, *Cylindrospermopsis*, *Aphanizomenon*, *Cylindrospermum*, *Planktolyngbya*, *Arthrospira*, *Trichodesmium*, *Oscillatoria* and *Limnothrix*. A total of 10 participants correctly identified seven of these genera in sample A, 13 participants correctly identified seven of these genera in sample B and 11 participants correctly identified seven of these genera in sample C.

*Pseudanabaena* was incorrectly reported by lab codes 1, 2, 6, 7, 8, 16, 21, 24A, 24B & 25 in sample A; lab codes 6, 7, 8, 16, 24A, 24B & 25 in sample B; and lab codes 1, 2, 6, 7, 8, 16, 21, 24A, 24B & 25 in sample C. These participants most likely misidentified trichomes of *Cylindrospermum* as *Pseudanabaena*, but thorough inspection of the sample would have revealed the characteristic presence of terminal heterocysts in a small proportion of trichomes. *Spirulina* was incorrectly reported by lab code 12 in sample A and lab code 1 in sample B. The genera *Spirulina* and *Arthrospira* are often interchangeable in some taxonomic texts, but Lab codes 12 and 1 identified both and *Spirulina* was therefore deemed an outlier. Lab code 12 incorrectly reported *Tychonema* in sample C and this genus is most likely a mis-identification of *Oscillatoria*. The presence of *Pseudanabaena*, *Spirulina* and *Tychonema* in the test samples can not be rejected absolutely, but they were not observed by the Technical Advisors at the time of sample preparation.

Lab codes 11 & 19 identified several species of *Anabaena* and only identified a total of five cyanobacterial genera, rather than the required seven genera. Lab code 11 only identified one genus of cyanobacteria in Sample C.

### Enumeration

Samples B and C were identical and so results were pooled for analysis. Single robust z-scores were calculated for sample A.

Statistical assessment of cell abundance estimates was reserved for the following genera and species. Counts of taxa other than the benchmark identification have been included where deemed to be the identical but otherwise identified organism.

- Organism 1 (green alga; Order Chlorococcales; Family Oocystaceae) – *Oocystis*
- Organism 2 (dominant filamentous green algal genus) – *Mougeotia*, *Hormidium* sp., *Chlorhormidium*
- Organism 3 (dominant unicellular motile algal genus; Order Volvocales) – *Chlamydomonas*
- Organism 4 (dominant cyanobacterial genus; Order Oscillatoriales) - *Arthrospira*

Results reported for genera other than these were not analysed and are considered extreme results.

### **Organism 1 (green alga; Order Chlorococcales; Family Oocystaceae)**

All participants correctly chose *Oocystis* for enumeration in all three samples. Two participants (lab codes 9 & 13) did not report any results.

For sample pair B & C, two within-laboratory outliers were reported by lab codes 2 & 3. For sample A, one outlier was reported by lab code 3. This outlier was a significant over-estimation compared with the consensus median and may have resulted from mis-identification and enumeration of another green colonial alga (cf *Rhombocystis*) that was abundant in all samples. Overall, a broad range of results was reported (5,451 to 43,644 cells/mL in Sample A and 6,500 to 40,760 cells/mL in Sample B/C).

### **Organism 2 (dominant filamentous green algal genus)**

A total of 19 of 23 participants correctly chose *Mougeotia* for enumeration in each of the three samples. Results for *Hormidium* (lab code 6) and *Chlorhormidium* (lab code 11) were also included in the analysis. Lab codes 9 & 13 did not report any results. The enumeration for this organism was performed well resulting in no outliers being reported. However, a considerable range in results was reported across all participants (34,429 to 87,696 cells/mL in Sample A and 32,798 to 94,667 cells/mL in Sample B/C).

### **Organism 3 (dominant unicellular motile algal genus; Order Volvocales)**

Only 13 of the 23 participants correctly chose *Chlamydomonas* for enumeration. Lab codes 2, 5, 11, 16, 20 & 25 enumerated *Volvox*, lab code 1 enumerated *Stenophora*, and lab code 22 enumerated *Lobomonus*. These mis-identifications have been classed as extreme results and so not included in the analysis. Lab codes 9 & 13 did not report any results.

For sample pair B & C, one between-laboratories outlier was reported by lab code 4. Lab code 4 also reported a robust z-score outlier for sample A. Both outliers were significant over-estimations (76,000-101,250 cells/mL) and are most likely attributable to enumeration of other single celled motile genera, including the disaggregated single cells of *Volvox*. Discounting the outliers of lab code 4, the results ranged from 2,275 to 7,560 cells/mL in Sample A and from 1,880 to 7,200 cells/mL in Sample B/C.

### **Organism 4 (dominant cyanobacterial genus; Order Oscillatoriales)**

All 23 participants reported results for organism 4, and all participants correctly chose *Arthrospira* for enumeration.

For sample pair B & C, three within-laboratory outliers were reported by lab codes 2, 7 & 15. For sample A, two robust z-score outliers were reported by lab codes 1 & 6 and these were significant under-estimations compared with the consensus median result. The robust outlier result reported by Lab Code 1 can be attributed to a very low estimate of cells per trichome, while both outliers were based on enumeration of a relatively small number of trichomes and/or cells. Overall, the results ranged from 101,881 to 590,000 cells/mL in Sample A and from 86,834 to 512,900 cells/mL in Sample B/C.

### **Variation Between Methods and Possible Sources of Error**

The majority of participants (16 of 23) chose to use a Sedgewick Rafter counting chamber for Round 9, and 4 of these provided a calibration factor for the nominal chamber volume of 1 mL. Another 5 participants chose to use a Lund Cell and 4 of these provided the measured volume of sample. One participant used a Utermohl chamber, while another used a custom built chamber of 3 mL nominal volume. The latter participant described a concentration technique using centrifugation, which is not generally regarded as a suitable technique for phytoplankton. The choice of counting chamber did not appear to have any bearing on outlier results in enumeration.



Magnification for identification and enumeration of algal taxa ranged from 100x to 1000x oil, although magnification of 100x was only used to enumerate trichomes of *Mougeotia* and *Arthrospira*. Lab codes 3, 11, 16, 20, 21 and 25 used 1000x oil to measure the length of *Arthrospira* cells, or alternatively to estimate the number of cells of *Arthrospira* in a defined segment of a trichome. A magnification of 200x was used by some participants (Lab codes 5, 8, 12, 14 and 19) for the same purpose. The choice of magnification for enumeration of *Arthrospira* cells did not appear to have any bearing on outlier results.

The majority of participants reported their results to 4, 5 or 6 significant figures, depending on the magnitude of results, suggesting that no rounding off was used for allowance of the error associated with enumeration estimates. Rounded off results were reported by Lab codes 13, 19, 21, 24A (2 significant figures), by Lab codes 8, 11, 16 and 25 (3 significant figures) and by Lab code 22 (4 significant figures).

The majority of participants chose to enumerate the colonial green alga *Oocystis* as cells only, while up to 6 participants chose to estimate cells per colony. Estimates ranged from 3.9 to 9.0 cells/colony and outlier results reported by Lab code 3 may be attributable to a high estimate. The total number of *Oocystis* cells enumerated ranged from 58 to 1082 and the number of colonies from 22 to 113. The majority of participants also chose to enumerate the filamentous green alga *Mougeotia* as cells only, while up to 8 participants estimated cells per filament. Estimates ranged from 2.6 to 6.0 cells/filament, but no outlier results were reported for *Mougeotia*. The total number of *Mougeotia* cells counted ranged from 103 to 4,260 and the number of filaments from 38 to 667.

*Chlamydomonas* was counted as single cells and the total number of cells counted in any one sample ranged from 18 to 585. The one outlier result (Lab code 4) enumerated 300-400 cells.

The methods used to enumerate *Arthrospira* varied considerably between participants, as might be expected given the sinusoidal shape of the trichomes and the difficulty in visualising individual cells. Up to 5 participants enumerated cells only and the total number of cells enumerated ranged from 611 (code 24B) to 7,970 (code 25). The robust outlier result reported by Lab code 6 for *Arthrospira* may be partly attributable to a low number of cells enumerated in the 3 samples (924-1332), although no outlier was reported by 24B for an even lower cell count. Estimates of cell per trichome were reported by the majority of participants and ranged from 31.8 (code 1) to 180.2 (code 11), with a median result for all participants of 87 cells/trichome. The robust outlier result reported by Lab code 1 can be attributed to a very low estimate of cells per trichome, whereas the highest estimate reported by 11 was not an outlier.

Up to 7 participants estimated cells of *Arthrospira* in a defined segment of the trichome (ie from crest to crest or from crest to trough or from a measured length) and then estimated the number of segments per trichome. None of these participants recorded a between laboratories or robust outlier. Lab code 11 estimated the mean length of trichomes by measuring from end to end and multiplying by a factor of 2 to account for the regular oscillation (crest and troughs) of trichomes. The mean trichome length was then divided by the mean cell length to estimate the mean number of cells per trichome. A variation of this method was used by Lab code 20, by estimating the mean length of trichome segments (crest to trough) and dividing by mean cell length. Lab code 22 estimated the number of cells between crest and trough by dividing spiral breadth by cell length. Although this method has obvious flaws, no outlier result was reported. The total number of *Arthrospira* trichomes that were enumerated in any one sample ranged from 16 to 774.

### General Comments by Participants

Lab codes 1, 6, 7 and 15 commented that the cells of some genera were very degraded, particularly the Volvocales, which made identification very difficult.

Lab codes 9 and 12 used a statistical software program for cell enumeration to provide an indication of when sufficient cells were counted to provide results of satisfactory precision.

## 7. REFERENCE

[1] "Guide to Proficiency Testing Australia" – January 2006

**TABLE B: EXTREME RESULTS - IDENTIFICATION**  
(by laboratory code number)

Classification Group	Sample A	Sample B	Sample C
Green Algae (Chlorococcales: Oocystaceae, Dictyosphaeraceae)	6, 22, 23, 24A, 24B	6, 22, 23, 24A, 24B	6, 22, 23, 24A, 24B
Filamentous Green Alga	6, 11	6, 11	6, 11
Motile Algae (Volvocales)	1, 4, 6, 7, 8, 9, 15, 19, 21, 22, 24A, 24B	1, 4, 6, 7, 8, 9, 15, 19, 21, 22, 24A, 24B	1, 4, 6, 7, 8, 9, 15, 19, 21, 22, 24A, 24B
Cyanobacteria (Nostocales, Oscillatoriales)	1, 2, 6, 7, 8, 11, 12, 16, 19, 21, 24A, 24B, 25	1, 6, 7, 8, 11, 16, 19, 24A, 24B, 25	1, 2, 6, 7, 8, 11, 19, 16, 21, 24A, 24B, 25

**TABLE C: EXTREME RESULTS – ENUMERATION**  
(by laboratory code number)

Classification Group	Mis-identification	Between-Laboratories Outlier	Within-Laboratory Outlier	Robust Outlier
ORGANISM 1 (green alga; Order Chlorococcales; Family Oocystaceae)	-	-	2, 3	3
ORGANISM 2 (dominant filamentous green algal genus)	-	-	-	-
ORGANISM 3 (dominant unicellular motile algal genus; Order Volvocales)	1, 2, 5, 11, 16, 20, 22, 25	4	-	4
ORGANISM 4 (dominant cyanobacterial genus; Order Oscillatoriales)	-	-	2, 7, 15	1, 6

# APPENDIX A

## Summary of Results

Appendix	A(i) - (ii)	Appendix Format
	<u>All Results</u>	
	A1 – A12	Identification
	A13 – A16	Enumeration
	<u>Z-Scores and Summary Results</u>	
	A17 – A19	Organism 1 – Sample Pair B & C
	A20 – A21	Organism 1 – Sample A
	A22 – A24	Organism 2 – Sample Pair B & C
	A25 – A26	Organism 2 – Sample A
	A27 – A29	Organism 3 – Sample Pair B & C
	A30 – A31	Organism 3 – Sample A
	A32 – A34	Organism 4 – Sample Pair B & C
	A35 – A36	Organism 4 - Sample A

## APPENDIX FORMAT

This appendix consists of all the results submitted by the participating laboratories for both the Identification and Enumeration components of the program, and the calculated z-scores and summary statistics for the enumeration component.

### (a) Table of Results and Z-Scores

Each of these tables contains the results returned by each laboratory, including the code number used for each participating laboratory, and the laboratory z-scores calculated for each laboratory's result.

Outliers are identified in the table by a marker (§) next to the relevant z-score. A positive between-laboratories or robust z-score indicates that the results are higher than the consensus median, while a negative between-laboratories or robust z-score indicates results are lower than the consensus median. Please see reference [1] for details on how these z-scores are calculated.

### (b) Summary Statistics

The list of summary statistics appears at the bottom of the table of results and consists of:

- (i) the number of results for that test/sample (*No. of Results*);
- (ii) the median of laboratories' results - i.e. the middle value (*Median*);
- (iii) the normalised interquartile range of the results (*Normalised IQR*) - the interquartile range times 0.7413;
- (iv) the robust coefficient of variation, expressed as a percentage (*Robust CV*) - i.e.  $100 \times \text{Normalised IQR} / \text{Median}$ ;
- (v) the minimum and maximum laboratory results; and
- (vi) the range (*Maximum - Minimum*).

The median is a measure of the centre of the data and replaces the previously used 'consensus mean'. The Normalised IQR is a measure of the spread of the results and replaces the 'standard deviation'.

Multiplying the IQR by the factor (0.7413) allows for comparison between the old and new types of coefficients of variation. Please see reference [1] for further details on these robust summary statistics.

(c) Ordered Z-Score Charts

On these charts each laboratory's z-score is shown, in order of magnitude, and is marked with its code number. From these each laboratory can readily compare its performance relative to the other laboratories.

The y-axis of these charts has been limited to +3 and -3, so very large or small (negative) z-scores appear to extend beyond the chart. The outliers are clearly identifiable as the laboratories whose "bar" extends beyond these "cutoff" lines.

(d) Youden Diagrams

Youden two-sample diagrams are presented to highlight laboratory systematic differences. They are based on a plot of each laboratory's pair of results, sample two versus sample one, represented by a black spot.

These diagrams also feature an approximate 95% confidence ellipse for the bivariate analysis of the results, and dashed lines which mark the median value for each of the samples.

All points which lie outside the ellipse are labelled with the laboratory's code number. Note however that these points may not correspond with those identified as outliers. This is because the outlier criteria ( $|z| > 3$ ) has a confidence level of approximately 99%, whereas the ellipse is an approximate 95% confidence region.

So, the points outside the ellipse on the Youden diagram will be those with z-scores greater than 2 or less than -2. The laboratories which are outside the ellipse but have not been identified as extreme (i.e. have  $2 < |z| < 3$ ) are encouraged to "take a close look at" their results.

As a guide to the interpretation of these diagrams:

- (i) laboratories with significant systematic error components (i.e. between-laboratories variation) will be outside the ellipse in either the upper right hand quadrant (as formed by the median lines) or the lower left hand quadrant, i.e. inordinately high or low results for both samples; and
- (ii) laboratories with random error components (i.e. within-laboratory variation) significantly more variable than other participants will be outside the ellipse and (usually) in either the upper left or lower right quadrants, i.e. an inordinately high result for one sample and low for the other.

Further details of the construction and interpretation of these diagrams is given in reference [1]. Please also refer to this document for a glossary of terms.

**Identification - All Results**

Lab Code	SAMPLE A			
	Green Algae (Chlorococcales: Oocystaceae, Dictyosphaeraceae )	Filamentous Green Alga	Motile Algae (Volvocales)	Cyanobacteria (Nostocales, Oscillatoriales)
1	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Gonium</i> ‡ <i>Stenophora</i> ‡	<i>Anabaena</i> <i>Planktolyngbya</i> <i>Arthrospira</i> <i>Aphanizomenon</i> <i>Cylindrospermopsis</i> <i>Anabaenopsis</i> <i>Pseudanabaena</i> ‡
2	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Volvox</i> <i>Chlamydomonas</i>	<i>Anabaena</i> <i>Aphanizomenon</i> <i>Cylindrospermopsis</i> <i>Cylindrospermum</i> <i>Arthrospira</i> <i>Pseudanabaena</i> ‡ <i>Planktolyngbya</i>
3	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Volvox</i> <i>Chlamydomonas</i>	<i>Anabaena</i> <i>Arthrospira</i> <i>Cylindrospermopsis</i> <i>Anabaenopsis</i> <i>Aphanizomenon</i> <i>Planktolyngbya</i> <i>Cylindrospermum</i>
4	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Chlamydomonas</i> <i>Pandorina</i> ‡	<i>Arthrospira</i> <i>Planktolyngbya</i> <i>Cylindrospermum</i> <i>Cylindrospermopsis</i> <i>raciborskii</i> <i>Anabaena</i> <i>Anabaenopsis</i> <i>Aphanizomenon</i>
5	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Volvox</i> <i>Chlamydomonas</i>	<i>Anabaena</i> <i>Anabaenopsis</i> <i>Cylindrospermopsis</i> <i>Cylindrospermum</i> <i>Aphanizomenon</i> <i>Arthrospira</i> <i>Planktolyngbya</i>
6	<i>Oocystis</i> <i>Dictyosphaerium</i> sp. ‡	<i>Hormidium</i> sp. ‡	<i>Gonium</i> sp. ‡ <i>Chlamydomonas</i>	<i>Anabaena</i> <i>Arthrospira</i> <i>Cylindrospermopsis</i> <i>Aphanizomenon</i> <i>Anabaenopsis</i> <i>Pseudanabaena</i> ‡ <i>Trichodesmium</i>
7	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Chlamydomonas</i> <i>Pandorina</i> ‡	<i>Arthrospira</i> <i>Anabaena</i> <i>Cylindrospermopsis</i> <i>Anabaenopsis</i> <i>Aphanizomenon</i> <i>Planktolyngbya</i> <i>Pseudanabaena</i> ‡

‡ Denotes a false result.

Lab Code	SAMPLE A			
	Green Algae (Chlorococcales: Oocystaceae, Dictyosphaeraceae)	Filamentous Green Alga	Motile Algae (Volvocales)	Cyanobacteria (Nostocales, Oscillatoriales)
8	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Gonium</i> ‡ <i>Chlamydomonas</i>	<i>Anabaena</i> <i>Anabaenopsis</i> <i>Cylindrospermopsis</i> <i>Aphanizomenon</i> <i>Arthrospira</i> <i>Pseudanabaena</i> ‡ <i>Planktolyngbya</i>
9	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Chlamydomonas</i> †	<i>Arthrospira</i> <i>Cylindrospermopsis</i> <i>Anabaena</i> <i>Planktolyngbya</i> <i>Anabaenopsis</i> <i>Cylindrospermum</i> <i>Aphanizomenon</i>
11	<i>Oocystis</i> <i>Botryococcus</i>	<i>Chlorhormidium</i> ‡	<i>Volvox</i> <i>Chlamydomonas</i>	<i>Arthrospira</i> <i>Cylindrospermopsis</i> <i>racib.</i> <i>Anabaenopsis</i> <i>Anabaena circinalis</i> <i>Anabaena crassa</i> † <i>Cylindrospermum</i> <i>Anabaena straight</i>
12	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Chlamydomonas</i> <i>Volvox</i>	<i>Arthrospira</i> <i>Planktolyngbya</i> <i>Spirulina</i> ‡ <i>Cylindrospermopsis</i> <i>Anabaena</i> <i>Aphanizomenon</i> <i>Anabaenopsis</i> <i>Cylindrospermum</i>
13	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i> or <i>Oedogonium</i> sp.		<i>Arthrospira</i> <i>Anabaena</i> <i>Anabaenopsis</i> <i>Aphanizomenon</i> <i>Cylindrospermum</i> <i>licheniforme</i> <i>Cylindrospermopsis</i> <i>raciborskii</i> <i>Fine Planktolyngbya</i>
14	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Volvox</i> <i>Chlamydomonas</i>	<i>Arthrospira</i> <i>Anabaena</i> <i>Cylindrospermopsis</i> <i>Cylindrospermum</i> <i>Planktolyngbya</i> <i>Anabaenopsis</i> <i>Aphanizomenon</i>

‡ Denotes a false result.

† Denotes participant reported less than the required number of genera.

Lab Code	SAMPLE A			
	Green Algae (Chlorococcales: Oocystaceae, Dictyosphaeraceae)	Filamentous Green Alga	Motile Algae (Volvocales)	Cyanobacteria (Nostocales, Oscillatoriales)
15	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Gonium</i> ‡ <i>Chlamydomonas</i>	<i>Anabaena</i> <i>Aphanizomenon</i> <i>Anabaenopsis</i> <i>Cylindrospermum</i> <i>Cylindrospermopsis</i> <i>Arthrospira</i> <i>Planktolyngbya</i>
16	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Volvox</i> <i>Chlamydomonas</i>	<i>Arthrospira</i> <i>Anabaena</i> <i>Cylindrospermopsis</i> <i>Pseudanabaena</i> ‡ <i>Anabaenopsis</i> <i>Aphanizomenon</i> <i>Planktolyngbya</i>
19	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Chlamydomonas</i> <i>Pandorina</i> ‡	<i>Anabaena circinalis</i> <i>Anabaena</i> <i>aphanizomioides</i> <i>Anabaena smithii</i> † <i>Anabaenopsis</i> <i>elenkinii</i> <i>Aphanizomenon</i> <i>issatschenkoi</i> <i>Cylindrospermum</i> <i>licheniforme</i> <i>Arthrospira</i> aff. <i>maxima</i>
20	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i> or <i>Debarya</i>	<i>Volvox</i> <i>Chlamydomonas</i>	<i>Arthrospira</i> <i>Anabaena</i> <i>Planktolyngbya</i> <i>Anabaenopsis</i> <i>Cylindrospermopsis</i> <i>Cylindrospermum</i> <i>Aphanizomenon</i>
21	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Gonium</i> ‡ <i>Chlamydomonas</i>	<i>Arthrospira</i> <i>Anabaena</i> <i>Anabaenopsis</i> <i>Aphanizomenon</i> <i>Cylindrospermopsis</i> <i>Planktolyngbya</i> <i>Pseudanabaena</i> ‡
22	<i>Oocystis</i> <i>Lobocystis</i> ‡	<i>Mougeotia</i>	<i>Lobomonas</i> ‡ <i>Chlamydomonas</i>	<i>Arthrospira</i> <i>Cylindrospermopsis</i> <i>Planktolyngbya</i> <i>Cylindrospermum</i> <i>Anabaenopsis</i> <i>Anabaena</i> <i>Aphanizomenon</i>

‡ Denotes a false result.

† Denotes participant reported less than the required number of genera.



Lab Code	SAMPLE A			
	Green Algae (Chlorococcales: Oocystaceae, Dictyosphaeraceae)	Filamentous Green Alga	Motile Algae (Volvocales)	Cyanobacteria (Nostocales, Oscillatoriales)
23	<i>Oocystis</i> <i>Dictyosphaerium</i> ‡	<i>Mougeotia</i>	<i>Chlamydomonas</i> <i>Volvox</i>	<i>Anabaena</i> <i>Cylindrospermopsis</i> <i>Anabaenopsis</i> <i>Aphanizomenon</i> <i>Cylindrospermum</i> <i>Arthrospira</i> <i>Planktolyngbya</i>
24A	<i>Oocystis</i> <i>Dictyosphaerium</i> ‡	<i>Mougeotia</i>	<i>Gonium</i> ‡ <i>Chlamydomonas</i>	<i>Anabaena</i> <i>Aphanizomenon</i> <i>Cylindrospermopsis</i> <i>Anabaenopsis</i> <i>Arthrospira</i> <i>Planktolyngbya</i> <i>Pseudanabaena</i> ‡
24B	<i>Oocystis</i> <i>Dictyosphaerium</i> ‡	<i>Mougeotia</i>	<i>Gonium</i> ‡ <i>Chlamydomonas</i>	<i>Arthrospira</i> <i>Anabaena</i> <i>Planktolyngbya</i> <i>Cylindrospermopsis</i> <i>Pseudanabaena</i> ‡ <i>Anabaenopsis</i> <i>Aphanizomenon</i>
25	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Volvox</i> <i>Chlamydomonas</i>	<i>Arthrospira</i> <i>Pseudanabaena</i> ‡ <i>Cylindrospermopsis</i> <i>Anabaena</i> <i>Anabaenopsis</i> <i>Aphanizomenon</i>

‡ Denotes a false result.

Lab Code	SAMPLE B			
	Green Algae (Chlorococcales: Oocystaceae, Dictyosphaeraceae)	Filamentous Green Alga	Motile Algae (Volvocales)	Cyanobacteria (Nostocales, Oscillatoriales)
1	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Gonium</i> ‡ <i>Stenophora</i> ‡	<i>Arthrospira</i> <i>Spirulina</i> ‡ <i>Planktolyngbya</i> <i>Anabaena</i> <i>Aphanizomenon</i> <i>Cylindrospermopsis</i> <i>Anabaenopsis</i>
2	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Volvox</i> <i>Chlamydomonas</i>	<i>Anabaena</i> <i>Anabaenopsis</i> <i>Aphanizomenon</i> <i>Cylindrospermopsis</i> <i>Cylindrospermum</i> <i>Arthrospira</i> <i>Planktolyngbya</i>
3	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Volvox</i> <i>Chlamydomonas</i>	<i>Anabaena</i> <i>Arthrospira</i> <i>Cylindrospermopsis</i> <i>Anabaenopsis</i> <i>Aphanizomenon</i> <i>Planktolyngbya</i> <i>Cylindrospermum</i>
4	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Chlamydomonas</i> <i>Pandorina</i> ‡	<i>Arthrospira</i> <i>Planktolyngbya</i> <i>Cylindrospermum</i> <i>Cylindrospermopsis</i> <i>raciborskii</i> <i>Anabaena</i> <i>Anabaenopsis</i> <i>Aphanizomenon</i>
5	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Volvox</i> <i>Chlamydomonas</i>	<i>Anabaena</i> <i>Anabaenopsis</i> <i>Cylindrospermopsis</i> <i>Cylindrospermum</i> <i>Aphanizomenon</i> <i>Arthrospira</i> <i>Planktolyngbya</i>
6	<i>Oocystis</i> <i>Dictyosphaerium</i> sp. ‡	<i>Hormidium</i> sp. ‡	<i>Gonium</i> sp. ‡ <i>Chlamydomonas</i>	<i>Anabaena</i> <i>Arthrospira</i> <i>Cylindrospermopsis</i> <i>Aphanizomenon</i> <i>Anabaenopsis</i> <i>Pseudanabaena</i> ‡ <i>Trichodesmium</i>
7	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Chlamydomonas</i> <i>Pandorina</i> ‡	<i>Arthrospira</i> <i>Pseudanabaena</i> ‡ <i>Anabaena</i> <i>Cylindrospermopsis</i> <i>Anabaenopsis</i> <i>Planktolyngbya</i> <i>Aphanizomenon</i>

‡ Denotes a false result.

Lab Code	SAMPLE B			
	Green Algae (Chlorococcales: Oocystaceae, Dictyosphaeraceae)	Filamentous Green Alga	Motile Algae (Volvocales)	Cyanobacteria (Nostocales, Oscillatoriales)
8	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Gonium</i> ‡ <i>Chlamydomonas</i>	<i>Anabaena</i> <i>Anabaenopsis</i> <i>Cylindrospermopsis</i> <i>Aphanizomenon</i> <i>Arthrospira</i> <i>Pseudanabaena</i> ‡ <i>Planktolyngbya</i>
9	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Chlamydomonas</i> †	<i>Arthrospira</i> <i>Cylindrospermopsis</i> <i>Anabaena</i> <i>Planktolyngbya</i> <i>Anabaenopsis</i> <i>Cylindrospermum</i> <i>Aphanizomenon</i>
11	<i>Oocystis</i> <i>Botryococcus</i>	<i>Chlorhormidium</i> ‡	<i>Volvox</i> <i>Chlamydomonas</i>	<i>Arthrospira</i> <i>Cylindrospermopsis</i> <i>racib.</i> <i>Anabaena circinalis</i> <i>Anabaena solitaria</i> † <i>Aphanizomenon</i> <i>Anabaenopsis</i> <i>Anabaena crassa</i>
12	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Chlamydomonas</i> <i>Volvox</i>	<i>Cylindrospermopsis</i> <i>Cylindrospermum</i> <i>Anabaena</i> <i>Anabaenopsis</i> <i>Aphanizomenon</i> <i>Arthrospira</i> <i>Planktolyngbya</i>
13	<i>Oocystis</i> <i>Botryococcus</i>			<i>Arthrospira</i> sp. <i>Anabaena</i> sp. <i>Anabaenopsis</i> sp. <i>Aphanizomenon</i> sp. <i>Cylindrospermum</i> sp. <i>Cylindrospermopsis</i> sp. fine <i>Planktolyngbya</i>
14	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Volvox</i> <i>Chlamydomonas</i>	<i>Arthrospira</i> <i>Cylindrospermum</i> <i>Cylindrospermopsis</i> <i>Planktolyngbya</i> <i>Anabaena</i> <i>Anabaenopsis</i> <i>Aphanizomenon</i>

‡ Denotes a false result.

† Denotes participant reported less than the required number of genera.

Lab Code	SAMPLE B			
	Green Algae (Chlorococcales: Oocystaceae, Dictyosphaeraceae)	Filamentous Green Alga	Motile Algae (Volvocales)	Cyanobacteria (Nostocales, Oscillatoriales)
15	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Gonium</i> ‡ <i>Chlamydomonas</i>	<i>Anabaena</i> <i>Aphanizomenon</i> <i>Anabaenopsis</i> <i>Cylindrospermopsis</i> <i>Cylindrospermum</i> <i>Arthrospira</i> <i>Planktolyngbya</i>
16	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Volvox</i> <i>Chlamydomonas</i>	<i>Arthrospira</i> <i>Anabaena</i> <i>Cylindrospermopsis</i> <i>Pseudanabaena</i> ‡ <i>Anabaenopsis</i> <i>Aphanizomenon</i> <i>Planktolyngbya</i>
19	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Chlamydomonas</i> <i>Pandorina</i> ‡	<i>Anabaena circinalis</i> <i>Anabaena</i> <i>aphanizomioides</i> <i>Anabaena smithii</i> † <i>Anabaenopsis</i> <i>elenkinii</i> <i>Aphanizomenon</i> <i>issatschenkoi</i> <i>Cylindrospermum</i> <i>licheniforme</i> <i>Arthrospira</i> aff. <i>maxima</i>
20	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i> or <i>Debarya</i>	<i>Volvox</i> <i>Chlamydomonas</i>	<i>Arthrospira</i> <i>Anabaena</i> <i>Planktolyngbya</i> <i>Anabaenopsis</i> <i>Cylindrospermopsis</i> <i>Cylindrospermum</i> <i>Aphanizomenon</i>
21	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Gonium</i> ‡ <i>Chlamydomonas</i>	<i>Arthrospira</i> <i>Anabaena</i> <i>Anabaenopsis</i> <i>Aphanizomenon</i> <i>Cylindrospermopsis</i> <i>Planktolyngbya</i> <i>Limnothrix</i>
22	<i>Oocystis</i> <i>Lobocystis</i> ‡	<i>Mougeotia</i>	<i>Lobomonas</i> ‡ <i>Chlamydomonas</i>	<i>Arthrospira</i> <i>Cylindrospermum</i> <i>Planktolyngbya</i> <i>Cylindrospermopsis</i> <i>Anabaenopsis</i> <i>Anabaena</i> <i>Aphanizomenon</i>

‡ Denotes a false result.

† Denotes participant reported less than the required number of genera.

Lab Code	SAMPLE B			
	Green Algae (Chlorococcales: Oocystaceae, Dictyosphaeraceae)	Filamentous Green Alga	Motile Algae (Volvocales)	Cyanobacteria (Nostocales, Oscillatoriales)
23	<i>Oocystis</i> <i>Dictyosphaerium</i> ‡	<i>Mougeotia</i>	<i>Chlamydomonas</i> <i>Volvox</i>	<i>Anabaena</i> <i>Cylindrospermopsis</i> <i>Anabaenopsis</i> <i>Aphanizomenon</i> <i>Cylindrospermum</i> <i>Arthrospira</i> <i>Planktolyngbya</i>
24A	<i>Oocystis</i> <i>Dictyosphaerium</i> ‡	<i>Mougeotia</i>	<i>Gonium</i> ‡ <i>Chlamydomonas</i>	<i>Anabaena</i> <i>Anabaenopsis</i> <i>Aphanizomenon</i> <i>Cylindrospermopsis</i> <i>Arthrospira</i> <i>Planktolyngbya</i> <i>Pseudanabaena</i> ‡
24B	<i>Oocystis</i> <i>Dictyosphaerium</i> ‡	<i>Mougeotia</i>	<i>Gonium</i> ‡ <i>Chlamydomonas</i>	<i>Arthrospira</i> <i>Anabaena</i> <i>Planktolyngbya</i> <i>Cylindrospermopsis</i> <i>Pseudanabaena</i> ‡ <i>Anabaenopsis</i> <i>Aphanizomenon</i>
25	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Volvox</i> <i>Chlamydomonas</i>	<i>Arthrospira</i> <i>Anabaenopsis</i> <i>Anabaena</i> <i>Aphanizomenon</i> <i>Cylindrospermopsis</i> <i>Pseudanabaena</i> ‡ <i>Oscillatoria</i>

‡ Denotes a false result.

Lab Code	SAMPLE C			
	Green Algae (Chlorococcales: Oocystaceae, Dictyosphaeraceae)	Filamentous Green Alga	Motile Algae (Volvocales)	Cyanobacteria (Nostocales, Oscillatoriales)
1	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Gonium</i> ‡ <i>Stenophora</i> ‡	<i>Arthrospira</i> <i>Anabaena</i> <i>Planktolyngbya</i> <i>Aphanizomenon</i> <i>Cylindrospermopsis</i> <i>Psuedanabaena</i> ‡ <i>Anabaenopsis</i>
2	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Volvox</i> <i>Chlamydomonas</i>	<i>Anabaena</i> <i>Aphanizomenon</i> <i>Cylindrospermopsis</i> <i>Cylindrospermum</i> <i>Arthrospira</i> <i>Psuedanabaena</i> ‡ <i>Planktolyngbya</i>
3	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Volvox</i> <i>Chlamydomonas</i>	<i>Anabaena</i> <i>Arthrospira</i> <i>Cylindrospermopsis</i> <i>Anabaenopsis</i> <i>Aphanizomenon</i> <i>Planktolyngbya</i> <i>Cylindrospermum</i>
4	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Chlamydomonas</i> <i>Pandorina</i> ‡	<i>Arthrospira</i> <i>Planktolyngbya</i> <i>Cylindrospermum</i> <i>Cylindrospermopsis</i> <i>raciborskii</i> <i>Anabaena</i> <i>Anabaenopsis</i> <i>Aphanizomenon</i>
5	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Volvox</i> <i>Chlamydomonas</i>	<i>Anabaena</i> <i>Anabaenopsis</i> <i>Cylindrospermopsis</i> <i>Cylindrospermum</i> <i>Aphanizomenon</i> <i>Arthrospira</i> <i>Planktolyngbya</i>
6	<i>Oocystis</i> <i>Dictyosphaerium</i> sp. ‡	<i>Hormidium</i> sp. ‡	<i>Gonium</i> sp. ‡ <i>Chlamydomonas</i>	<i>Anabaena</i> <i>Arthrospira</i> <i>Cylindrospermopsis</i> <i>Aphanizomenon</i> <i>Anabaenopsis</i> <i>Pseudanabaena</i> ‡ <i>Trichodesmium</i>
7	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Chlamydomonas</i> <i>Pandorina</i> ‡	<i>Arthrospira</i> <i>Cylindrospermopsis</i> <i>Aphanizomenon</i> <i>Pseudanabaena</i> ‡ <i>Anabaena</i> <i>Anabaenopsis</i> <i>Planktolyngbya</i>

‡ Denotes a false result.

Lab Code	SAMPLE C			
	Green Algae (Chlorococcales: Oocystaceae, Dictyosphaeraceae)	Filamentous Green Alga	Motile Algae (Volvocales)	Cyanobacteria (Nostocales, Oscillatoriales)
8	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Gonium</i> ‡ <i>Chlamydomonas</i>	<i>Anabaena</i> <i>Cylindrospermopsis</i> <i>Aphanizomenon</i> <i>Arthrospira</i> <i>Pseudanabaena</i> ‡ <i>Planktolyngbya</i> <i>Tychonema</i> ‡
9	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Chlamydomonas</i> †	<i>Arthrospira</i> <i>Cylindrospermopsis</i> <i>Anabaena</i> <i>Planktolyngbya</i> <i>Anabaenopsis</i> <i>Cylindrospermum</i> <i>Aphanizomenon</i>
11	<i>Oocystis</i> <i>Botryococcus</i>	<i>Chlorhormidium</i> ‡	<i>Volvox</i> <i>Chlamydomonas</i>	<i>Arthrospira</i> †
12	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Chlamydomonas</i> <i>Volvox</i>	<i>Anabaena</i> <i>Anabaenopsis</i> <i>Aphanizomenon</i> <i>Cylindrospermopsis</i> <i>Cylindrospermum</i> <i>Arthrospira</i> <i>Planktolyngbya</i>
13	<i>Oocystis</i> <i>Botryococcus</i>			<i>Arthrospira</i> sp. <i>Anabaena</i> sp. <i>Anabaenopsis</i> sp. <i>Aphanizomenon</i> sp. <i>Cylindrospermum</i> sp. <i>Cylindrospermopsis</i> sp. fine <i>Planktolyngbya</i>
14	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Volvox</i> <i>Chlamydomonas</i>	<i>Arthrospira</i> <i>Aphanizomenon</i> <i>Cylindrospermopsis</i> <i>Anabaena</i> <i>Cylindrospermum</i> <i>Anabaenopsis</i> <i>Planktolyngbya</i>

‡ Denotes a false result.

† Denotes participant reported less than the required number of genera.

Lab Code	SAMPLE C			
	Green Algae (Chlorococcales: Oocystaceae, Dictyosphaeraceae)	Filamentous Green Alga	Motile Algae (Volvocales)	Cyanobacteria (Nostocales, Oscillatoriales)
15	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Gonium</i> ‡ <i>Chlamydomonas</i>	<i>Anabaena</i> <i>Aphanizomenon</i> <i>Anabaenopsis</i> <i>Cylindrospermum</i> <i>Cylindrospermopsis</i> <i>Arthrospira</i> <i>Planktolyngbya</i>
16	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Volvox</i> <i>Chlamydomonas</i>	<i>Arthrospira</i> <i>Anabaena</i> <i>Cylindrospermopsis</i> <i>Pseudanabaena</i> ‡ <i>Anabaenopsis</i> <i>Aphanizomenon</i> <i>Planktolyngbya</i>
19	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Chlamydomonas</i> <i>Pandorina</i> ‡	<i>Anabaena circinalis</i> <i>Anabaena</i> <i>aphanizomoides</i> † <i>Anabaena smithii</i> <i>Anabaenopsis</i> <i>elenkinii</i> <i>Aphanizomenon</i> <i>issatschenkoi</i> <i>Cylindrospermum</i> <i>licheniforme</i> <i>Arthrospira</i> aff. <i>maxima</i>
20	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i> or <i>Debarya</i>	<i>Volvox</i> <i>Chlamydomonas</i>	<i>Arthrospira</i> <i>Anabaena</i> <i>Planktolyngbya</i> <i>Anabaenopsis</i> <i>Cylindrospermopsis</i> <i>Cylindrospermum</i> <i>Aphanizomenon</i>
21	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Gonium</i> ‡ <i>Chlamydomonas</i>	<i>Arthrospira</i> <i>Anabaena</i> <i>Anabaenopsis</i> <i>Aphanizomenon</i> <i>Cylindrospermopsis</i> <i>Planktolyngbya</i> <i>Pseudanabaena</i> ‡
22	<i>Oocystis</i> <i>Lobocystis</i> ‡	<i>Mougeotia</i>	<i>Lobomonas</i> ‡ <i>Chlamydomonas</i>	<i>Arthrospira</i> <i>Cylindrospermum</i> <i>Planktolyngbya</i> <i>Cylindrospermopsis</i> <i>Anabaenopsis</i> <i>Anabaena</i> <i>Aphanizomenon</i>

‡ Denotes a false result.

† Denotes participant reported less than the required number of genera.



Lab Code	SAMPLE C			
	Green Algae (Chlorococcales: Oocystaceae, Dictyosphaeraceae)	Filamentous Green Alga	Motile Algae (Volvocales)	Cyanobacteria (Nostocales, Oscillatoriales)
23	<i>Oocystis</i> <i>Dictyosphaerium</i> ‡	<i>Mougeotia</i>	<i>Chlamydomonas</i> <i>Volvox</i>	<i>Anabaena</i> <i>Cylindrospermopsis</i> <i>Anabaenopsis</i> <i>Aphanizomenon</i> <i>Cylindrospermum</i> <i>Arthrospira</i> <i>Planktolyngbya</i>
24A	<i>Oocystis</i> <i>Dictyosphaerium</i> ‡	<i>Mougeotia</i>	<i>Gonium</i> ‡ <i>Chlamydomonas</i>	<i>Anabaena</i> <i>Anabaenopsis</i> <i>Aphanizomenon</i> <i>Cylindrospermopsis</i> <i>Arthrospira</i> <i>Planktolyngbya</i> <i>Pseudanabaena</i> ‡
24B	<i>Oocystis</i> <i>Dictyosphaerium</i> ‡	<i>Mougeotia</i>	<i>Gonium</i> ‡ <i>Chlamydomonas</i>	<i>Arthrospira</i> <i>Anabaena</i> <i>Planktolyngbya</i> <i>Cylindrospermopsis</i> <i>Pseudanabaena</i> ‡ <i>Anabaenopsis</i> <i>Aphanizomenon</i>
25	<i>Oocystis</i> <i>Botryococcus</i>	<i>Mougeotia</i>	<i>Volvox</i> <i>Chlamydomonas</i>	<i>Arthrospira</i> <i>Pseudanabaena</i> ‡ <i>Anabaenopsis</i> <i>Anabaena</i> <i>Cylindrospermopsis</i> <i>Aphanizomenon</i> <i>Planktolyngbya</i>

‡ Denotes a false result.

**Enumeration – All Results - Organism 1**

Lab Code	Genus enumerated	Sample A Cells/ml	Sample B Cells/ml	Sample C Cells/ml
<b>1</b>	<i>Oocystis</i>	12498	29731	28279
<b>2</b>	<i>Oocystis</i>	10733	6933	9600
<b>3</b>	<i>Oocystis</i>	43644	26635	40760
<b>4</b>	<i>Oocystis</i>	14533	11692	14533
<b>5</b>	<i>Oocystis</i>	15943	14935	17006
<b>6</b>	<i>Oocystis</i>	5451	12405	12499
<b>7</b>	<i>Oocystis</i>	13244	24044	19422
<b>8</b>	<i>Oocystis</i>	34600	35700	29800
<b>11</b>	<i>Oocystis</i>	12800	12800	13500
<b>12</b>	<i>Oocystis</i>	12025	15502	16967
<b>14</b>	<i>Oocystis</i>	8912	10840	10840
<b>15</b>	<i>Oocystis</i>	15106	18875	14691
<b>16</b>	<i>Oocystis</i>	8000	11385	12424
<b>19</b>	<i>Oocystis</i>	6700	6700	6500
<b>20</b>	<i>Oocystis</i>	10925	11720	10425
<b>21</b>	<i>Oocystis</i>	37000	39000	38000
<b>22</b>	<i>Oocystis</i>	14500	12360	13160
<b>23</b>	<i>Oocystis</i>	29480	16480	18840
<b>24A</b>	<i>Oocystis</i>	18000	27000	27000
<b>24B</b>	<i>Oocystis</i>	17393	27463	27100
<b>25</b>	<i>Oocystis</i>	7050	10850	9950

**Enumeration – All Results - Organism 2**

Lab Code	Genus enumerated	Sample A Cells/ml	Sample B Cells/ml	Sample C Cells/ml
<b>1</b>	<i>Mougeotia</i>	45575	57568	44565
<b>2</b>	<i>Mougeotia</i>	58667	64000	61333
<b>3</b>	<i>Mougeotia</i>	49243	59996	54681
<b>4</b>	<i>Mougeotia</i>	34429	40077	39231
<b>5</b>	<i>Mougeotia</i>	64184	60077	62747
<b>6</b>	<i>Hormidium sp.</i>	36181	42665	32798
<b>7</b>	<i>Mougeotia</i>	79467	78044	94667
<b>8</b>	<i>Mougeotia</i>	59200	51800	51090
<b>11</b>	<i>Chlorhormidium</i>	45500	45400	53250
<b>12</b>	<i>Mougeotia</i>	53649	56623	59297
<b>14</b>	<i>Mougeotia</i>	53985	55270	54499
<b>15</b>	<i>Mougeotia</i>	41958	54000	60600
<b>16</b>	<i>Mougeotia</i>	68334	74334	51334
<b>19</b>	<i>Mougeotia</i>	52000	58000	53000
<b>20</b>	<i>Mougeotia</i>	59570	64620	63060
<b>21</b>	<i>Mougeotia</i>	64000	58000	57000
<b>22</b>	<i>Mougeotia</i>	80320	66040	92640
<b>23</b>	<i>Mougeotia</i>	87696	63516	75238
<b>24A</b>	<i>Mougeotia</i>	56000	47000	54000
<b>24B</b>	<i>Mougeotia</i>	56758	47147	54547
<b>25</b>	<i>Mougeotia</i>	46250	51350	52250

**Enumeration – All Results - Organism 3**

Lab Code	Genus enumerated	Sample A Cells/ml	Sample B Cells/ml	Sample C Cells/ml
<b>1</b>	<i>Stenophora</i> *	48479	37874	28784
<b>2</b>	<i>Volvox</i> *	40333	40750	31750
<b>3</b>	<i>Chlamydomonas</i>	6528	3156	2862
<b>4</b>	<i>Chlamydomonas</i>	101250	91000	76000
<b>5</b>	<i>Volvox</i> *	279228	234247	217919
<b>6</b>	<i>Chlamydomonas</i>	1692	2349	3007
<b>7</b>	<i>Chlamydomonas</i>	6133	4356	3333
<b>8</b>	<i>Chlamydomonas</i>	4300	3930	2970
<b>11</b>	<i>Volvox</i> *	256000	220000	240000
<b>12</b>	<i>Chlamydomonas</i>	4469	3087	2733
<b>14</b>	<i>Chlamydomonas</i>	3713	2635	2442
<b>15</b>	<i>Chlamydomonas</i>	2275	2700	2075
<b>16</b>	<i>Volvox</i> *	191000	149000	173000
<b>19</b>	<i>Chlamydomonas</i>	6900	6700	7200
<b>20</b>	<i>Volvox</i> *	260250	228200	209375
<b>21</b>	<i>Chlamydomonas</i>	5000	3700	3900
<b>22</b>	<i>Lobomonus</i> *	188700	166300	195000
<b>23</b>	<i>Chlamydomonas</i>	7560	2260	1880
<b>24A</b>	<i>Chlamydomonas</i>	3000	2600	2600
<b>24B</b>	<i>Chlamydomonas</i>	3662	3051	3204
<b>25</b>	<i>Volvox</i> *	193000	129000	90000

Notes:

\* Classed as mis-identification and so not included in statistical analysis

**Enumeration – All Results - Organism 4**

Lab Code	Genus enumerated	Sample A Cells/ml	Sample B Cells/ml	Sample C Cells/ml
1	<i>Arthrospira</i>	101881	115705	116273
2	<i>Arthrospira</i>	390000	269444	149733
3	<i>Arthrospira</i>	547216	347124	306062
4	<i>Arthrospira</i>	525643	261769	227077
5	<i>Arthrospira</i>	300428	171785	175471
6	<i>Arthrospira</i>	125177	86834	90030
7	<i>Arthrospira</i>	364267	96533	235200
8	<i>Arthrospira</i>	240000	177000	147000
9	<i>Arthrospira</i>	259395	131090	148910
11	<i>Arthrospira</i>	560588	512900	414460
12	<i>Arthrospira</i>	536518	319813	293767
13	<i>Arthrospira</i>	490000	190000	230000
14	<i>Arthrospira</i>	261183	158398	151676
15	<i>Arthrospira</i>	387400	283750	165170
16	<i>Arthrospira</i>	459667	249667	322000
19	<i>Arthrospira</i>	300000	190000	180000
20	<i>Arthrospira</i>	374070	288500	272780
21	<i>Arthrospira</i>	590000	250000	240000
22	<i>Arthrospira</i>	366200	148300	156000
23	<i>Arthrospira</i>	379236	159975	225592
24A	<i>Arthrospira</i>	340000	280000	270000
24B	<i>Arthrospira</i>	335258	281504	280000
25	<i>Arthrospira</i>	398500	261000	202000

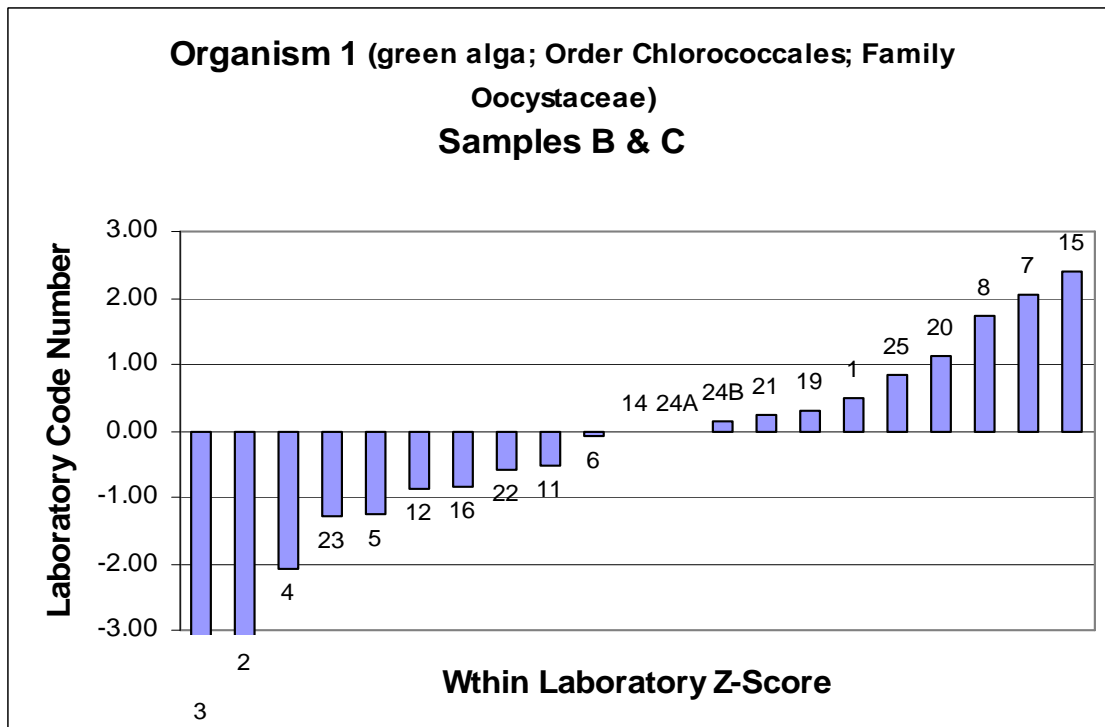
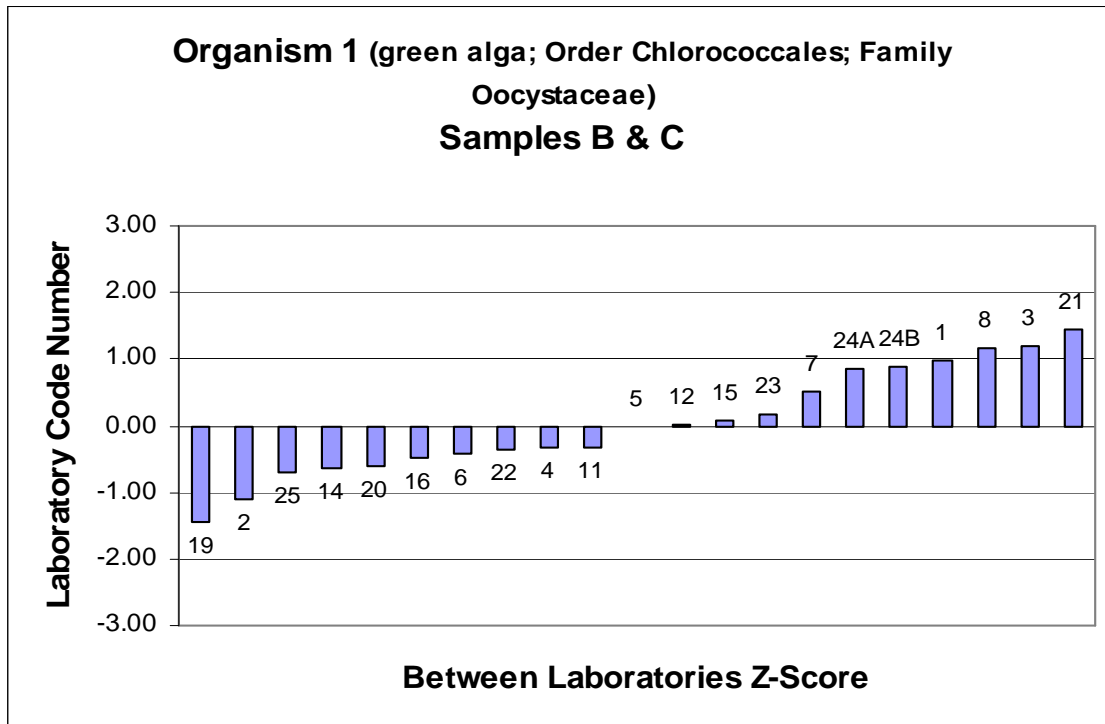
**Z-Scores and Summary Results****Organism 1 (green alga; Order Chlorococcales; Family Oocystaceae)****Sample Pair B & C**Accepted genus: *Oocystis*

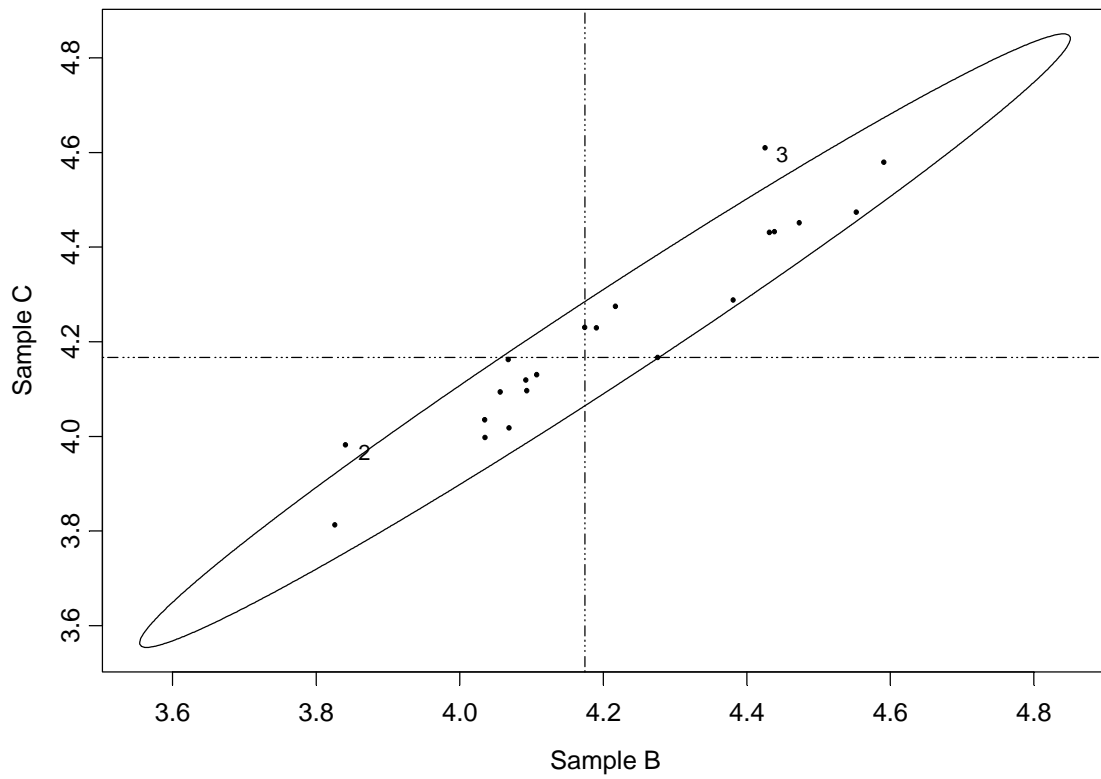
Lab Code	Log10 Sample B	Log10 Sample C	Between Laboratories Z-Score	Within Laboratory Z-Score
<b>1</b>	4.47	4.45	0.98	0.48
<b>2</b>	3.84	3.98	-1.10	-3.13 §
<b>3</b>	4.43	4.61	1.20	-4.09 §
<b>4</b>	4.07	4.16	-0.33	-2.09
<b>5</b>	4.17	4.23	0.00	-1.25
<b>6</b>	4.09	4.10	-0.41	-0.07
<b>7</b>	4.38	4.29	0.50	2.05
<b>8</b>	4.55	4.47	1.18	1.74
<b>11</b>	4.11	4.13	-0.32	-0.51
<b>12</b>	4.19	4.23	0.03	-0.87
<b>14</b>	4.04	4.04	-0.63	0.00
<b>15</b>	4.28	4.17	0.07	2.41
<b>16</b>	4.06	4.09	-0.48	-0.84
<b>19</b>	3.83	3.81	-1.45	0.29
<b>20</b>	4.07	4.02	-0.60	1.13
<b>21</b>	4.59	4.58	1.45	0.25
<b>22</b>	4.09	4.12	-0.37	-0.60
<b>23</b>	4.22	4.28	0.17	-1.29
<b>24A</b>	4.43	4.43	0.87	0.00
<b>24B</b>	4.44	4.43	0.88	0.13
<b>25</b>	4.04	4.00	-0.70	0.83

No. of Results	21	21
Median	4.174	4.167
Normalised IQR	0.265	0.250
Robust CV	6.35%	6.00%
Minimum	3.83	3.81
Maximum	4.59	4.61
Range	0.76	0.80

**Notes:**

§ denotes an outlier.

**Z-Score Charts**

**Youden Diagram****Organism 1****Notes:**

The following results are highlighted as outliers:

<u>Code</u>	<u>Sample B</u>	<u>Sample C</u>	<u>Between Z-score</u>	<u>Within Z-score</u>
2	3.840921	3.982271	-1.10174	-3.12778
3	4.425453	4.610234	1.195062	-4.08883



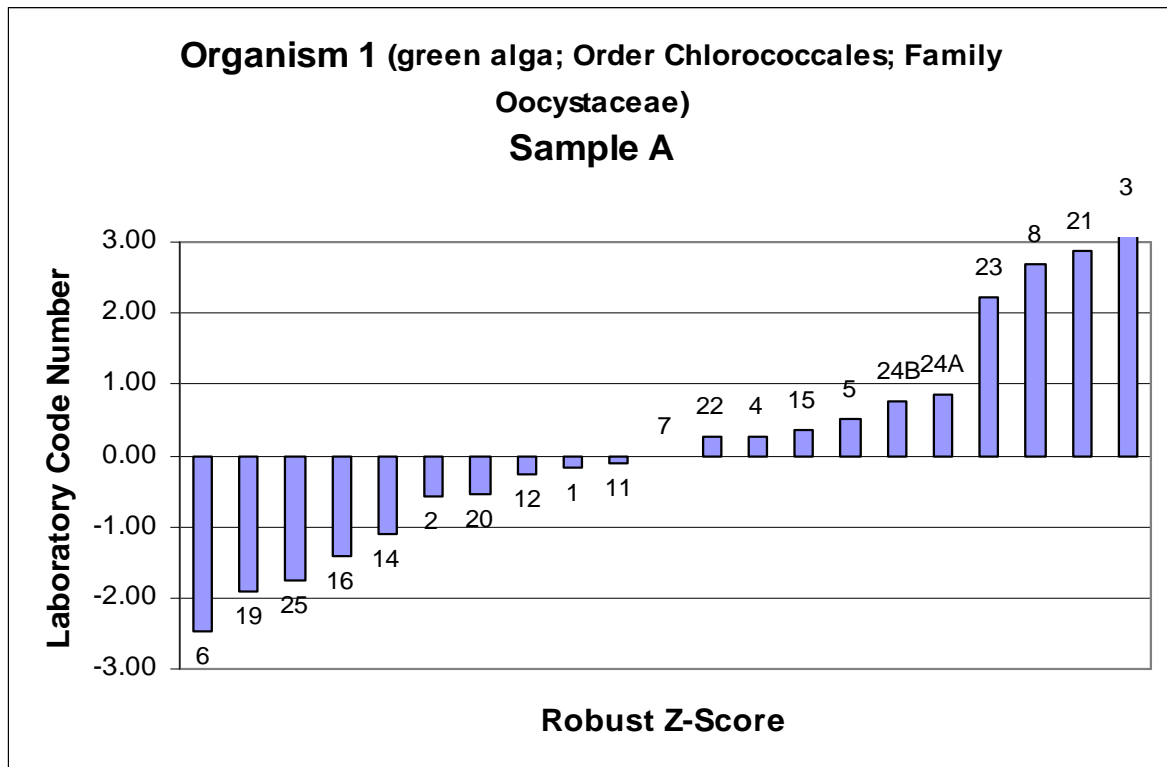
**Z-Scores and Summary Results****Organism 1 (green alga; Order Chlorococcales; Family Oocystaceae)****Sample A**Accepted genus: *Oocystis*

Lab Code	Log10 Sample A	Robust Z-Score
<b>1</b>	4.10	-0.16
<b>2</b>	4.03	-0.59
<b>3</b>	4.64	3.33 §
<b>4</b>	4.16	0.26
<b>5</b>	4.20	0.52
<b>6</b>	3.74	-2.48
<b>7</b>	4.12	0.00
<b>8</b>	4.54	2.68
<b>11</b>	4.11	-0.10
<b>12</b>	4.08	-0.27
<b>14</b>	3.95	-1.11
<b>15</b>	4.18	0.37
<b>16</b>	3.90	-1.41
<b>19</b>	3.83	-1.90
<b>20</b>	4.04	-0.54
<b>21</b>	4.57	2.87
<b>22</b>	4.16	0.25
<b>23</b>	4.47	2.24
<b>24A</b>	4.26	0.86
<b>24B</b>	4.24	0.76
<b>25</b>	3.85	-1.76

No. of Results	21
Median	4.122
Normalised IQR	0.155
Robust CV	3.77%
Minimum	3.74
Maximum	4.64
Range	0.90

**Notes:**

§ denotes an outlier

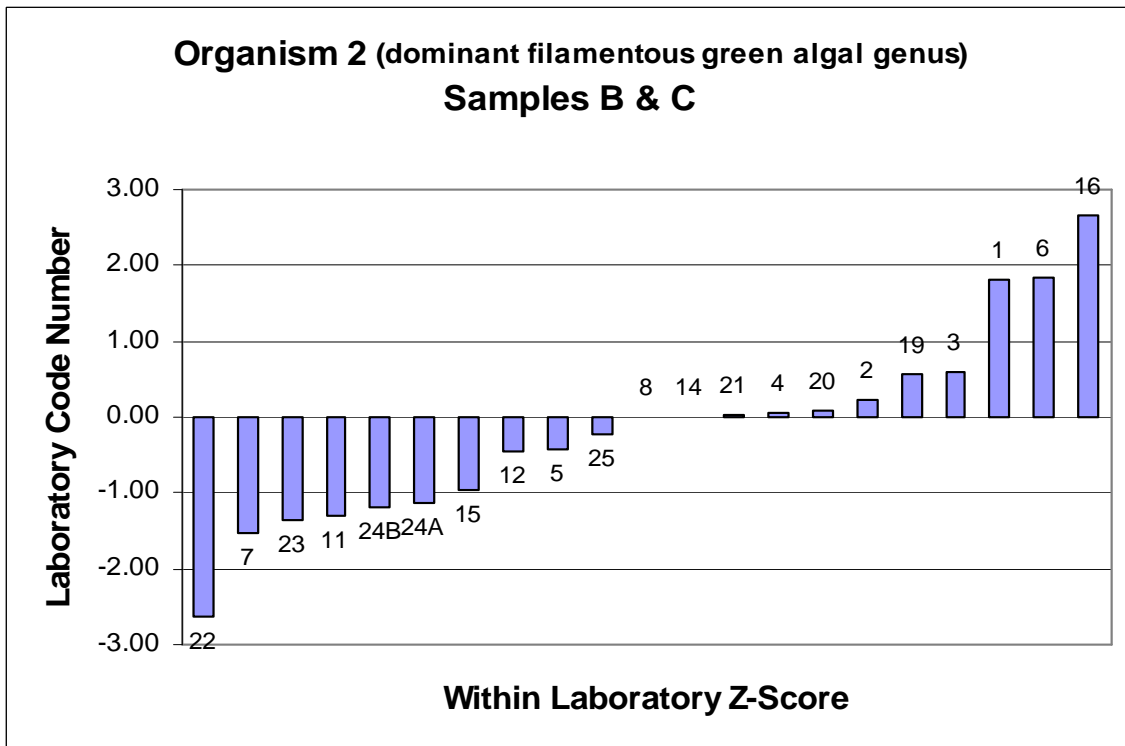
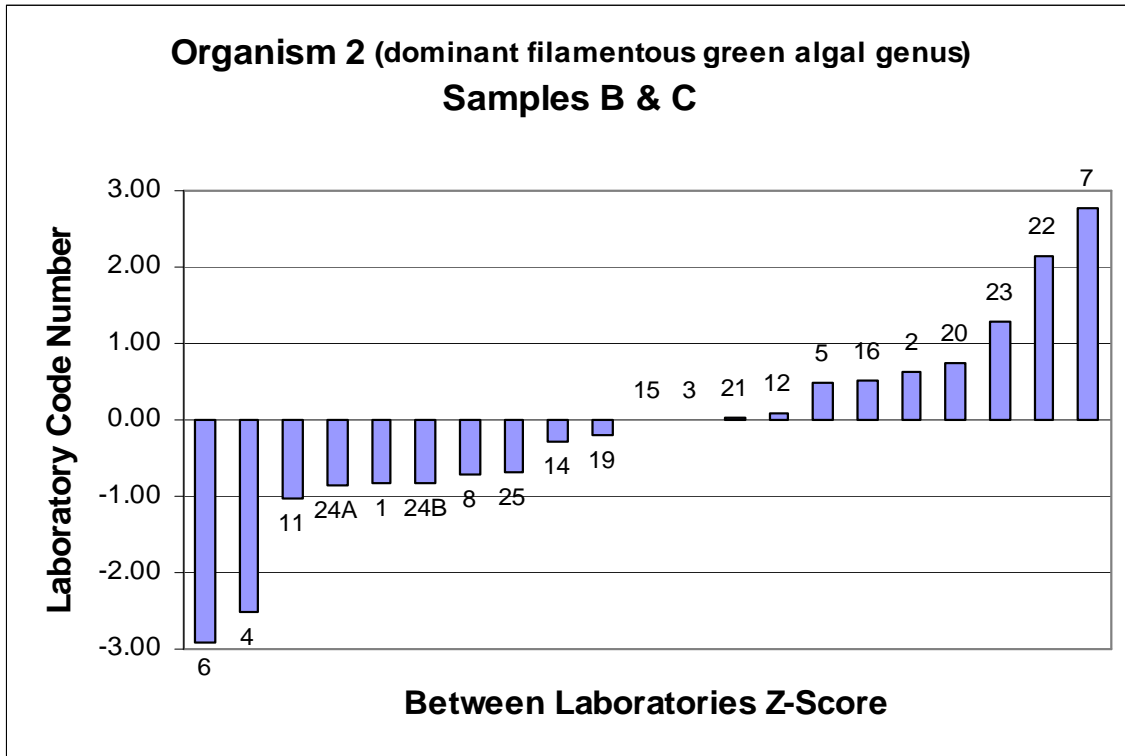
**Z-Score Chart**

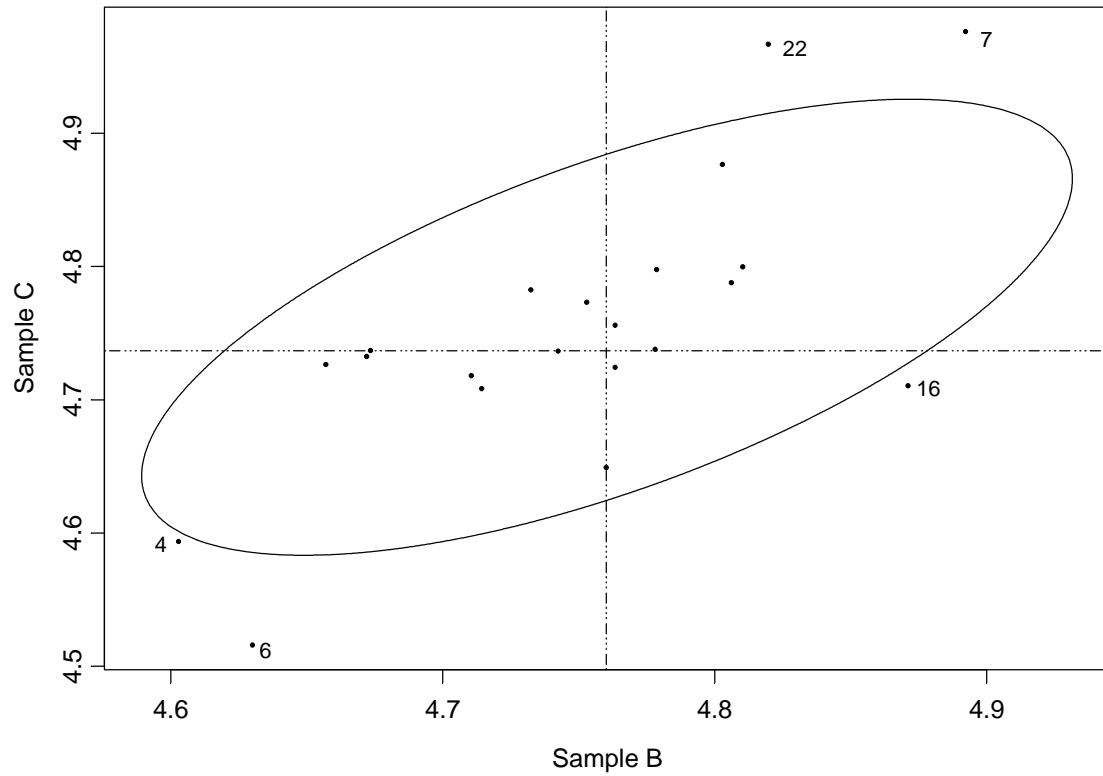
**Z-Scores and Summary Results**  
**Organism 2 (dominant filamentous green algal genus)**  
**Sample Pair B & C**

Accepted genera: *Mougeotia*, *Hormidium* sp., *Chlorhormidium*

Lab Code	Log10 Sample B	Log10 Sample C	Between Laboratories Z-Score	Within Laboratory Z-Score
<b>1</b>	4.76	4.65	-0.83	1.80
<b>2</b>	4.81	4.79	0.62	0.21
<b>3</b>	4.78	4.74	0.01	0.59
<b>4</b>	4.60	4.59	-2.51	0.06
<b>5</b>	4.78	4.80	0.48	-0.43
<b>6</b>	4.63	4.52	-2.90	1.85
<b>7</b>	4.89	4.98	2.78	-1.54
<b>8</b>	4.71	4.71	-0.73	0.00
<b>11</b>	4.66	4.73	-1.04	-1.29
<b>12</b>	4.75	4.77	0.09	-0.45
<b>14</b>	4.74	4.74	-0.28	0.00
<b>15</b>	4.73	4.78	0.00	-0.96
<b>16</b>	4.87	4.71	0.53	2.65
<b>19</b>	4.76	4.72	-0.21	0.57
<b>20</b>	4.81	4.80	0.75	0.08
<b>21</b>	4.76	4.76	0.03	0.03
<b>22</b>	4.82	4.97	2.14	-2.62
<b>23</b>	4.80	4.88	1.29	-1.36
<b>24A</b>	4.67	4.73	-0.87	-1.14
<b>24B</b>	4.67	4.74	-0.82	-1.19
<b>25</b>	4.71	4.72	-0.68	-0.23

No. of Results	21	21
Median	4.760	4.737
Normalised IQR	0.068	0.052
Robust CV	1.44%	1.09%
Minimum	4.60	4.52
Maximum	4.89	4.98
Range	0.29	0.46

**Z-Score Charts**

**Youden Diagram****Organism 2****Notes:**

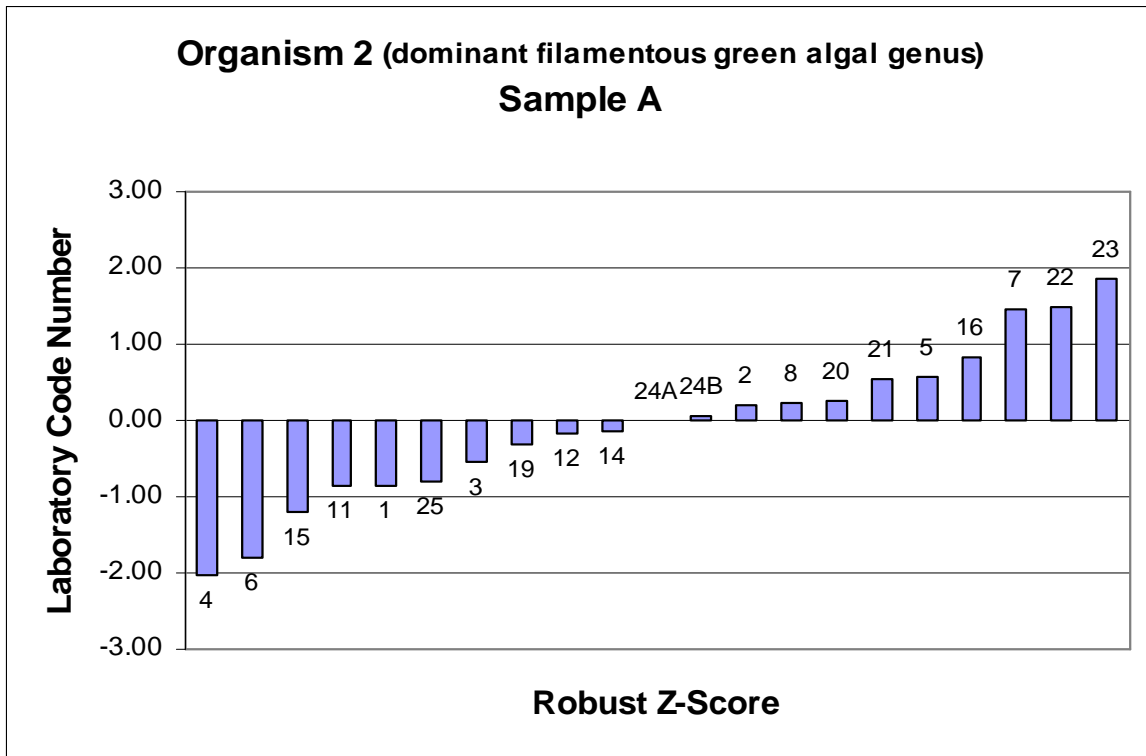
There were no outliers for this organism. The highlighted lab codes which are outside the ellipse obtained z-scores that were greater than 2 or less than -2.

**Z-Scores and Summary Results**  
**Organism 2 (dominant filamentous green algal genus)**  
**Sample A**

Accepted genera: *Mougeotia*, *Hormidium* sp., *Chlorhormidium*

Lab Code	Log10 Sample A	Robust Z-Score
<b>1</b>	4.66	-0.86
<b>2</b>	4.77	0.19
<b>3</b>	4.69	-0.53
<b>4</b>	4.54	-2.02
<b>5</b>	4.81	0.57
<b>6</b>	4.56	-1.81
<b>7</b>	4.90	1.45
<b>8</b>	4.77	0.23
<b>11</b>	4.66	-0.86
<b>12</b>	4.73	-0.18
<b>14</b>	4.73	-0.15
<b>15</b>	4.62	-1.20
<b>16</b>	4.83	0.83
<b>19</b>	4.72	-0.31
<b>20</b>	4.78	0.26
<b>21</b>	4.81	0.55
<b>22</b>	4.90	1.50
<b>23</b>	4.94	1.86
<b>24A</b>	4.75	0.00
<b>24B</b>	4.75	0.06
<b>25</b>	4.67	-0.79

No. of Results	21
Median	4.748
Normalised IQR	0.105
Robust CV	2.20%
Minimum	4.54
Maximum	4.94
Range	0.41

**Z-Score Chart**

**Z-Scores and Summary Results****Organism 3 (dominant unicellular motile algal genus; Order Volvocales)****Sample Pair B & C**Accepted genus: *Chlamydomonas*

Lab Code	Log10 Sample B	Log10 Sample C	Between Laboratories Z-Score	Within Laboratory Z-Score
<b>1*</b>				
<b>2*</b>				
<b>3</b>	3.50	3.46	0.00	0.00
<b>4</b>	4.96	4.88	11.81 §	0.48
<b>5*</b>				
<b>6</b>	3.37	3.48	-0.44	-2.00
<b>7</b>	3.64	3.52	0.84	0.98
<b>8</b>	3.59	3.47	0.46	1.06
<b>11*</b>				
<b>12</b>	3.49	3.44	-0.12	0.14
<b>14</b>	3.42	3.39	-0.60	-0.13
<b>15</b>	3.43	3.32	-0.85	0.96
<b>16*</b>				
<b>19</b>	3.83	3.86	2.98	-0.98
<b>20*</b>				
<b>21</b>	3.57	3.59	0.83	-0.87
<b>22*</b>				
<b>23</b>	3.35	3.27	-1.34	0.50
<b>24A</b>	3.41	3.41	-0.52	-0.57
<b>24B</b>	3.48	3.51	0.14	-0.85
<b>25*</b>				

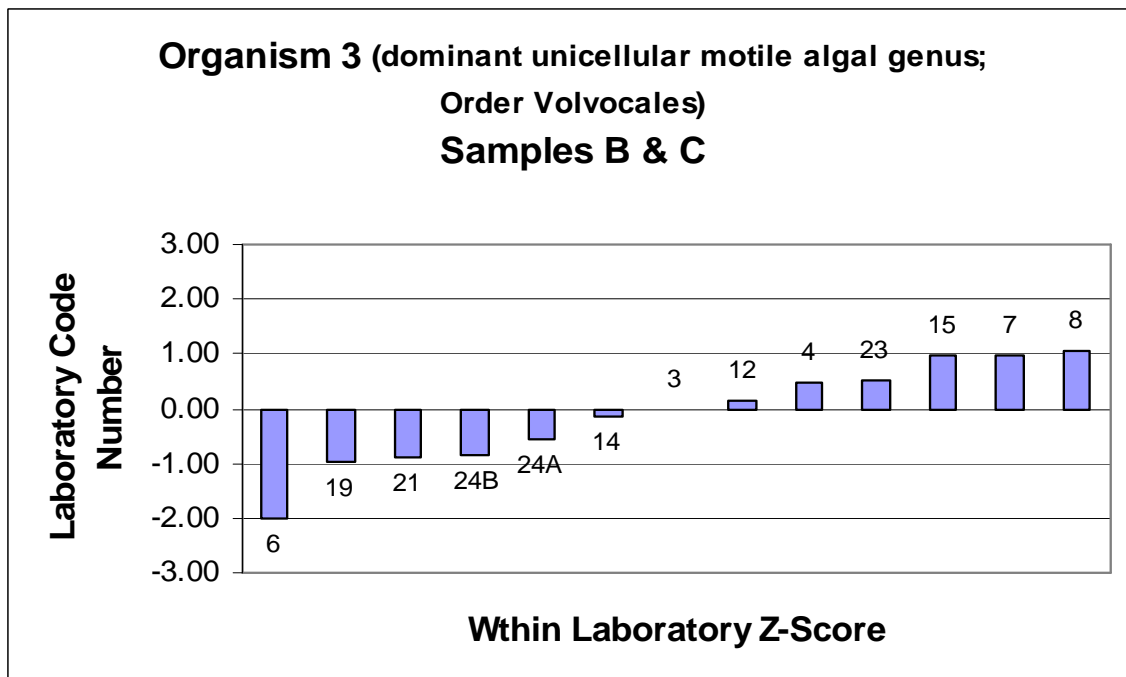
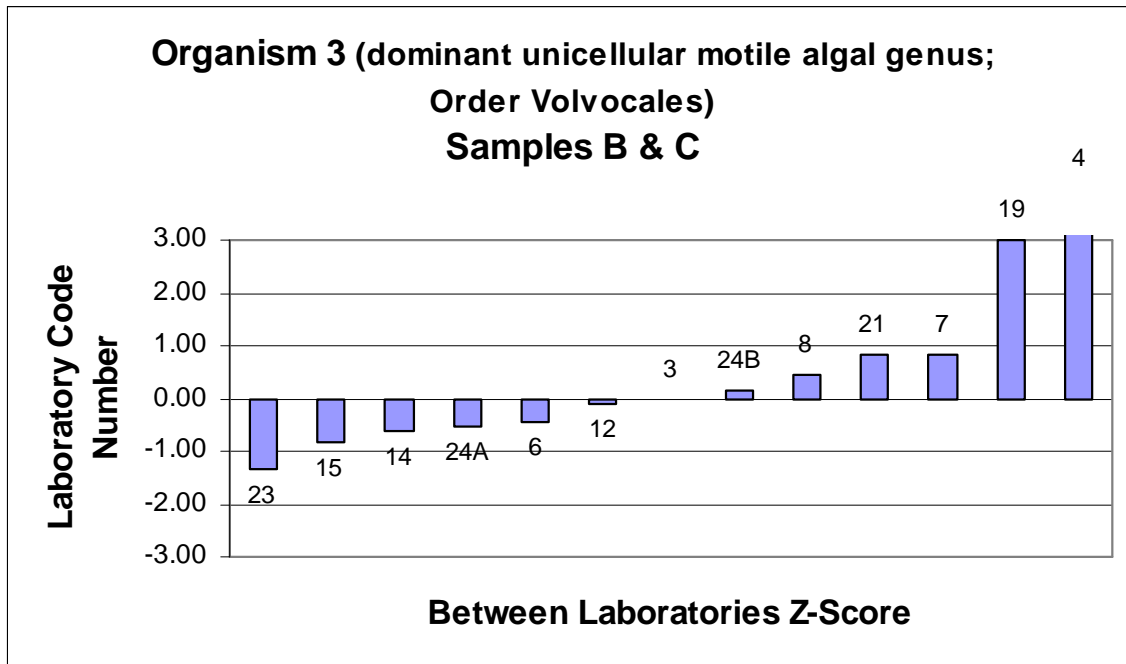
No. of Results	13	13
Median	3.490	3.473
Normalised IQR	0.129	0.080
Robust CV	3.69%	2.30%
Minimum	3.35	3.27
Maximum	4.96	4.88
Range	1.60	1.61

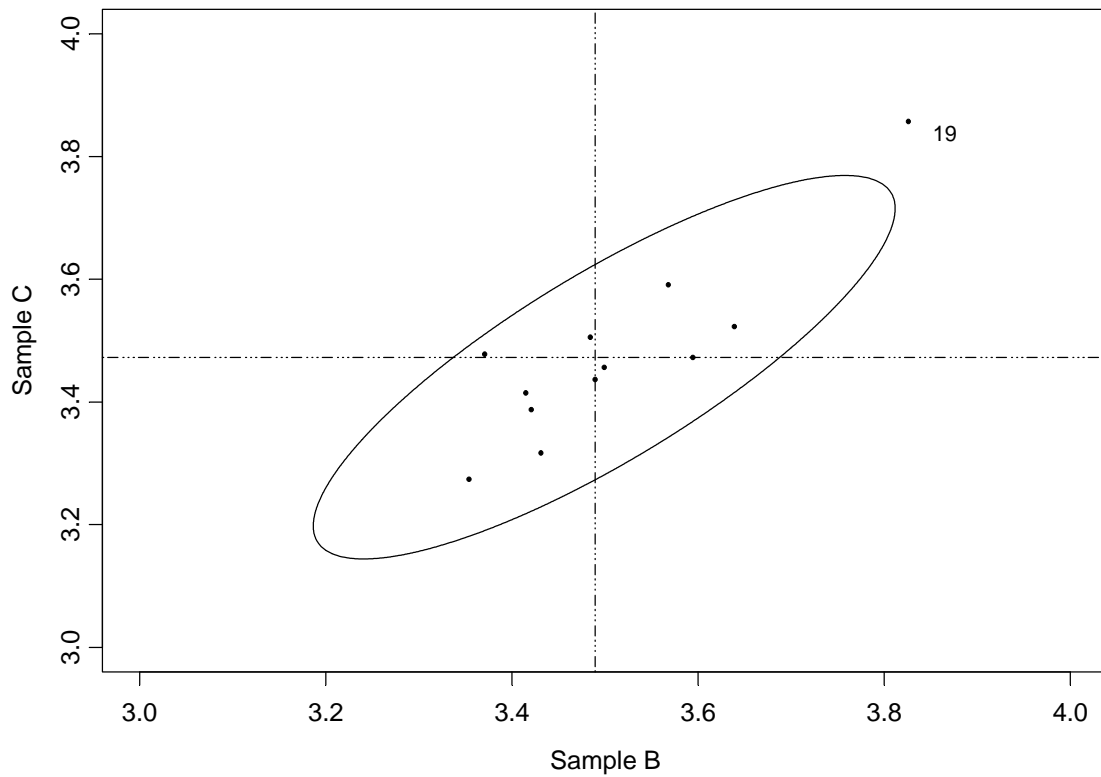
**Notes:**

§ denotes an outlier

\*Lab codes 2, 5, 11, 16, 20 & 25 enumerated *Volvox*; lab code 1 enumerated *Stenophora*; and lab code 22 enumerated *Lobomonas*. These mis-identifications have been classed as extreme results and so not included in the analysis.



**Z-Score Charts**

**Youden Diagram – Organism 3****Organism 3****Notes:**

The following results were excluded from the Youden diagram:

<u>Code</u>	<u>Sample B</u>	<u>Sample C</u>	<u>Between Z-score</u>	<u>Within Z-score</u>
4	4.959041	4.880814	11.81382	0.476677

The highlighted lab code which is outside the ellipse obtained a z-score greater than 2 or less than -2.

**Z-Scores and Summary Results**  
**Organism 3 (dominant unicellular motile algal genus; Order Volvocales)**  
**Sample A**

Accepted genus: *Chlamydomonas*

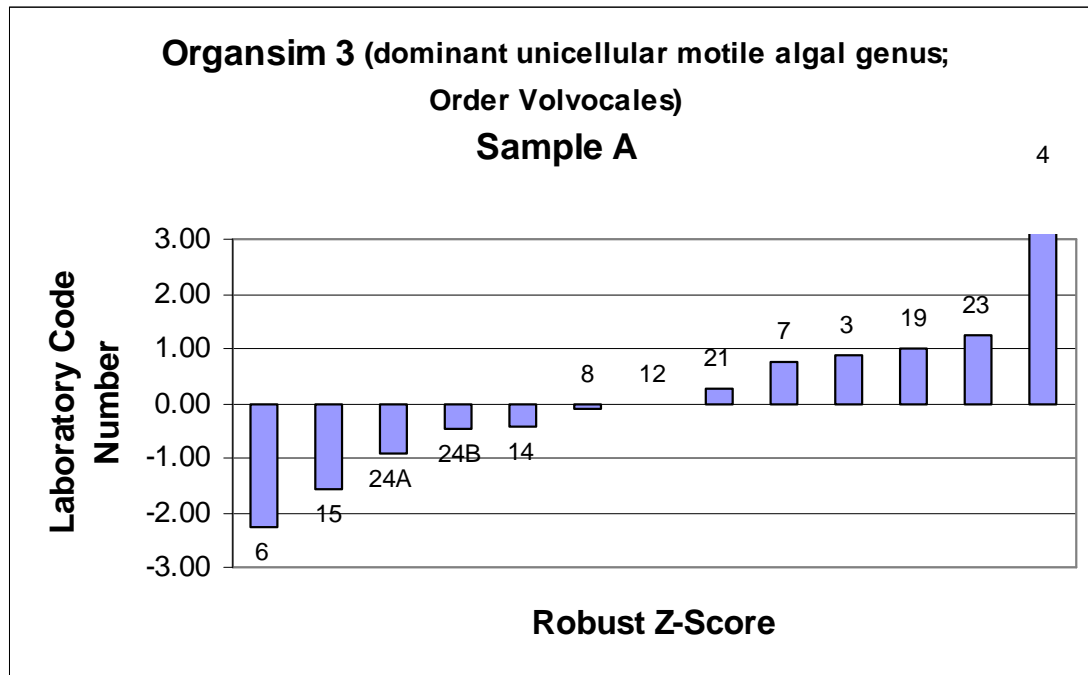
Lab Code	Log10 Sample A	Robust Z-Score
<b>1*</b>		
<b>2*</b>		
<b>3</b>	3.81	0.88
<b>4</b>	5.01	7.28 §
<b>5*</b>		
<b>6</b>	3.23	-2.27
<b>7</b>	3.79	0.74
<b>8</b>	3.63	-0.09
<b>11*</b>		
<b>12</b>	3.65	0.00
<b>14</b>	3.57	-0.43
<b>15</b>	3.36	-1.58
<b>16*</b>		
<b>19</b>	3.84	1.01
<b>20*</b>		
<b>21</b>	3.70	0.26
<b>22*</b>		
<b>23</b>	3.88	1.23
<b>24A</b>	3.48	-0.93
<b>24B</b>	3.56	-0.46
<b>25*</b>		

No. of Results	13
Median	3.650
Normalised IQR	0.186
Robust CV	5.10%
Minimum	3.23
Maximum	5.01
Range	1.78

**Notes:**

§ denotes an outlier

\*Lab codes 2, 5, 11, 16, 20 & 25 enumerated *Volvox*; lab code 1 enumerated *Stenophora*; and lab code 22 enumerated *Lobomonus*. These have been classed as mis-identifications and so not included in the analysis.

**Z-Score Chart**

**Z-Scores and Summary Results**  
**Organism 4 (dominant cyanobacterial genus; Order Oscillatoriales)**  
**Sample Pair B & C**

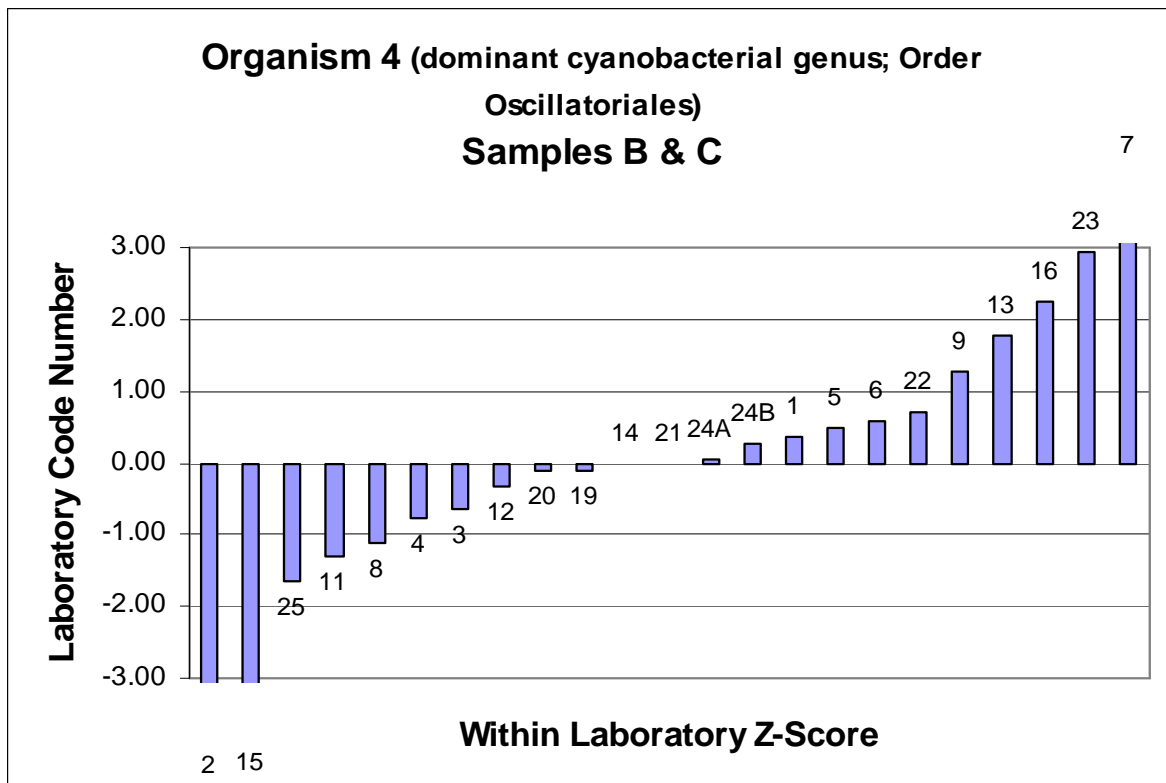
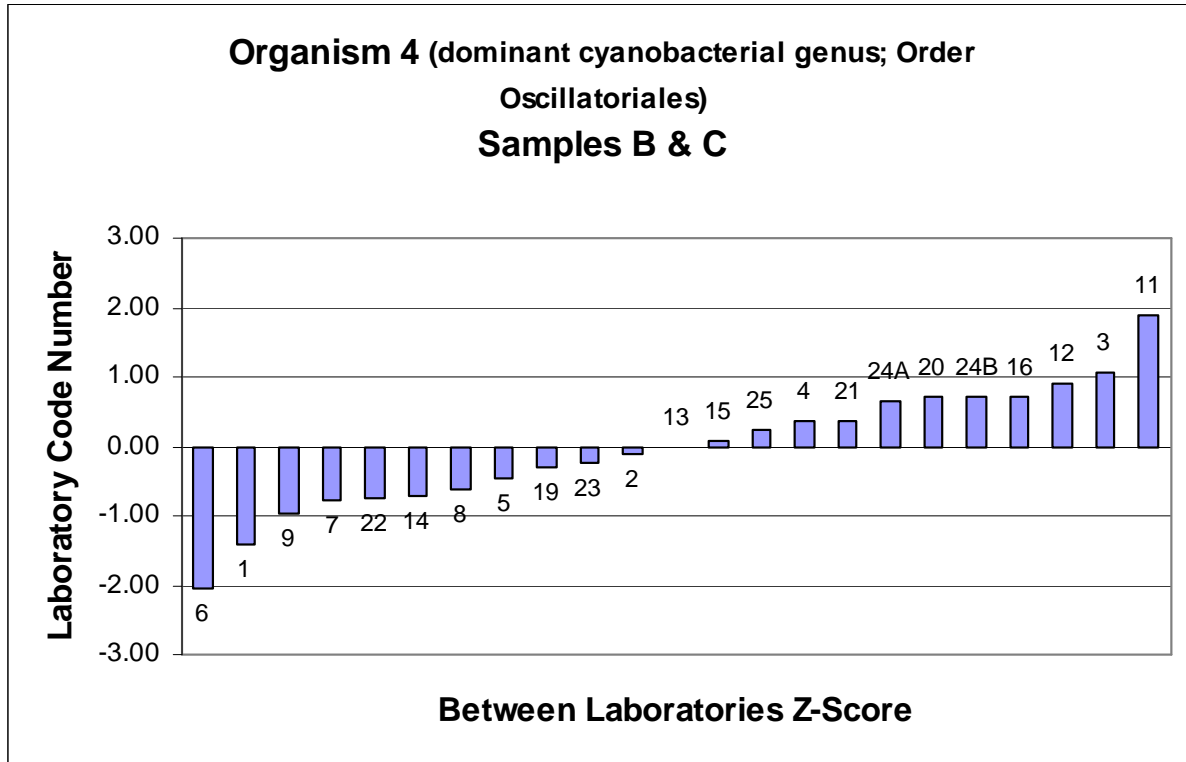
Accepted genus: *Arthrospira*

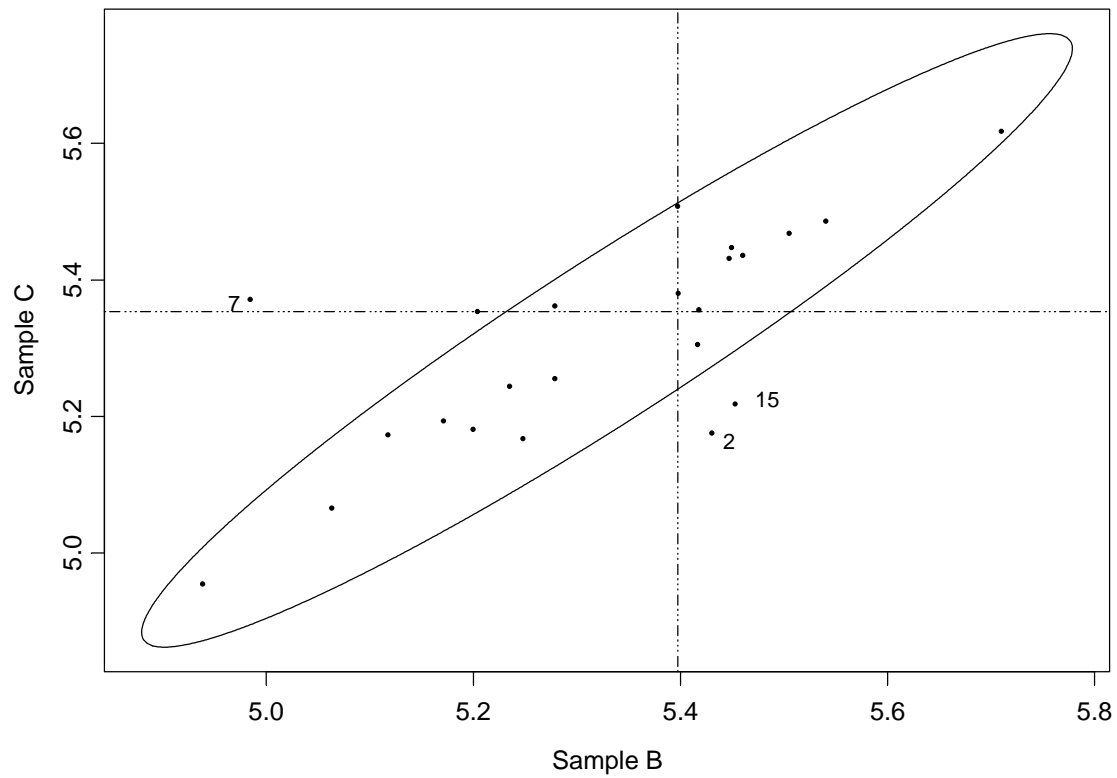
Lab Code	Log10 Sample B	Log10 Sample C	Between Laboratories Z-Score	Within Laboratory Z-Score
1	5.06	5.07	-1.41	0.35
2	5.43	5.18	-0.10	-4.16 §
3	5.54	5.49	1.06	-0.65
4	5.42	5.36	0.37	-0.77
5	5.23	5.24	-0.44	0.47
6	4.94	4.95	-2.06	0.59
7	4.98	5.37	-0.78	7.08 §
8	5.25	5.17	-0.62	-1.10
9	5.12	5.17	-0.96	1.28
11	5.71	5.62	1.89	-1.31
12	5.50	5.47	0.92	-0.34
13	5.28	5.36	0.00	1.76
14	5.20	5.18	-0.72	-0.02
15	5.45	5.22	0.08	-3.80 §
16	5.40	5.51	0.73	2.24
19	5.28	5.26	-0.29	-0.10
20	5.46	5.44	0.70	-0.12
21	5.40	5.38	0.38	0.00
22	5.17	5.19	-0.76	0.70
23	5.20	5.35	-0.23	2.92
24A	5.45	5.43	0.66	0.03
24B	5.45	5.45	0.71	0.27
25	5.42	5.31	0.22	-1.64

No. of Results	23	23
Median	5.397	5.353
Normalised IQR	0.183	0.183
Robust CV	3.38%	3.41%
Minimum	4.94	4.95
Maximum	5.71	5.62
Range	0.77	0.66

**Notes:**

§ denotes an outlier

**Z-Score Charts**

**Youden Diagram****Organism 4****Notes:**

The following results are highlighted as outliers:

<u>Code</u>	<u>Sample B</u>	<u>Sample C</u>	<u>Between Z-score</u>	<u>Within Z-score</u>
2	5.430469	5.175318	-0.09566	4.15672
7	4.984676	5.371437	-0.78405	-7.0817
15	5.452936	5.217931	0.083778	3.804004

**Z-Scores and Summary Results**  
**Organism 4 (dominant cyanobacterial genus; Order Oscillatoriales)**  
**Sample A**

Accepted genus: *Arthrospira*

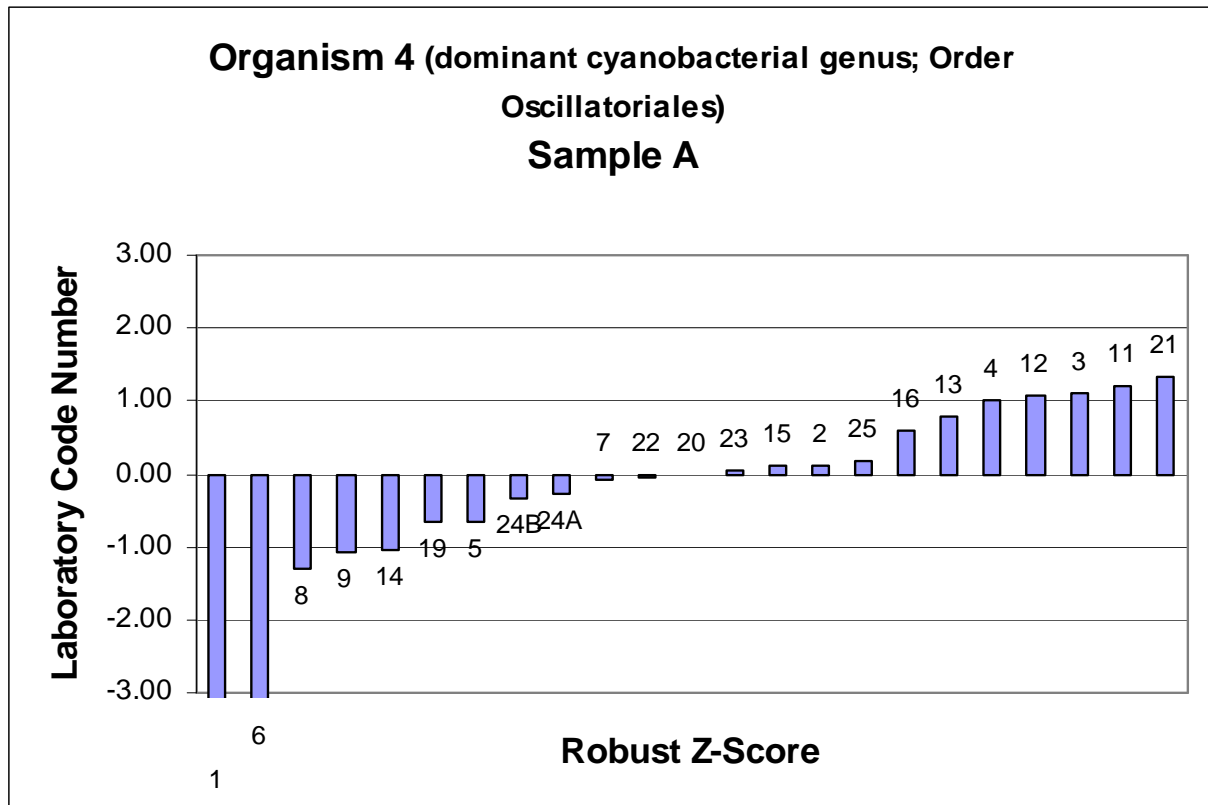
Lab Code	Log10 Sample A	Robust Z-Score
<b>1</b>	5.01	-3.83 §
<b>2</b>	5.59	0.12
<b>3</b>	5.74	1.12
<b>4</b>	5.72	1.00
<b>5</b>	5.48	-0.65
<b>6</b>	5.10	-3.22 §
<b>7</b>	5.56	-0.08
<b>8</b>	5.38	-1.31
<b>9</b>	5.41	-1.08
<b>11</b>	5.75	1.19
<b>12</b>	5.73	1.06
<b>13</b>	5.69	0.80
<b>14</b>	5.42	-1.06
<b>15</b>	5.59	0.10
<b>16</b>	5.66	0.61
<b>19</b>	5.48	-0.65
<b>20</b>	5.57	0.00
<b>21</b>	5.77	1.34
<b>22</b>	5.56	-0.06
<b>23</b>	5.58	0.04
<b>24A</b>	5.53	-0.28
<b>24B</b>	5.53	-0.32
<b>25</b>	5.60	0.19

No. of Results	23
Median	5.573
Normalised IQR	0.147
Robust CV	2.65%
Minimum	5.01
Maximum	5.77
Range	0.76

Notes:

§ denotes an outlier



**Z-Score Charts**

## **APPENDIX B**

**Sample Preparation and Distribution**

**and**

**Homogeneity Testing**

## **SAMPLE PREPARATION AND DISTRIBUTION**

The samples utilised in this program were prepared by the Australian Water Quality Centre (SA).

The test sample was made to 2.5 L in volume and was mixed by a paddle stirrer in a square perspex box for 30 minutes. Subsamples of 20mL volume were then dispensed from the tap into amber glass bottles. A square box was used rather than a flask to prevent a vortex effect when mixing.

Each participant was provided with three sample bottles, labelled Sample A, Sample B and Sample C. The samples were delivered to participants on the week ending 28 October 2005 by overnight prepaid courier.

## **HOMOGENEITY TESTING**

For this program, 8 bottles of Sample B/C and 8 of Sample A were randomly selected and tested for homogeneity.

### **Samples B & C (Blind duplicates)**

Replicate	<i>Arthrospira</i>	Log10	<i>Oocystis</i>	Log10	<i>Mougeotia</i>	Log10	<i>Chlamydomonas</i>	Log10
1	307,030	5.487	14,700	4.167	65,000	4.813	1,817	3.259
2	304,520	5.484	11,500	4.061	55,533	4.745	1,850	3.267
3	324,650	5.511	13,150	4.119	61,314	4.788	1,767	3.247
4	349,820	5.544	11,950	4.077	64,667	4.811	1,900	3.279
5	327,170	5.515	9,700	3.987	74,768	4.874	1,575	3.197
6	402,670	5.605	14,800	4.170	65,867	4.819	1,212	3.084
7	347,300	5.541	11,400	4.057	77,572	4.890	1,325	3.122
8	359,880	5.556	12,900	4.111	64,350	4.809	1,225	3.088

Mean	5.530	4.094	4.818	3.193
SD	0.040	0.061	0.046	0.083
CV	0.7%	1.5%	1.0%	2.6%

### **Sample A**

Replicate	<i>Arthrospira</i>	Log10	<i>Oocystis</i>	Log10	<i>Mougeotia</i>	Log10	<i>Chlamydomonas</i>	Log10
1	639,925	5.806	9,950	3.998	61,750	4.791	2550	3.407
2	622,050	5.794	10,350	4.015	57,600	4.760	2575	3.411
3	593,450	5.773	11,300	4.053	52,000	4.716	2,675	3.427
4	582,725	5.765	11,950	4.077	59,200	4.772	2,425	3.385
5	643,500	5.809	11,300	4.053	64,071	4.807	2,500	3.398
6	593,450	5.773	10,800	4.033	63,000	4.799	2,425	3.385
7	668,525	5.825	10,650	4.027	57,750	4.762	2,475	3.394
8	736,450	5.867	10,700	4.029	57,000	4.756	2,550	3.407

Mean	5.802	4.036	4.770	3.402
SD	0.034	0.025	0.029	0.014
CV	0.6%	0.6%	0.6%	0.4%

**STABILITY TESTING**

Three samples of B/C and three samples of A were randomly selected and tested for stability twice. The first was at the time most labs tested (1/11/05) and the second time was when results were due (8/11/05). All three samples were deemed to be stable during the testing period.

**Samples B & C (Blind duplicates) 1/11/05**

Replicate	<i>Arthrospira</i>	Log10	<i>Oocystis</i>	Log10	<i>Mougeotia</i>	Log10	<i>Chlamydomonas</i>	Log10
1	322,500	5.509	7,300	3.863	57,085	4.757	1,425	3.154
2	437,500	5.641	8,000	3.903	51,775	4.714	1,130	3.053
3	377,500	5.577	8,600	3.934	49,030	4.690	1,340	3.127

Mean	5.575	3.900	4.720	3.111
SD	0.066	0.036	0.033	0.052
CV	1.2%	0.9%	0.7%	1.7%

**Sample A 1/11/05**

Replicate	<i>Arthrospira</i>	Log10	<i>Oocystis</i>	Log10	<i>Mougeotia</i>	Log10	<i>Chlamydomonas</i>	Log10
1	604,450	5.781	9,400	3.973	54,110	4.733	1785	3.252
2	700,700	5.846	10,800	4.033	53,810	4.731	1,260	3.100
3	727,650	5.862	8,000	3.903	52,500	4.720	1,460	3.164

Mean	5.830	3.970	4.728	3.172
SD	0.043	0.065	0.007	0.076
CV	0.7%	1.6%	0.1%	2.4%

**Samples B & C (Blind duplicates) 8/11/05**

Replicate	<i>Arthrospira</i>	Log10	<i>Oocystis</i>	Log10	<i>Mougeotia</i>	Log10	<i>Chlamydomonas</i>	Log10
1	224,250	5.351	9,200	3.964	56,270	4.750	2,750	3.439
2	366,275	5.564	10,600	4.025	63,430	4.802	1,950	3.290
3	361,290	5.558	10,600	4.025	54,970	4.740	1,870	3.272

Mean	5.491	4.005	4.764	3.334
SD	0.121	0.036	0.033	0.092
CV	2.2%	0.9%	0.7%	2.8%

**Sample A 8/11/05**

Replicate	<i>Arthrospira</i>	Log10	<i>Oocystis</i>	Log10	<i>Mougeotia</i>	Log10	<i>Chlamydomonas</i>	Log10
1	598,940	5.777	8,000	3.903	56,570	4.753	2,250	3.352
2	579,510	5.763	8,400	3.924	51,775	4.714	2,550	3.407
3	582,750	5.765	9,600	3.982	50,875	4.707	2,150	3.332

Mean	5.769	3.937	4.724	3.364
SD	0.008	0.041	0.025	0.038
CV	0.1%	1.0%	0.5%	1.1%

## **APPENDIX C**

### **Instructions to Participants & Results Sheet**

**NATA ALGAE PROFICIENCY TESTING PROGRAM - ROUND 9****INSTRUCTIONS TO PARTICIPANTS**

OCTOBER 2005

Participants are asked to carefully note the following **BEFORE** commencing the analysis of the samples.

**1. Samples**

Three samples (labelled Sample A, Sample B and Sample C) have been provided, containing a range of algal and cyanobacterial genera, representing the major algal groups: green algae, diatoms, cyanobacteria (blue-green algae) and phytoflagellates (motiles).

**2. Analysis**

The analysis consists of two parts; (i) Identification, (ii) Enumeration.

**(i) Identification**

Examine all three samples (A, B and C) and identify the algae or cyanobacteria that are present in each, fitting the following criteria.

1. Two (2) genera of green algae that represent the order Chlorococcales; one of the Family Oocystaceae and the other of the Family Dictyosphaeraceae.
2. The dominant green filamentous algal genus.
3. Two (2) dominant genera of motile algae representing the order Volvocales.
4. Seven (7) genera of cyanobacteria representing the orders Nostocales and Oscillatoriales.

**(ii) Enumeration**

Each of the three samples (A, B and C) are to be enumerated. The samples are NOT to be sedimented or otherwise concentrated prior to enumeration.

For each sample, pipette a sub-sample from the bottle and place into a counting chamber. Each sample is to be enumerated using the counting chamber of choice in each laboratory. Results are to be reported for each sample.

An identification and an estimate of cell abundance (reported as cells/mL for each) is required for :

1. The abundant green alga representing the Order Chlorococcales; Family Oocystaceae
2. The dominant (by cell number) filamentous green algal genus.
3. The dominant (by cell number) unicellular motile genus representing the Order Volvocales.
4. The dominant (by cell number) cyanobacterium representing the Order Oscillatoriales.

**Please note** that species differences within the same genus should be disregarded for the purposes of enumeration (ie all morphological forms of a genus should be counted as one).

Participants are requested to perform the analysis according to their routine method. Information on the method used to enumerate each genus or species should be written in the spaces provided on the Results Sheet.

Please note that cells can be counted in either transects (strips), squares or fields of view, whichever is more appropriate, and at a magnification which is appropriate to the cell size and abundance of each genus. An estimate of cells per colony or trichome may be determined if deemed appropriate.

The concentration of each alga or cyanobacterium is to be given as **cells per mL** in the space provided on the Results Sheet.

### 3. Reporting

- (i) Please submit results on the Results Sheet provided.
- (ii) The following information must be recorded on the results sheet:
  - (a) The genera identified.
  - (b) The total magnification used for enumeration of each designated alga.
  - (c) The number of cells, colonies, trichomes enumerated for each genus, depending on which is appropriate.
  - (d) The number of transects, squares or fields of view examined.
  - (e) The type of counting chamber used and its total volume.
  - (f) The method used (if applicable) to estimate cells in colonies and/or trichomes.
  - (g) Any additional method / technique used.

- 4. Since one aim of the program is to obtain an estimate of within-laboratory variation, NATA requests that one analyst performs the enumeration on all samples.

- 5. Testing should commence as soon as possible after receiving samples, and results reported NO LATER THAN **4 November 2005** to:

Mail: Ms Kate Wiggins  
 Proficiency Testing  
 National Association of Testing Authorities, Australia  
 7 Leeds Street  
 RHODES NSW 2138

Phone: (02) 9736 8222 Fax: **(02) 9743 6664** or (02) 9743 5311

- 6. For this program your laboratory has been allocated the following code number [ ]. All reference to your laboratory in reports associated with the program will be through this code number, thus ensuring the confidentiality of your results.

**NATA ALGAE PROFICIENCY TESTING PROGRAM - ROUND 9 (OCTOBER 2005)**

## RESULTS SHEET

Laboratory Code	
-----------------	--

**(i) IDENTIFICATION**

### Sample A

[illegible]

### **Sample B**

[illegible]

### Sample C

[illegible]





**NATA ALGAE PROFICIENCY TESTING PROGRAM - ROUND 9 (OCTOBER 2005)**  
RESULTS SHEET

Laboratory Code

**(ii) ENUMERATION****(a) Organism 1**

(Table for the enumeration of the green alga; Order Chlorococcales; Family Oocystaceae)

Sample	Name of Genus enumerated	Magnification	Total no. of units counted		No. of replicate counts **			Estimate of cells/colony or trichome *	Cells/mL
			cells	trichomes or colonies *	transects	squares	fields of view		
A									
B									
C									

**(b) Organism 2**

(Table for the enumeration of the dominant filamentous green algal genus)

Sample	Name of Genus enumerated	Magnification	Total no. of units counted		No. of replicate counts **			Estimate of cells/colony or trichome *	Cells/mL
			cells	trichomes or colonies *	transects	squares	fields of view		
A									
B									
C									

\* Only complete this column if the method used included an estimation of cells per colony or trichome.

\*\* Enter result for only one column (number of complete transects, squares or fields of view), whichever is appropriate.

**(c) Organism 3****(Table for the enumeration of the dominant unicellular motile algal genus; Order Volvocales)**

Sample	Name of Genus enumerated	Magnification	Total no. of units counted		No. of replicate counts **			Estimate of cells/colony *	Cells/mL
			cells	colonies *	transects	squares	fields of view		
A									
B									
C									

**(d) Organism 4****(Table for the enumeration of the dominant cyanobacterial genus; Order Oscillatoriales)**

Sample	Name of Genus enumerated	Magnification	Total no. of units counted		No. of replicate counts **			Estimate of cells per trichome *	Cells/mL
			cells	trichomes *	transects	squares	fields of view		
A									
B									
C									

\* Only complete this column if the method used included an estimation of cells per colony or trichome.

\*\* Enter result for only one column (number of complete transects, squares or fields of view), whichever is appropriate.

Please confirm the type of chamber used and its volume (mL):

Please provide details of method used (if applicable) to estimate cells in colonies and or trichomes:

Any comments relating specifically to the method used

Date of sample receipt:

Date of Analysis:

Analysts name:

(please print)

Signature:

**Please return results NO LATER THAN 4 November 2005 to:**

Ms Kate Wiggins,

Scientific Officer

Proficiency Testing

National Association Of Testing Authorities, Australia

7 Leeds Street

RHODES NSW 2138

Telephone: (02) 9736 8222

**Fax: (02) 9743 6664 or (02) 9743 5311**

## **APPENDIX D**

### **Z-Score Calculation Parameters**

TABLE D: Z-SCORE CALCULATION PARAMETERS

Test	Sample Pair	Standardised Sum ( <b>S</b> )		Standardised Difference ( <b>D</b> )	
		<i>Median</i>	<i>Normalised IQR</i>	<i>Median</i>	<i>Normalised IQR</i>
Genus 1	B & C	5.943	0.373	0.000	0.032
Genus 2	B & C	6.728	0.090	0.004	0.041
Genus 3	B & C	4.918	0.173	0.030	0.053
Genus 4	B & C	7.524	0.256	-0.013	0.040

**w**                      **x**                      **y**                      **z**

NOTE 1: Each laboratory's between-laboratories z-score (**ZB**) is calculated as follows:

$$\mathbf{ZB} = (\mathbf{S} - \mathbf{w}) / \mathbf{x}$$

Where **S** refers to the standardised sum of the laboratory's pair of results (i.e. Samples B & C), while **w** and **x** refer to the median(**S**) and normalised IQR(**S**) respectively (see above table).

NOTE 2: Each laboratory's within-laboratory z-score (**ZW**) is calculated as follows:

$$\mathbf{ZW} = (\mathbf{D} - \mathbf{y}) / \mathbf{z}$$

Where **D** refers to the standardised difference between the laboratory's pair of results (i.e. Samples B & C), while **y** and **z** refer to the median (**D**) and normalised IQR(**D**) respectively (see above table).