The National Marine Biological Analytical Quality Control Scheme
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Macroalgae Identification Component Report – RM RT07 2013 Year 19

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April 2013
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ALGAL COMPONENT REPORT FROM THE CONTRACTOR SCHEME OPERATION – YEAR 19 - 2013

1 Introduction

To enable correct water quality classification and good management decision-making, quality control of biological data is a high priority. This extends through all biological elements including macroalgae and seagrass. Good quality control ensures consistency of data being reported for management purposes, and for macroalgae and marine angiosperms this has been driven primarily by the requirements of the Water Framework Directive. This QC scheme aims to facilitate improvements in biological assessment whilst maintaining the standard of marine biological data. The scheme should help to ensure consistency between analysts with improved confidence in ecological quality status.

The National Marine Biological Analytical Quality Control (NMBAQC) Scheme addresses several issues relating to macroalgae and seagrass data, this report focuses on one of these:

- The identification of macroalgae species

This is the seventh year in which the identification of intertidal macroalgae has been included as an element of the NMBAQC scheme. The test consisted of a single component with the format following that of the sixth year. Test material was labelled and distributed to participating laboratories using previously employed procedures, from which species identification forms were completed and returned for analysis.

Eight laboratories subscribed to the macroalgae ring test with six laboratories submitting results with a total of fourteen participants. One laboratory decided against submitting results due to collaborative efforts and one laboratory failed to submit results; no reasons were provided. Five of the subscribing laboratories were government organisations and three were private consultancies. To ensure consistency between scheme years, each participating laboratory was assigned the same laboratory code as in previous years except where a laboratory was new to the scheme. Individual codes may, however, change slightly due to variations in individual participants. Due to the nature of the exercise there was no limit on the number of participants per lab.

Currently this scheme does not specify a definite qualifying performance level, and NMBAQC ring tests may be treated as training exercises. However, a pass rate of 80% is suggested as a n indicator of good performance, which may be used by competent monitoring authorities for internal monitoring of performance. Ring tests offer 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.

1.1 Summary of Performance.

This report presents the findings of the macroalgae identification component for the seventh year of operation within the National Marine Biological Analytical Quality Control (NMBAQC) Scheme. This component consisted of a single macroalgae exercise the analytical procedures of which remained consistent with round six of the scheme (RM RT06). The results for the exercise are presented and discussed with comments provided on the overall participant performance.

A macroalgae ring test of twenty macroalgae specimens was distributed to the eight subscribing laboratories. Round seven of the ring test produced an acceptable degree of agreement between identification made by participating laboratories and initial identification as made by Wells Marine. However, the ring test incorporated more challenging species than in previous tests resulting in a greater degree of conflicting results.

2 Summary of Macroalgae Component

2.1 Introduction

There was one module for the macroalgae identification component for scheme year seven. This module is described in full below to include details of distribution and logistics, completion of test result forms and full analysis and comparison of final submitted results.

2.2.1 Logistics

The test material was distributed on CD to each laboratory with labelling and distribution procedures following those of previous years. Each disc contained the full identification module including photos and habitat details from which to identify specimens, description of methods and data submission forms. Participants were primarily given a month to complete the test and return the results, however this was extended to 6 weeks due to late interest and subscription to the test. There were no restrictions on the number of participants per laboratory.

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

2.2.2 Analysis and Data Submissions

A prepared results sheet was distributed with the exercise instructions to standardise the format in which the results were submitted as per previous years. All returned data was done so in Excel and has been stored and analysed in this format. In this and previous scheme years slow or missing returns for exercises lead to delays in data processing data, reporting and feedback of results, therefore reminders were distributed shortly before the exercise deadline.

2.2.3 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, the final two digits represent the laboratory. For those laboratories where multiple submissions were provided the four digit code is followed by a letter allocated to each participant of that laboratory. For example, participant c from laboratory twelve in scheme year twenty will be recorded as MA2012c.

2.3 Macroalgae Ring Test (RM RT07) Module

2.3.1 Description

This training module enables the inter-laboratory comparisons of participants' ability to correctly identify macroalgae taxa and whether errors may be attributed to inadequate keys, lack of reference material or incorrect use of satisfactory keys.

One set of photographs for twenty specimen was distributed in January 2013. The specimens included a range of Chlorophyta, Rhodophyta and Phaeophyta and a mix of macroscopic and microscopic specimens from a variety of habitats including epilithic, epiphytic and endozoic species. There were a number of photographs per taxon showing different aspects of the alga and its habitat. Some supplementary information on habitat was included.

2.3.1.1 Preparation of the Sample

Each specimen was to be identified through a number of in-situ, macroscopic and microscopic photographs. In total a minimum of five photographs was used for each specimen collected by Wells Marine for the purpose of this exercise. Specimen photographs were obtained from a range of surveys from around the coast of the UK. Photographs were selected to represent sufficiently each specimen including in-situ (where possible), overall structure, branching patterns, cellular arrangements and cell contents making sure to include key characteristics for accurate identification. Scale bars were included where appropriate. Attempts were also made to ensure a high quality of photographs primarily focusing on clean specimens with sharp photographs.

Using a photographic test is considered a more practical means of testing macroalgal identification skills, than preserved samples,. These are known to lose colour rapidly and cell contents may become distorted making key characteristics more difficult to distinguish. Equally, fresh samples would not last a sufficient period to enable identification.

2.3.1.2 Analysis Required

The participating laboratories were required to identify each of the macroalgae specimens from the photographs provided. Additional information could also be submitted including brief notes, information on keys used or possible problems with identification or quality of photograph provided. If a laboratory was unfamiliar with the specimen then the level of confidence of identification could also be detailed. Participating laboratories were permitted to supply multiple data entries for each exercise to maximise results and allow sufficient comparisons of data entries. The protocol for circulating and completing the module followed that of previous years with four weeks allowed for the identification and submission of species identification results.

2.3.2 Results

2.3.2.1 General Comments

The scheme has taken on the same format as previous years (RT06) this includes the format of the test and method of data analysis and scoring. The macroalgae ring test can act as a training aid in the identification of species allowing those difficult taxa to be revealed and further identifying problematic areas.

For this current round of the scheme (RM RT07) specimen photographs were circulated to a total of eight laboratories. As with previous scheme years, multiple data entries were permitted from each participating laboratory. Six of the eight laboratories returned data entries with a total of fourteen individual data sets.

2.3.2.2 Analysis and Scoring of Data Returns

Laboratories returned lists of their species identifications within the format provided, these were compared against AQC identification as determined by Wells Marine to assess the number of differences. The method of data comparison was achieved by comparing both the genus and species names and identifying where these differed with the AQC names. Such comparison included differences in spelling or use of a valid synonym for example:

- Use of different synonym for a taxon, e.g. *Audouinella purpurea* for *Rhodochorton purpureum*
- Mis-spelling of taxa name, e.g. *Rhodomela lycopodioides* for *Rhodomela Lycopoides*

Such differences were not taken into account during calculation of the total number of differences in identification.

Data entries were tabulated (as seen in RM RT07 Preliminary Results Bulletin, Table 2) in order of specimen number and laboratory. The individuals' data entries are only given where they differ from the AQC identification. This includes those entries for which species are incorrectly spelled or where an appropriate synonym is provided as well as those instances in which the specimen has been identified incorrectly. For those entries in which a synonym or mis-spelling was supplied by the participant but for which the identification was consistent with that of the AQC, the name was presented in brackets [species name]. Those entries in which the identification was considered different to the AQC the species or genus name that did not correspond to the AQC was provided in the table. If part or the entire species name entered was correct this was indicated by a dash "-" any incorrect name was included in the table e.g. where *Ceramium virgatum* was identified as *Ceramium secundatum* this would be entered as " – *secundatum*". Further exceptions were granted to *Ceramium virgatum*. Due to the lack of distinguishing features between *Ceramium virgatum* and *Ceramium botrycarpum* both species were accepted for the identification of species RT0714.

The data entries were scored by increasing the score of the individual by one where the entry was consistent with that of the AQC. For instance where text other than a dash "-" or a bracketed name [name] is provided no score was given. This includes differences at both genus and species level, although these can be considered independent values it is often the case that where the generic identification was incorrect then the species identification would usually also be incorrect. Therefore where the full genus and species name was correct a score of two would be given where either genus or species name was incorrect a score of one would be given. The method of scoring applied to those species in which a correct identification was provided and included those instances where synonyms were used or species/genus names incorrectly spelled.

2.3.2.3 Ring Test Results

Results were forwarded to each of the participating laboratories four weeks after data submission. These results are documented in the preliminary results bulletin (RM RT07) which detailed individual scores and highlighted incorrect identifications, mis-spellings and use of synonyms. The bulletin also outlined reasons for identification discrepancies by comparing incorrect species and genus names with those of the AQC with the aid of photographs to pick out key characteristics.

RM RT07 contained twenty specimens for identification for which there was a good general level of agreement through all fourteen participants. At the generic level there were a total of thirty five differences (from a potential two hundred and eighty) across the fourteen sets of data received from the six participating laboratories. At the specific level, agreement was also considered good with a total of fifty six differences. These differences could be attributed to just a few taxa. A total of 28% of all errors were from one species (*Callithamnion tetragonum*) contributing to 37% of all generic differences and 23% of all specific differences. *Cladophora laetevirens* contributed to a further 11% of differences with ten specific errors. A further five specimens contributed individually between 7% and 9% of both generic and specific differences attributing to 40% of overall errors (*Elachista scutulata, Lomentaria clavellosa, Chordaria flagelliformis, Chorda filum* and *Callithamnion tetricum*). A final seven taxa were responsible for the remaining thirteen specific and six generic errors (*Furcellaria lumbricalis, Halopteris filicina, Rhizoclonium riparium, Boergeseniella fruticulosa, Rhodomela lycopodioides, Ceramium virgatum* and *Rhodochorton purpureum*). The remaining six species received no generic or specific identification errors.

The difference between participants' entries and AQC identifications was generally well distributed with all participants identifying at least one genera and one species incorrectly. The overall scores and number of incorrect identifications ranged from two to fourteen with no one participant identifying all genera and species correctly. At this stage the levels of low, medium and high have not been established for this particular ring test so participants and laboratories cannot be allocated a level of acceptance based on their overall score.

2.4 Discussion

This is the seventh macroalgae identification ring test as circulated through the NMBAQC scheme. Although the results were comparable with those of previous years (RT06) there is a noticeable decrease in the level of agreement between participating laboratories and the AQC. As per last year's suggestions an increased number of cryptic and taxonomically challenging species were included in the test. Such genera included *Cladophora*, *Callithamnion* and *Ceramium* which are difficult to identify to species level. These genera require an increased depth of knowledge on the cellular attributes and other characteristics, which can be remarkably similar between species. As intended by the scheme these tests aim to challenge participants and assist with training by stimulating the use of various keys and increasing familiarity with taxonomic terminology. Further, it allows problem taxa to be identified stimulating areas for inclusion in workshops, and targeting such taxa within future exercises. Photographs used within the ring tests may be retained within the participating laboratories for future reference, with some descriptions allowing the comparison of taxonomically similar species.

No laboratory or individual managed to identify all species and genera correctly and there were only 6 species for which all laboratories were successful in their identification (Table 1 and Figure 1). The two most problematic species were *Callithamnion tetragonum* and *Cladophora laetevirens* both of which are considered relatively difficult to identify due to lack of conclusive characteristics and the occurrence of morphologically similar species. Those characteristics which are considered more specific and may be used to distinguish such species were detailed within the Bulletin. The most common error was for *Callithamnion tetragonum,* which in eleven instances was confused with *Aglaothamnion*. The genera are differentiated primarily on the number of nuclei. In contrast *Cladophora* provided problems at the species level with all fourteen laboratories identifying the genus correctly. Although the largest portion of incorrect identifications could be attributed to these two species, there was equally a high number of species for which at least one laboratory identified incorrectly; however, there no specimens which were not identified successfully by at least one laboratory.

Another issue arose with *Ceramium virgatum*. This is a common species, but highly variable morphologically. Similar species have overlapping characteristics, and it was considered that this overlap between *C.virgatum* and *C.botryocarpum* was sufficient to justify accepting both names on this occasion. It is widely recognised that a number of identification works would benefit from up to date revision.

At this time the use of a photographic test is considered the most effective means of testing macroalgal identification skills. Preserved samples are known to rapidly to lose colour with cells becoming distorted making key characteristics more difficult to distinguish. Equally, fresh samples would not last a sufficient period to enable identification. However, it is possible that some photographs were not considered to be of sufficient quality to correctly identify the specimens despite all efforts. This may have attributed to some confusion over the identification of some more cryptic species.

Table 1: Summary of Differences

Specimen	Genera	Species	Total differences for 14 returns	
			Genus	Species
RT0701	*Saccorhiza*	*polyschides*	0	0
RT0702	*Hypoglossum*	*hypoglossoides*	0	0
RT0703	*Cladophora*	*rupestris*	0	0
RT0704	*Furcellaria*	*lumbricalis*	1	1
RT0705	*Elachista*	*scutulata*	2	6
RT0706	*Lomentaria*	*clavellosa*	3	3
RT0707	*Halopteris*	*filicine*	1	1
RT0708	*Callithamnion*	*tetragonum*	13	13
RT0709	*Bifurcaria*	*bifurcata*	0	0
RT0710	*Rhizoclonium*	*riparium*	0	2
RT0711	*Plumaria*	*plumosa*	0	0
RT0712	*Boergeseniella*	*fruticulosa*	1	2
RT0713	*Rhodomela*	*lycopodioides*	1	1
RT0714	*Ceramium*	*virgatum*	0	3
RT0715	*Chordaria*	*flagelliformis*	3	3
RT0716	*Chorda*	*filum*	4	4
RT0717	*Cladophora*	*laetevirens*	0	10
RT0718	*Callithamnion*	*tetricum*	4	4
RT0719	*Rhodochorton*	*purpureum*	2	3
RT0720	*Asparagopsis*	*armata*	0	0
		Total differences	35	56
		Average differences per species	1.750	2.800

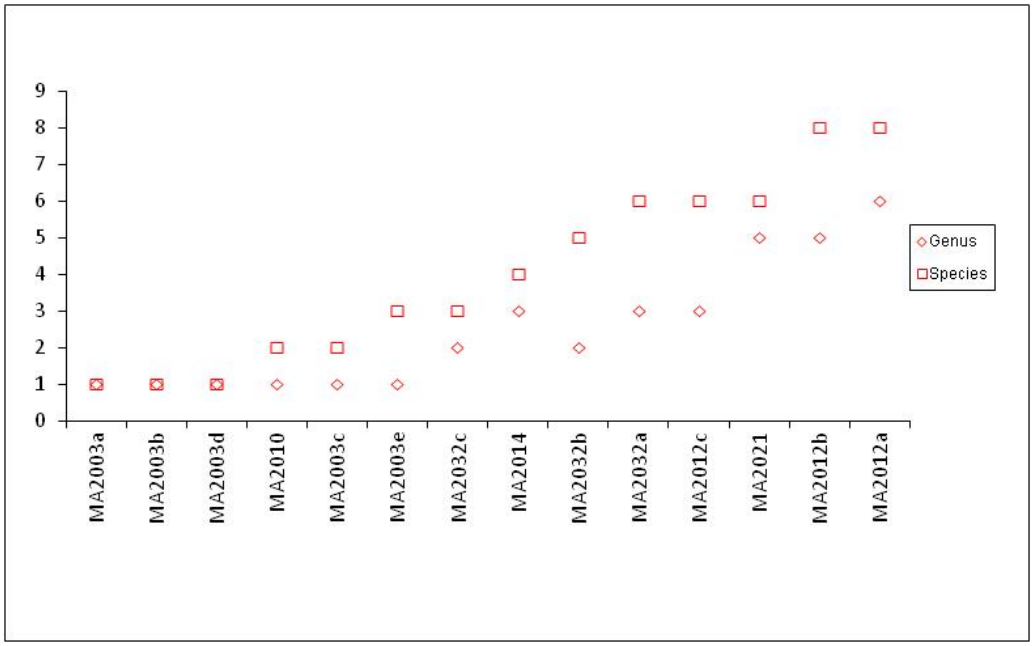

Figure 1: The number of differences from the AQC identification of intertidal macroalgae specimens, for each of the participating laboratories for RT07, arranged in order of increasing number of differences.

3 Conclusions and Recommendations

1. The seventh macroalgae ring test exercise was successfully implemented and completed by most participants with a general agreement of the format. All feedback has been reviewed and will be considered for subsequent exercises, such feedback is encouraged to enable the protocols to be refined.

2. The good level of agreement within this test provides evidence that macroalgae identification skills are increasing, however there are still a number of problematic areas. This is to be expected, as some taxa are inherently more difficult than others. The errors occurring were generally at the specific level, however where generic errors occurred these were most often with taxonomically similar species which share similar characteristics and are therefore hard to separate. Such species will be noted for possible future workshops and will be targetted in future exercises.

3. There were still a number of incorrect spellings; therefore more care should be taken prior to submitting results to ensure all species are spelled correctly. This is equally important when submitting data records or reports where scientific names are incorporated.

4. As with some previous tests there was some conflict as to the correct identification of some species. Descriptions of some species have recently changed some of which have resulted in nomenclatural changes or use of more specific characteristics that were previously considered more generic. New studies in species taxonomy are regularly highlighting previously unidentified species, splitting one species into two based on a previously unknown characteristic. In these instances both species identification have been accepted. More specifically within the test there has been conflict over the acceptance of *C. botrycarpum* for species RT0714 (*Ceramium virgatum).* Keying out the two species shows very little difference except for the actual forms of the species, i.e. turf and tuft, which may not be wholly clear in the photos (no sizes given). As with the overlap of habitat and morphological form there can also be overlap of cell sizes depending on where in the plant the measurement is taken. Much of this can be quite subjective, especially where descriptions of cell size are very broad, varying considerably along the frond of the plant (i.e. from base to tip). Given the number of participants identifying No.14 as *C. botrycarpum*, and the overlap of criteria, a decision was made to accept both identifications. This highlights the need for more definitive photos and descriptions to be provided in future exercises so as to save confusion.

5. All laboratories are encouraged to keep all test photographs within a reference collection. This has a number of benefits particularly with regards to improving identification ability, training new staff and maintaining consistency of identification between surveys and staff. This reference collection should also be extended through to literature to ensure current keys are used and up to date nomenclature. A list of identification works will be given on the NMBAQC website. However, this is not exhaustive, and does not necessarily include unpublished keys provided at workshops unless specifically authorised by the key's author.

6. During this seventh cycle of the macroalgae identification exercise all participants submitted results within the designated (extended) timescale. However, one laboratory was unable to submit their results within the designated time period due to late subscription requiring a 2 week extension deadline for all laboratories. In future exercises all laboratories should continue to submit results within the requested deadlines as detailed at the beginning of the exercise. In

subsequent years reminders will continue to be distributed prior to the completion of the exercise.

7. There are still some issues over the timing of the test and there are suggestions that the time allowed for completion of the test should be extended to accommodate increased workloads. Although this is still the most appropriate time of year to complete the tests, a longer time scale within which to complete the exercises would allow more laboratories to complete the exercise in full. Six weeks has been suggested as a more appropriate time scale in which to successfully complete the ring test and this shall be considered for future years.

8. Although there was general approval on the quality, detail and use of photographs with most participants agreeing on the levels of difficulty, there were some areas which require some improvement. In some instances the *in situ* specimen photographs would have benefitted further from a scale and additional details of habitat, general location, exposure of shore, height present on shore etc than were provided. This additional information will be included in subsequent tests to allow more accurate identification and to reduce error or confusion. Some more specific cellular information was also requested within the photos, and where possible this will be achieved. However, even when looking at fresh specimens not all such characteristics may be present, e.g. reproductive structures. All attempts will be made in the future to ensure that sufficient material is provided, allowing correct identification to species level.

If anyone has further comments on this, or disagrees with any of the interpretation, please pass forward your comments to Dr Emma Wells (emma@wellsmarine.org) or Dr Clare Scanlan (clare.scanlan@sepa.org.uk). This ring test is continually being refined to ensure it provides the best opportunity to test macroalgae identification skills so all suggestions and comments are welcomed.

4. References

Bunker, F.StP.D., Brodie, J.A., Maggs, C.A. & Bunker, A.R. (2010). *Seasearch guide to seaweeds of Britain and Ireland*. pp. [1] 5-224, many colour photographs. Ross-on-Wye: Marine Conservation Society.

Brodie, J. & Irvine, L.M., 2003. Seaweeds of the British Isles. Volume 1. Rhodophyta. Part 3B. Bangiophycidae. Pp. i-xiii, 1 – 167, map. Andover: Intercept.

Dixon, P.S. and Irvine, L.M., 1977. Seaweeds of the British Isles. Vol. 1 Rhodophyta. Part 1. Introduction, Nemaliales, Gigartinales. British Museum (Natural History), London.

Guiry, M.D., 1997. Benthic red, brown and green algae. In: Howson, C.M. and Picton, B.E. The species directory of the marine fauna and flora of the British Isles and surrounding seas. The Ulster Museum and the Marine Conservation Society, Belfast and Ross-on-Wye.

Hiscock, S., 1979. A field guide to the British brown seaweeds (Phaeophyta). Field Studies. **5**, 1 – 44.

Hiscock, S., 1986. A Field Guide to the British Red Seaweeds (Rhodophyta). Field Studies Council Occasional Publications no. 13.

Irvine, L.M., 1983. Seaweeds of the British Isles. Vol. 1 Rhodophyta. Part 2a. Cryptonemiales (sensu stricto), Palmariales, Rhodymeniales. British Museum (Natural History), London.

Irvine, L.M. and Chamberlain, Y., 1993. Seaweeds of the British Isles. Vol. 1. Rhodophyta. Part 2b. Corallinales. British Museum (Natural History), London.

Kornmann, P. and Sahling, P.H., 1983. Meeresalgen von Helgoland: Erganzung. Helgoländer Wissenschaftliche. Meeresuntersuchungen. **36**, 1 – 65.

Maggs, C.A. and Hommersand, M., 1993. Seaweeds of the British Isles. Vol. 1. Rhodophyta. Part 3a. Ceramiales.

Wells, E. 2007. Water Framework Directive – coastal water rocky shore monitoring: Field guide to British seaweeds. Environment Agency, Bristol.