The National Marine Biological Analytical Quality Control Scheme
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Ring Test Bulletin – OMB RT02

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RING TEST DETAILS

Ring Test - OMB 02

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Number of Results Received - 12

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1 Introduction

For a number of years there has been a quality control over the submission of biological data. This now extends through all biological elements including macroalgae. This ensures consistency of data being reported for management purposes and has been primarily driven by international analytical standards due to the Water Framework Directive. The QC scheme aims to facilitate improvements in biological assessment whilst maintaining the standard of marine biological data. The scheme is able to ensure consistency between laboratories and field staff with improved confidence in ecological quality status.

The National Marine Biological Analytical Quality Control (NMBAQC) Scheme addresses one main issue relating to macroalgae data collection:

- The determination of algal biomass

This is the second year in which biomass of macroalgae has been included as an element of the NMBAQC scheme and was included a single exercise. Test material was distributed to participating laboratories from which data forms were completed with algal biomass results and returned.

Twelve laboratories completed the macroalgae biomass component of the NMBAQC scheme. All of the participating laboratories were government; no private consultancy took part in this particular exercise.

Due to the limited number of samples distributed only a single set of results was permitted per Laboratory. It was possible for each sample to be completed by a different participant, however, this was not recorded within the final results.

Currently this scheme does not provide a means of qualifying performance levels. It offers a means of assessing personal and laboratory performance from which continued training requirements may be identified or from which improvements in current field and laboratory procedures may be addressed. Certain targets have been applied to the assessment of the results based on Z-scores allowing “Pass” or “Fail” flags to be assigned accordingly; however, these have no weighting and merely act to identify those results which were considered significantly different based on comparisons between laboratories. These flags have no current bearing on the acceptability of data from such participating laboratories.

1.1 Summary of Performance.

This report presents the findings of the macroalgae biomass component for the second year of operation within the National Marine Biological Analytical Quality Control (NMBAQC) Scheme. This component consisted of a single exercise producing a single set of results from each laboratory.

The results for each for the exercise are presented and discussed with comments provided on the overall participant performance.

2 Summary of Macroalgae Biomass Component

2.1 Introduction

There was one exercise for the assessment of biomass of macroalgae which took the form of three representative artificial samples. This exercise is described in full below to include details of distribution and logistics, procedures for determination of biomass, completion of test result forms and full analysis and comparison of final submitted results.

In order to assess the accuracy of determining biomass of opportunistic macroalgae, samples were supplied that consisted of j-cloth and wool material that had been cut and finely shredded in order to mimic species of *Ulva* (and *Enteromorpha*). Three representative samples were been supplied to be processed. (It was not considered practicable to obtain reliably replicate samples of natural material). Sediment and debris commonly found within areas of algal growth were mixed into the samples with small amounts of water. For each sample wet weight and dry weight had to be calculated.

2.2 Logistics

Each sample was distributed within an airtight plastic container. Each sample within the container was separately sealed within a ziplock plastic bag to retain moisture. The samples were distributed either via first class mail or recorded delivery, depending upon personal requirements. All instructions and additional test material was distributed on CD, within the parcel, to each laboratory. Each disc contained description of methods and data submission forms. Participants were provided a month to complete the test and return the results. Only one set of results could be submitted from each laboratory although it was possible to have up to three participants complete the sample analysis.

Email has been the primary means of communication for all participating laboratories subsequent to the initial postal distribution of test material.

2.3 Methods

Three samples were provided and labelled from A to C. Identical weights were provided for all participants.

Sample A – 42g

Sample B – 66g

Sample C – 23g

Due to the nature of the samples they could be kept for several days retaining most of the moisture. However, much of the water was removed prior to distribution to reduce weight during transport therefore it was necessary to add additional water to each of the samples prior to commencement of the tests to enable rehydration of the material and aid with rinsing.

2.3.1 Method for Wet Weight

The laboratory instructions stipulated that each of the samples required rinsing free of all sediment. The samples were fully washed in a bucket to ensure no loss of sample until the water ran clear and all debris was removed. Once the samples were adequately washed they were squeezed of excess water. This was achieved by hand using samples no larger than the size of a tennis ball to ensure it fit in the palm of the hand and be properly squeezed. Where the sample was large it was divided into smaller clumps for squeezing. The samples were squeezed until no additional running water could be removed by hand (over-enthusiastic squeezing of actual algal samples might damage cell membranes and lose 'genuine' weight). At this stage the whole sample was weighed on a calibrated balance to two decimal places. The exact method used for rinsing and squeezing should be consistent with that used in the field which may vary between laboratories.

2.3.2 Method for Dry Weight

Once each of the samples had been wet weighed they were laid and spread out on a sorting tray or similar container. By spreading the samples this aided with the drying process. The samples were left to air dry for 24 hours. The samples were checked regularly and the drying/weighing process was continued until constant mass was achieved. The unchanged dry weight was the final weight to be used.

2.4 Analysis and Data Submissions

Spreadsheet based forms were distributed with the test material to standardise the format in which the results were submitted. These results will be retained and stored appropriately. Each participant was required to submit a dry weight and a wet weight for each of the 3 samples provided.

2.5 Confidentiality

To preserve the confidentiality of participating laboratories, each participant is allocated a four digit laboratory code from which they can identify their results. These codes are randomly assigned. The initial letters (MA) refer to the scheme, this is followed by the scheme year which refers to the year in which the NMBAQC scheme original commenced, and the final two digits represent the laboratory. For example, laboratory twelve in scheme year eighteen will be recorded as MA1812.

2.5 Results

The results have been collated and represented in various formats to enable full comparisons between participants and samples. These are detailed in the following section with a summary of these results provided at the end of the report.

The raw data shows the range of results submitted from each laboratory with details of the range of values (Table 1). The raw data indicates the range of results increased as the sample weight increased, this was evident for both the wet and dry weights. Table 2 provides further evidence of the level of deviation between submitted dry weight results and actual results. For most laboratories the level of deviation was between 0.34 and 7.43. However, one set of results showed significant deviation from actual results indicting some error during processing. This may be due to procedures used, inadequate rinsing or incomplete drying. Although the wet weight from this laboratory did deviate slightly from the average, for which increased pressure during rinsing may rectify this, it was

the dry weight that showed a significant difference in weight compared with the other of the laboratories. It may be concluded from this that the samples were insufficiently dried.

Table 1. Raw Data results from each laboratory including both dry and wet weights.

	Sample A		Sample B		Sample C	
Lab Code	Wet weight	Dry Weight 42g	Wet weight	Dry Weight 66g	Wet weight	Dry Weight 23g
MA1830	171	115	259	201	129	92
MA1817	141		207		68	
MA1831	124.5	47.1	193.1	72.7	71.8	25.4
MA1806	150.9	44.28	216.28	69.16	78.62	23.9
MA1810	131.5	46.0	194.0	70.3	68.0	25.0
MA1811	124.21	45.35	195.95	73.43	67.85	24.63
MA1818	116.32	43.45	180.53	66.34	59.02	23.48
MA1805	149.1	47.1	232.8	68.6	76.1	23.9
MA1802	146.01	45.75	230	73.07	80.01	24.6
MA1809	134.9	42.9	276.7	70.7	75.6	25.2
MA1801	179.4		259.7		91.9	
MA1803	155.4	39.7	140.8	61.4	80.5	21.6
Max	179.4	115	276.7	201	129	92
Min	116.32	39.7	140.8	61.4	59.02	21.6
Range	63.08	75.3	135.9	139.6	69.98	70.4
Average	143.69	51.66	215.49	82.67	78.87	30.97

Table 2. Deviation of dry weight results from actual dry weight including average deviation per laboratory.

	Sample A	Sample B	Sample C	
Lab Code	Dry Weight 42	Dry Weight 66	Dry Weight 23	
MA1730	73	135	69	92.33333333
MA1717				
MA1731	5.1	6.7	2.4	4.733333333
MA1706	2.28	3.16	0.9	2.113333333
MA1710	4	4.3	2	3.433333333
MA1711	3.35	7.43	1.63	4.136666667
MA1718	1.45	0.34	0.48	0.756666667
MA1705	5.1	2.6	0.9	2.866666667
MA1702	3.75	7.07	1.6	4.14
MA1709	0.9	4.7	2.2	2.6
MA1801				
MA1803	-2.3	-4.6	-1.4	-2.766666667

Z-scores were calculated to indicate how much each participant's weight results deviated from the mean. It uses the following formula:

$$Z = \frac{X - \mu}{\delta}$$

A Z-score of greater than +/- 2.0 was considered to be outside an acceptable limit of deviation from the mean. This value was assigned a 'Fail' or 'Pass' flag on the data (Table 3). In total five results were flagged as 'Fail', most z-scores were within +/- 1.0. The full range of Z-score results can be seen in Figure 1.

A second Z-score was calculated based on deviation from the actual known dry weight using the same criteria to flag 'Pass' and 'Fail'. Table 4 indicates a total of three 'Fails'. The Z-scores are relatively low for all bar one laboratory, the high dry weight values submitted has produced a higher standard deviation for the population mean and has prevented any small deviation from the actual weight becoming evident in this analysis.

Table 3. Z-scores for wet and dry weight based on the mean per sample.

	Sample A						Sample B						Sample C					
Lab Code	WW	Z-score	Flag	DW (42g)	Z-score	Flag	WW	Z-score	Flag	DW (66g)	Z-score	Flag	WW	Z-score	Flag	DW (23g)	Z-score	Flag
MA1730	171	1.989	Pass	115	2.661	Fail	259	1.302	Pass	201	2.663	Fail	129	2.689	Fail	92	2.666	Fail
MA1717	141	0.128	Pass				207	-0.371	Pass				68	-0.490	Pass			
MA1731	124.5	-0.896	Pass	47.1	-0.253	Pass	193.1	-0.818	Pass	72.7	-0.283	Pass	71.8	-0.292	Pass	25.4	-0.294	Pass
MA1706	150.9	0.742	Pass	44.28	-0.374	Pass	216.3	-0.073	Pass	69.16	-0.364	Pass	78.62	0.064	Pass	23.9	-0.360	Pass
MA1710	131.5	-0.462	Pass	46.0	-0.300	Pass	194.0	-0.789	Pass	70.3	-0.338	Pass	68.0	-0.490	Pass	25.0	-0.312	Pass
MA1711	124.21	-0.914	Pass	45.35	-0.328	Pass	196	-0.727	Pass	73.43	-0.266	Pass	67.85	-0.498	Pass	24.63	-0.328	Pass
MA1718	116.32	-1.404	Pass	43.45	-0.410	Pass	180.5	-1.223	Pass	66.34	-0.429	Pass	59.02	-0.958	Pass	23.48	-0.379	Pass
MA1705	149.1	0.630	Pass	47.1	-0.253	Pass	232.8	0.459	Pass	68.6	-0.377	Pass	76.1	-0.068	Pass	23.9	-0.360	Pass
MA1702	146.01	0.438	Pass	45.75	-0.311	Pass	230	0.369	Pass	73.07	-0.275	Pass	80.01	0.136	Pass	24.6	-0.329	Pass
MA1709	134.9	-0.251	Pass	42.9	-0.433	Pass	276.7	1.871	Pass	70.7	-0.329	Pass	75.6	-0.094	Pass	25.2	-0.303	Pass
MA1801	179.4	2.510	Fail				259.7	1.324	Pass				91.9	0.756	Pass			
MA1803	155.4	1.021	Pass	39.7	-0.570	Pass	140.8	-2.501	Fail	61.4	-0.543	Pass	80.5	0.162	Pass	21.6	-0.463	Pass
Mean	138.94			52.99			218.54			85.03			77.40			32.01		
StDev	16.115			23.300			31.086			43.549			19.186			22.505		

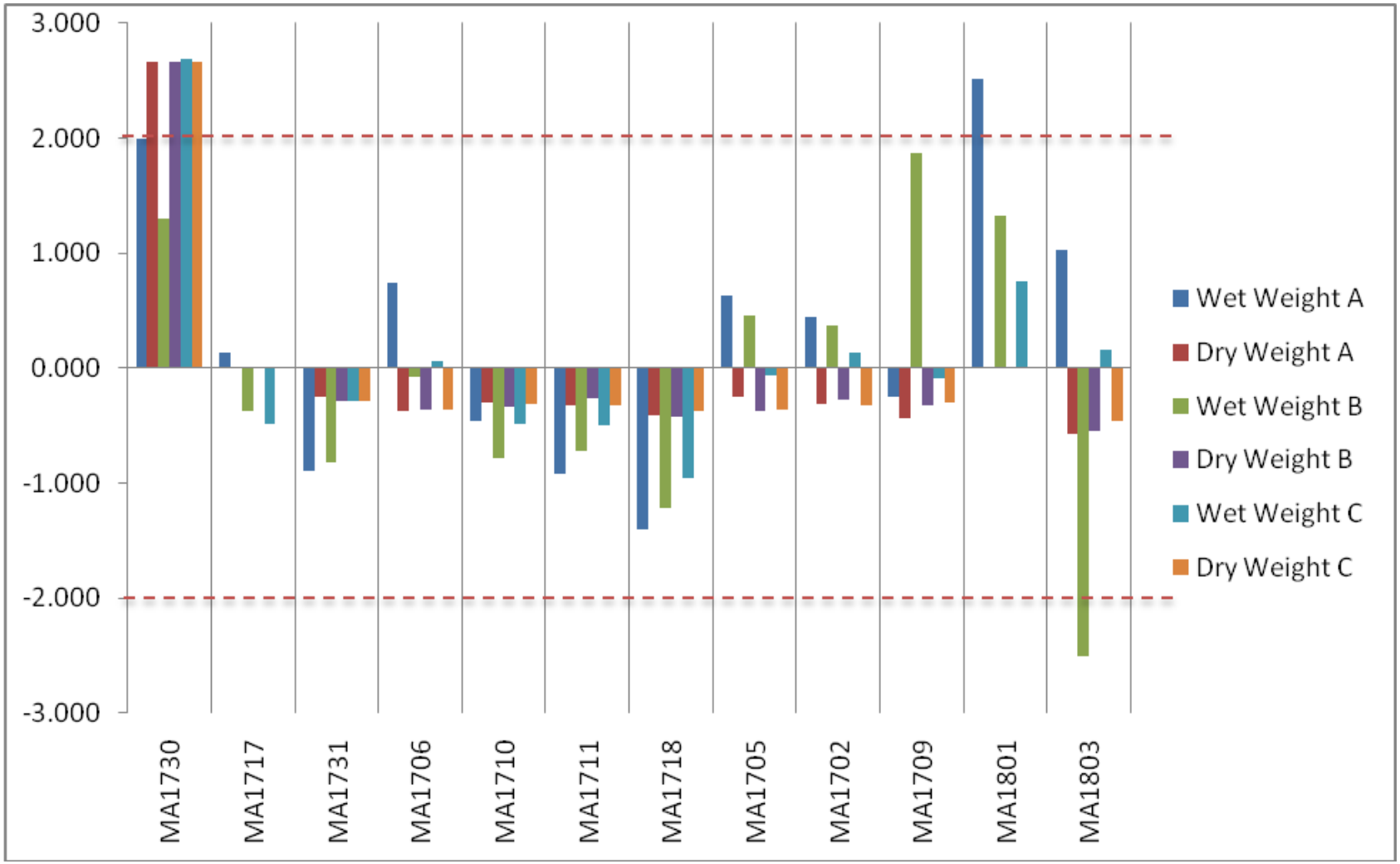

Figure 1. Graph of Z-scores based on deviation from mean

Table 4. Z-scores for dry weight based on the actual weight per sample.

	Sample A			Sample B			Sample C		
Lab Code	DW (42g)	Z-score	Flag	DW (66g)	Z-score	Flag	DW (23g)	Z-score	Flag
MA1730	115	3.133	Fail	201	3.100	Fail	92	3.066	Fail
MA1717									
MA1731	47.1	0.219	Pass	72.7	0.154	Pass	25.4	0.107	Pass
MA1706	44.28	0.098	Pass	69.16	0.073	Pass	23.9	0.040	Pass
MA1710	46.0	0.172	Pass	70.3	0.099	Pass	25.0	0.089	Pass
MA1711	45.35	0.144	Pass	73.43	0.171	Pass	24.63	0.072	Pass
MA1718	43.45	0.062	Pass	66.34	0.008	Pass	23.48	0.021	Pass
MA1705	47.1	0.219	Pass	68.6	0.060	Pass	23.9	0.040	Pass
MA1702	45.75	0.161	Pass	73.07	0.162	Pass	24.6	0.071	Pass
MA1709	42.9	0.039	Pass	70.7	0.108	Pass	25.2	0.098	Pass
MA1801									
MA1803	39.7	-0.099	Pass	61.4	-0.106	Pass	21.6	-0.062	Pass
Mean	52.99			85.03			32.01		
StDev	23.300			43.549			22.505		

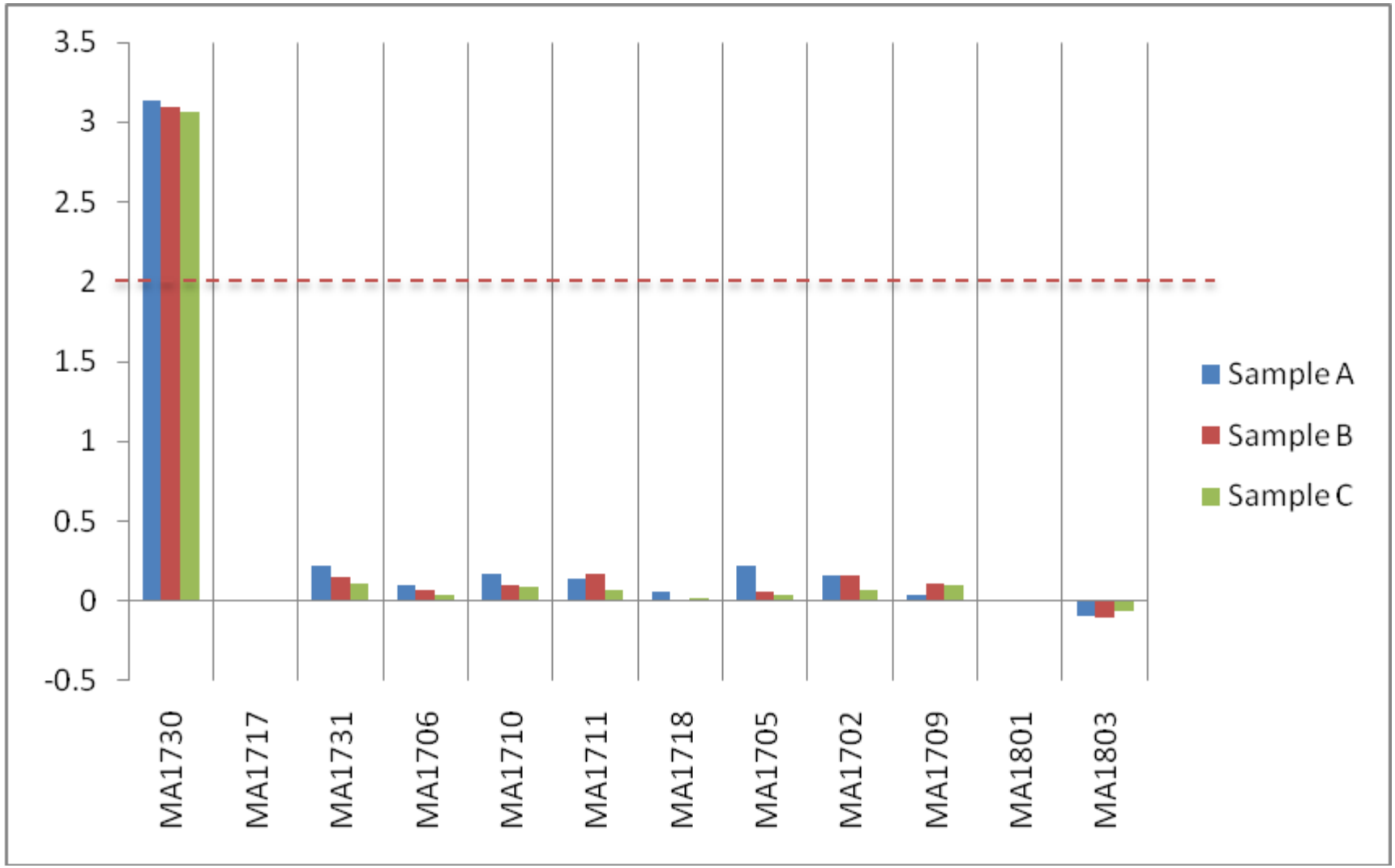

Figure 2. Graph of Z-scores based on deviation from actual weight

3 Conclusions and Recommendations

Excluding the extreme outliers produced by one laboratory, the results do indicate a much higher level of accuracy associated with dry weight than wet weight. This suggests the techniques used between laboratories to rinse and squeeze vary considerable and may also do so between participants within the same laboratory. The wet weights appear to be highly depended upon the participant involved with the processing. There may be future requirements to include biomass analysis within a workshop to further discuss processing procedures and levels of intensity for manual removal of debris and water.

Most laboratories produced a dry weight greater than that of the actual result, this would be due to insufficient drying or rinsing of the sample. However, one laboratory produced dry weights less than that of the actual result which suggests possible loss of material during the rinsing processing. The significant deviation in results from one laboratory from both the mean and actual weight produced an exceptionally high standard deviation making it impossible for the analysis to pick up any smaller deviations from mean and actual without removing the outlier.

It is appreciated that the use of artificial material to mimic opportunist algal species does not fully represent the conditions experienced within the field. However, in order to assimilate an exercise within which a standard sample weight can be distributed, there are few alternatives. Each sample is required to have identical dry weight free of any additional debris prior to distribution. On arrival to the participating laboratory, assurance needs to be provided that the sample will not be degrade in any way and will be identical to both its distribution state and to other samples distributed at the same time. This is necessary for results to be accurately compared both against other laboratories and against the original weight. It may be possible in the future to utilise alternative materials that may be more representative of the texture and general nature of opportunist but at this stage alternative materials have not been tested with the same success rate.

During this second cycle of the macroalgae biomass scheme there were slow and missing returns from some laboratories which lead to some delays in processing and subsequent reporting and feedback of results. In subsequent years reminders will be distributed prior to the completion deadline for the exercise.

Two laboratories only completed the wet weight of samples. Although many in house field procedures do not incorporate dry weight of algal samples these values are included within NMBAQC scheme to enable analysis of laboratory procedures. The values provide evidence of insufficient rinsing of samples, whereby the dry would be considerably higher than the actual weight. Therefore it is recommended that the test be completed fully.