

NE ATLANTIC MARINE BIOLOGICAL ANALYTICAL QUALITY CONTROL SCHEME

Annual Report 2020/2021

A report prepared by the NMBAQC Scheme Coordinating Committee March 2022

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This Annual Report provides synopsis of the scheme year's activities over 2020/2021, the 27th year of the NMBAQC scheme. Detailed information about each of the scheme components is now available as separate reports or bulletins on the scheme's website. The relevant documents are all cited here and the reader is directed via hyperlinks to the NMBAQC website as appropriate.

The NMBAQC Scheme is jointly run by academic, advisory, commercial, conservation and regulatory bodies of the UK and Ireland. As the current scheme treasurers, the Environment Agency wishes to acknowledge the financial assistance of JNCC Support Co. Representatives from these agencies and competent monitoring authorities (CMAs) for the NMBAQC coordinating committee.

The NMBAQC coordinating committee held one meeting during the 2020-2021 reporting period. This was on the 6th October 2020; subsequent meetings will be covered in the next Annual Report. Minutes of this meeting are on the NMBAQC website: <u>http://www.nmbaqcs.org/reports/</u>

Committee Membership for 2020/2021 is shown in Appendix 1.

1 Scheme Review

The scope of the NMBAQC scheme continued to develop in 2020/2021 to encompass the requirement to provide quality assurance for assessments under the Water Framework Directive (WFD), for which monitoring commenced in the UK in 2007. The scheme still maintains its role to provide Analytical Quality Control for Invertebrate and Particle Size data collected for the UK CSEMP (Clean Seas Environment Monitoring Programme). Under the UK Marine Monitoring and Assessment Strategy (UKMMAS) the NMBAQC scheme coordinating committee reports to the Healthy and Biologically Diverse Seas Evidence Group (HBDSEG).

Where possible other components followed a similar format to the previous year and involved training and testing exercises for the Invertebrate, Particle Size, Fish, and

Macroalgae components. The Zooplankton component is held every two years with the last ring test undertaken during the autumn of 2021.

The 2020-2021 participation level in the NMBAQC scheme was slightly lower than the previous year. This was due to the ongoing Covid-19 pandemic. (See Appendix 2)

Summaries of all the component activities are provided in this document.

2 Invertebrate component

Contract Manager: Myles O'Reilly, Scottish Environment Protection Agency. Component Administrator: David Hall, APEM Ltd.

2.1 Summary of activities

Scheme year 2020/ 2021 (year 27) followed the format of year 2019 / 2020. A series of components, modules and exercises involved the distribution of test materials to participating laboratories and the centralised examination of returned data and samples. The labelling and distribution procedures employed previously have been maintained except for a modification to accommodate disruption caused by the Covid-19 pandemic. Specific details can be found in previous Scheme annual reports.

Forty-two laboratories (with multiple participants from some organizations counted separately) participated in the Benthic Invertebrate Component of the NMBAQC Scheme in 2020 / 2021 (year 27). Thirteen of the participants were UK Competent Monitoring Authorities (CMAs), responsible for the Clean Seas Environment Monitoring Programme (CSEMP) or Water Framework Directive (WFD) sample analysis; nineteen were UK private consultancies. Ten of the participants were non-UK laboratories (including three government organizations and seven private consultancies). Laboratory Codes were assigned in a single series for all laboratories participating in the Benthic Invertebrate component. Separate Laboratory Codes were assigned for the other scheme components, such as the particle size component.

As in previous years, some laboratories elected to be involved in limited aspects of the scheme. UK Competent Monitoring Authorities (CMAs) completing benthic biological analyses for monitoring programmes, including the assessment of MPAs (Marine Protected Areas), as evidence under MSFD (Marine Strategy Framework Directive), WFD (Water Framework Directive) and CSEMP (Clean Seas Environmental Monitoring Programme), must participate in the Benthic Invertebrate component. CSEMP / WFD laboratories are no longer required to participate in all components / modules of the scheme.

This component comprised three modules (each with one or more exercises):

- 1. Own Sample module (OS) re-analysis by APEM Ltd. of three samples supplied by participating laboratories.
- 2. Invertebrate Ring Test module (RT) identification of two sets of twenty-five invertebrate specimens.

3. Laboratory Reference module (LR) - re-identification by APEM Ltd. of a set of up to twenty-five specimens supplied by participating laboratories.

The analytical procedures of the various modules were the same as for 2019 / 2020 (year 26) of the Scheme.

2.2 Summary of results

Two **Ring Tests (RT)**, each of 25 specimens, were distributed (RT59 and RT60). The first (RT59) was a general ring test and the second (RT60) was targeted on biotope-defining species and similar. The methods and policies used in the module followed the Ring Test Protocol (<u>Worsfold & Hall, 2017a</u>).

For RT59, the average numbers of differences per participating laboratory (for a total of 22 laboratories with 18 submissions) were 2.2 generic differences and 4.2 specific differences. Three species (all amphipod crustaceans) were responsible for just over a third (36%) of the specific differences.

For RT60, the average numbers of differences per participating laboratory (for a total of 22 participants with 17 submissions) were 1.4 generic differences and 2.4 specific differences. Five specimens (a sponge, a cnidarian, a mollusc, a bryozoan and an ascidian), were responsible for half (50%) of the specific differences.

Laboratory Reference (LR): Five laboratories signed up for the LR25 module and four laboratories submitted specimens for confirmation. Most misidentifications were for Annelida (82%), followed by Arthropoda (9%). The methods and policies used in the module followed the Laboratory Reference Protocol (<u>Hall & Worsfold, 2017</u>).

The methods and policies used in the **Own Sample (OS)** module followed the <u>Own</u> <u>Sample Exercise Protocol</u> (Worsfold & Hall, 2017b), produced to explain and standardise policies, including details of audit sample selection and determination of 'associated samples' for subsequent remedial actions. Laboratories were asked to submit full completed data matrices from their previous year's CSEMP / WFD, or similar alternative sampling programmes. The OS 'Pass / Fail' flagging system, introduced in Scheme Year 8, was continued (<u>see Hall, 2010: Description of the Scheme Standards for the Benthic</u> <u>Invertebrate Component</u>). In OS74-76, extraction efficiency (of individuals) was better than 90% in 90% of the comparisons and better than 95% in 87% of all comparisons. 100% of countable taxa were extracted from the sample residues in 70% of samples. The Bray-Curtis similarity index ranged from 26% to 100% with an average of 94.8%. The Bray-Curtis similarity index was greater than 95% in 83% of comparisons; in 94% of cases, the value of the index was greater than 90% and, therefore, achieved 'Pass' flags. Six samples (9%) achieved 'Pass- Excellent' flags with Bray-Curtis similarity scores of 100%.

2.3 Issues and recommendations

Several observations may be made from the results of the exercises described above. The following is a summary of the major points of importance:

- The majority of participating laboratories submit data / samples in accordance with the Scheme's timetable. Late submissions, however, are still the major contributing factor for delaying the production of exercise bulletins / reports. Laboratories should endeavour to report their results within the requested time, according to the deadlines circulated at the beginning of each Scheme year.
- 2. The number of samples in data sets provided for selection of Own Samples varied considerably, with several laboratories offering less than the minimum 20 samples for audit selection (due to low volumes of sample processing) and other laboratories offering a full year's benthic data across multiple projects. Best practice for commercial laboratories should be to use the Scheme as an external auditor for most or all of their samples and no 'cherry picking', pre-analysis selection, or pre-submission re- working of samples should be undertaken. Retention of sample residues will be required to facilitate this and to ensure that any subsequent remedial actions can be adequately completed.
- 3. There were continued **problems associated with the measurement of biomass** for individual species in the Own Sample module. In this and previous Scheme years, several laboratories, despite using blotted wet weight biomass techniques, rendered some of their specimens too damaged to be re-identified. Additionally, some laboratories had erroneous results where it appeared that biomass had been estimated or mis-transcribed. The initial processing of a sample should in no way compromise the effectiveness of an audit. Biomass procedures should not render the specimens unidentifiable. Biomass must be reported to four decimal places with nominal weights recorded as 0.0001g. A standardised protocol is available in the NMBAQC guidance document (Worsfold, Hall & O'Reilly (Ed.) 2010) and must be followed for CSEMP / WFD analysis.
- 4. There were some instances (OS & LR modules) of specimens being provided in vials / containers that were not airtight and, as a consequence, specimens were dry and in some case identification was impossible. Participants are reminded that specimens should be stored in suitable air-tight containers so that viability is maintained for the audit process. Participants should also ensure that OS & LR samples are transported to APEM in accordance with the H&S regulations. Participants should use rigid crates when submitting heavy sample residues to prevent damage in transit.
- 5. The maintenance of a comprehensive reference collection has numerous benefits for improving identification ability, maintaining consistency of identification between surveys and access to growth series material. The LR exercise can be used as a means of verifying reference specimens. Laboratories are strongly recommended to **implement and expand in-house reference collections of biota**. The inclusion of growth series material is extremely useful for certain groups, *e.g.* molluscs. All surveys should have an associated reference collection to enable ease of cross-checking or adopting future taxonomic developments.
- 6. Participants submitting data for **laboratory reference exercises should add a note on habitat / location** of samples, to aid identification. A similar 'Habitat

Notes' section to that distributed with the ring test exercises was distributed for completion in this year's exercise and should continue into the next exercise to support AQC identifications.

- 7. Laboratories participating in the ring test exercises should attempt to identify all specimens to species and complete the 'confidence level' section of their ring test datasheets to enable additional information to be gathered regarding the difficulty of ring test specimens.
- 8. There are problems of individuals and taxa missed at the sorting stage of Own Sample analysis. This is an area that is often the major contributing factor in samples with 'Fail' flags or low Bray-Curtis similarity indices. When taxa and individuals are missed during the extraction of biota from the sediment, laboratories should determine why certain taxa have not been extracted. This could be due to the taxon not being recognised as countable, or due to problems with the effect of stains upon the specimens. There may also be a problem within certain taxonomic groups (e.g. crustaceans floating within samples or molluscs settled within the coarser sediment fractions). Additional training may be required and a review of existing extraction techniques and internal quality control measures may be beneficial. Remedial action should concentrate on the specific causes of the failure and should be targeted accordingly e.g. analyst or method related discrepancies.
- 9. It is apparent that some laboratories are not utilizing the NMBAQC guidelines for processing macrobenthic samples (Worsfold, Hall & O'Reilly (Ed.), 2010) issued with MB18 in Scheme Year 17 to improve the consistency of analysis, i.e. all analysts extracting and recording all biota. A detailed taxonomic discrimination policy (TDP) is now under development and will be added to the processing requirement protocol (PRP) to ensure that macrobenthic data from multiple analysts are as consistent and inter-comparable as possible. The Own Sample pass / fail criteria will be reviewed to ensure that they are fit for purpose and uphold data consistency between the Scheme participants.
- 10. Since the beginning of the scheme, continual improvement to the learning structure of the Scheme reports has been maintained. For the LR and OS modules, detailed results have been forwarded as **individual exercise reports** to each participating laboratory as soon after the exercise deadlines as practicable. The **Laboratory Reference Module Summary Reports introduced in 2017** show identification problems found in all LR submissions and should benefit all participants. In the RT module, after each RT exercise a bulletin was circulated, reviewing the literature used, detailing the accepted identification of the taxa circulated, and including images of relevant specimens. Participants are encouraged to review their exercise reports **and provide feedback concerning content and format** wherever appropriate.
- 11. The primary aim of the Benthic Invertebrate Component of the Scheme is to improve the quality of biological data via training and audit modules. An informal constructive reporting system exists to assist in the overall improvement of data quality. For example, laboratories struggling with particular taxonomic groups in

their Own Samples often receive additional support, as well as receiving their returned OS material separated, according to the AQC identifications, for future reference. APEM will continue to proactively follow up outstanding remedial actions from previous scheme years to enable these data to be NMBAQC scheme quality assured. Participants are reminded that completion of remedial action is mandatory for CMA labs and labs submitting data to CMAs. **Participants are encouraged to provide feedback and request further information for any of the scheme exercises to improve the quality and consistency of their data.**

- 12. Additional guidance for Own Sample 'next steps' following audit results has been created to ensure that all participants and other stakeholders are aware of the route to quality assured data (<u>Hall, 2016; Own Sample Interim Report Review</u> and Remedial Action Processes).
- 13. There remain some misconceptions about the nature of the Scheme and the services it provides. It is not an accreditation scheme but provides quality assurance for the UK's CSEMP/WFD programme and other benthic monitoring programmes. In addition, the Scheme can provide **audits of samples** for any marine biological programme or development. It also provides **project-level audits** by applying the OS and LR protocols to examine project data. These services require more extensive communication (Scheme website, information note etc.) to notify all potential users and maintain consistent quality assurance for European marine biological data. A best practice guidance protocol for NMBAQC project-level audits needs to be produced and published on the scheme website. Meanwhile, it should be understood that a project level audit includes a review of data and check of reference collection specimens for the whole project, as well as for selected samples. Audits of samples from a project without more extensive reviews of data and other material do not constitute quality control of the whole project through the Scheme.
- 14. APEM Ltd. strives to ensure smooth running **and transparency of the Scheme** at all times. APEM Ltd. log and make available all correspondence to the Benthic Invertebrate Component Technical Manager (Myles O'Reilly, SEPA). Participants can be assured that their anonymity will be protected if this correspondence is required to be shared with the Committee.
- 2.4 Reports

Benthic Invertebrate Component Annual Report, 2020/2021 (Year 27)

Worsfold, T.M., Hall, D.J., and O'Reilly, M. (Ed.), 2022. Benthic Invertebrate Component Annual Report. Scheme Operation 2020/2021 (Year 27). A report from the contractor to the NMBAQC Scheme co-ordinating committee. 28pp, January 2022

Own Sample Module Summary Report OS74,75,76 – December 2021

Hall, D. 2021. NE Atlantic Marine Biological Analytical Quality Control Scheme. Own Sample Module Summary Report OS74, 75 & 76. Report to the NMBAQC Scheme participants. 17pp, December 2021.

Laboratory Reference Module Summary Report LR25 – June 2021

Worsfold, T., Hall, D. and O'Reilly, M., 2020. NE Atlantic Marine Biological Analytical Quality Control Scheme. Laboratory Reference Module Summary Report LR24. Report to the NMBAQC Scheme participants. 10 pp, April 2020.

RTB60 – March 2020 (Targeted – Biotope-defining species)

Worsfold, T., Hall, D. & Pears, S., 2021. NE Atlantic Marine Biological Analytical Quality Control Scheme. Ring Test Bulletin: RTB#60. Report to the NMBAQC Scheme participants. APEM Report NMBAQC RTB#60, 39pp, Mar, 2021.

RTB59 – Feb 2021 (General/Mixed taxa)

Worsfold, T., Hall, D. & Pears, S., 2021. NE Atlantic Marine Biological Analytical Quality Control Scheme. Ring Test Bulletin: RTB#59. Report to the NMBAQC Scheme participants. APEM Report NMBAQC RTB#97, 40pp, Feb, 2021.

3 Particle Size Analysis component

Contract Manager: Claire Mason, Cefas. Component Administrator: Lydia McIntyre-Brown and David Hall, APEM Ltd.

3.1 Summary of activities

The particle size component of the scheme comprises of two modules:

- 1. The PS Ring Test (PS) analysis of four sediment samples circulated to participant.
- 2. The PS Own Sample (PS-OS) submission of three analysed sediment samples from participant.

The PS module followed the same format of 2019/20; a series of exercises involved the distribution of test materials to participating laboratories and the centralised examination of returned data and samples.

The PS-OS module, introduced in the 2014/15 Scheme year, followed the same logistical format as the previous year. Selected participant samples are re-analysed by the NMBAQC Scheme PSA contractor and the results are compared. The Particle Size Own Sample module is a training / audit module and the purpose of this module is to examine the accuracy of particle size analysis for participants' in-house samples.

Seventeen laboratories signed up to participate in the 2020/21 PS module exercises (<u>PS76</u>, <u>PS77</u>, <u>PS78</u> and <u>PS79</u>); seven were government laboratories and ten were private consultancies. Ten laboratories signed up to participate in the PS-OS module exercises (PS-OS19, PS-OS20 and PS-OS21); seven were government laboratories and three were private consultancies. One government laboratory had four Lab Codes to submit twelve PS-OS samples for AQC analysis.

To reduce potential errors and simplify administration, Lab Codes were assigned with a prefix to determine the Scheme component; all codes for the Particle Size component were prefixed with "PSA_".

As in previous years, some laboratories elected to be involved in limited aspects of the Scheme. Competent monitoring authorities (CMAs) completing PSA in support of biological analysis for monitoring programmes (including in assessment of MPA (Marine Protected Areas), as evidence under MSFD (Marine strategy framework directive) and WFD (Water framework directive), as well as the CSEMP (Clean Seas Environmental Monitoring programme), must participate in this component of the Scheme. The Scheme is aware of other PSA methodologies (*e.g.* those used in the Regional Seabed Monitoring Plan) and encourages those involved in any relevant PSA monitoring programmes to participate in this Scheme, especially where pass/fail criteria can be used to assess overlapping aspects of different methodologies.

Seventeen laboratories subscribed to the exercises in 2020/21. For the first circulation (PS76 and PS77) sixteen subscribing participants provided results; for the second circulation (PS78 and PS79) fifteen participants provided results. PSA_2710 communicated that they were not participating due to Covid-19 restrictions and PSA_2703 communicated that they were not participating as both samples contained a large proportion of gravel and they do not undertake sieve analysis. PSA_2718 appear in PS78, this was a lab participating in a trial to understand whether the scheme was suitable for them and if they would participate in future scheme years.

Most participating laboratories now provide data in the requested format, although some variations remain. As reported previously, it should be remembered that the results presented may be from a more limited number of analytical laboratories than is immediately apparent since this component of the Scheme is often sub-contracted by participants to one of a limited number of specialist laboratories. Detailed results for each exercise (<u>PS76</u>, <u>PS77</u>, <u>PS78</u> and <u>PS79</u>) have been reported to the participating laboratories.

3.3 Conclusions and recommendations

A number of observations may be made based on the results of the exercises described above. The following is a summary of the major points of importance.

1. Laboratories should ensure that they follow the NMBAQC methodology when participating in the Particle Size (PS) Ring Test. The PS Ring Test is designed to test that all participants are getting comparable results when they follow the same methodology. It is therefore important that only the NMBAQC methodology (Mason, 2016) is used where possible and that results for 3 x 3 laser analyses are provided. Participants who do not have access to a laser analyser will be permitted to use alternate methods for samples that contain sediment less than 1mm as long as the method used is detailed in the summary section of the workbook. Participants can choose to opt out of either the sieve or laser aspects if they do not routinely undertake that type of analysis. The participant must let the administrator know at the start of the scheme year if they wish to opt out of any analysis. Results will only be provided for the analysis that was undertaken and a note will be put on the Statement of Performance that the participant has opted out of certain points.

Samples for the PS-OS module can be analysed following alternative in-house methods however, these must be thoroughly described and the participant should be aware that re-analysis will be undertaken following the NMBAQC methodology. Samples provided for PS-OS which have been routinely analysed do not necessarily have to provide 3 x 3 laser analysis data but should show that appropriate QC checks have been carried out, including on the final data set.

- 2. Participants should review their data prior to submission. Errors in datasets can often be spotted in the summary statistics, e.g. percentage gravel, sand and silt/clay, before the data are submitted. All parts of the workbook should be double checked before submission to ensure that they are all filled in correctly. This will help eradicate typing and transcription errors. The workbook has been updated for the next Scheme Year (Year 28) to help enable the continuity of data through the workbook. Conditional formatting will flag up red cells where there are possible data entry errors.
- 3. The current NMBAQC Scheme Pass/Fail criteria for the PS modules are under review. Currently results are broken down for review, including methodology, sieve processing, laser processing, data merging and summary statistics. Laboratories then received a "Good" or "Review" flag based on their results; "Review" flags came with accompanying comments as to where mistakes have been made and how to correct them. This approach was thought to be more informative and would help participants to identify errors and correct any issues for future exercises. Lydia McIntyre-Brown (APEM), PSA component Technical Manager Claire Mason (Cefas) and Jon Barry (Cefas) are currently researching a statistical method to compare participant results with the Benchmark data. Although this year's data is not ready to be trialled yet there is the possibility of a report detailing the outcomes available in the next couple of scheme years.
- Possible workshop looking at sample preparation and presentation to laser. Covid-19 restrictions put an end to any possible face-to-face workshops in 2020-21 (Year 27), but as restrictions ease this may become an option in the next couple of Scheme years.

Most participants now use the recommended laser parameters of an optical model of Mie Theory with Particle Refractive index of 1.55 and a Particle Absorption Index of 0.1; however, the results can still differ from the Benchmark data and other participants. One possible reason for this could be due to sample preparation and homogenisation as well as presentation of the sample to the laser. Another issue that has occurred is whether muddy samples need only laser analysis or whether sieve analysis should be undertaken too. There were incidents where participants recorded less than 1g of sediment greater than 1mm causing sample descriptions to become "slightly gravelly". The NMBAQC guidance states in "5.4.2 Laser diffraction analysis of <1mm sediment fraction" that "...if no sediment >1mm is left on the 1mm mesh [when preparing a laser sub-sample from the bulk], then no further analysis is required". With such small amounts of sediment greater than 1mm found in the entire sample it is unlikely that significant amounts of sediment greater than 1mm were present on the mesh when preparing a laser sub-sample and therefore sieve analysis did not have to be undertaken. A workshop, either in person or a webinar detailing how to create and homogenise a laser sub-sample, particularly looking at the use of ultrasonics may be useful in forth coming years.

- 5. Health and Safety. Recently the presence of asbestos in marine samples has been brought to light. Although safe when the sample is wet, asbestos particles could become air-borne when analysing a particle size sample particularly during the dry sieving process. At the PSA workshop in December 2017, laboratories were informed how to mitigate the hazards associated with analysing samples that may contain asbestos. All the natural material used to create PS ring test samples continues to be sent for presence/absence of asbestos before being distributed to participating laboratories. This will continue for subsequent years and participants can request to see the results of the tests by emailing <u>nmbaqc@apem.co.uk</u>
- 3.4 Reports

PSA Component Annual Report 2020/2021 (Year 27)

McIntyre-Brown, L., Pye, K. and Hall, D. Particle Size Analysis Component Annual Report Scheme Operation 2020/2021 (Year 27). 36pp, August 2021.

PS76 April 2021

McIntyre-Brown, L. & Hall, D., 2021. National Marine Biological Analytical Quality Control Scheme. Particle Size Results: PS76. Report to the NMBAQC Scheme participants. Apem Report NMBAQCps76, April 2021.

PS77 April 2021

McIntyre-Brown, L. & Hall, D., 2021. National Marine Biological Analytical Quality Control Scheme. Particle Size Results: PS77. Report to the NMBAQC Scheme participants. Apem Report NMBAQCps77, April 2021.

PS78 April 2021

McIntyre-Brown, L. & Hall, D., 2021. National Marine Biological Analytical Quality Control Scheme. Particle Size Results: PS78. Report to the NMBAQC Scheme participants. Apem Report NMBAQCps78, April 2021.

PS79 April 2021

McIntyre-Brown, L. & Hall, D., 2021. National Marine Biological Analytical Quality Control Scheme. Particle Size Results: PS79. Report to the NMBAQC Scheme participants. Apem Report NMBAQCps79, April 2021.

4 Fish component

Contract Manager: Jim Ellis, Cefas. Component Administrator: Stephen Duncombe-Smith and David Hall, APEM Ltd.

4.1 Summary of activities

This component consisted of two modules, each with a single exercise:

1. Fish Reverse Ring Test (F_RRT) - Re-identification of a set of up to fifteen fish specimens supplied by each of the participating laboratories.

2. Fish Ring Test (F_RT) - Identification of fifteen fish specimens supplied with images.

Scheme year 2020/2021 (Year 27) followed the format of year 2019, with a ring test (RT) and a reverse ring test (RRT) being organised. The Fish Component of the Scheme is currently in its sixteenth year (start 2005/06). It involved the distribution of test specimens to participating laboratories and the centralised examination of returned data for the first module (RT), and re-analysis of fish specimens submitted by participants for the second exercise (RRT). The labelling and distribution procedures employed previously have been maintained. Specific details can be found in the fish reverse ring test protocol and fish ring test protocol (<u>FRRT Protocol</u> and <u>FRT Protocol</u>).

Fourteen laboratories signed up for Scheme year 2020/2021 (with multiple participants from some organisations counted separately). Ten participants were government laboratories, two private consultancies, one University and one chartered laboratory. Although some fish are sampled under the Clean Seas Environment Monitoring Programme (CSEMP), the number of target species is relatively few. However, the requirement to monitor fish assemblages in transitional waters for the Water Framework Directive (WFD) provides a major impetus for the Fish Component exercise. As in previous years, some laboratories elected to be involved in either one or both exercises of the scheme.

4.2 Summary of results

Fish Reverse Ring Test (F_RRT): The identification of fifteen fish specimens selected and supplied by the nine participating laboratories was very accurate (<u>F_RRT12</u>) (only two taxonomic errors for 131 specimens submitted). Seven participants supplied collection dates for specimens, these were all collected between November and December 2020. Most participants used this as a test for confirming voucher specimens; two participants included a problematic specimen in their submission, misidentification of a problematic specimen is not counted as a taxonomic error.

Fish Ring Test (F_RT): Samples of 15 specimens were distributed <u>(FRT14)</u>. The FRT was not a targeted ring test and most species included are commonly caught in routine monitoring surveys. Some specimens were relatively small but could still be expected to be caught using standard monitoring methods (e.g. seine netting).

For <u>FRT14</u>, the average numbers of differences per participating laboratory (for a total of 8 laboratories with 10 submissions) were 0.8 generic differences (5%) and 1.3 specific differences (9%). Three families (Gobiidae, Clupeidae and Ammodytidae) were responsible for 7 of the 8 generic errors and 12 of the 13 specific errors.

4.3 Issues and recommendations

A number of observations may be made from the results of the exercises described above. The following is a summary of the major points of importance:

- 1. The latest Fish Reverse Ring Test (<u>FRRT12</u>) and Fish Ring (<u>FRT 14</u>) were successfully implemented and their format can be continued in the next scheme year. **Participants are encouraged to provide feedback to enable protocols and implementation to be improved where possible.**
- Most participating laboratories submitted data / specimens in accordance with the Scheme's timetable. There were only two slightly late submissions, although they did not delay initial analysis and distribution of interim reports.
 Participants should endeavour to supply data / specimens according to the exercise deadlines to ensure timely summary reporting.
- 3. Some identification differences might be the results of inadequate literature. Participants are encouraged to collate fish identification literature for problematic groups or juvenile specimens and follow the most recent taxonomy. Participants are encouraged to review the bibliography of taxonomic literature available on the NMBAQC website (Section 3 in Worsfold *et al.* 2020) and give details of additions where possible. Reference to online databases for the validity of scientific names (FishBase, WoRMS and Eschmeyer's Catalog of Fishes) is also recommended.
- 4. The maintenance of a comprehensive reference collection has numerous benefits for improving identification ability, maintaining consistency of identification between surveys and access to growth series material. The FRRT exercise can be used as a means of verifying reference specimens. Laboratories are strongly recommended to **implement and expand in-house reference collections of fish; these should include images alongside physical specimens**. The inclusion of juvenile material is useful for certain groups, *e.g.* clupeids. Ideally **all surveys should include a photographic reference of all species encountered as a minimum.**
- 5. Laboratories participating in the ring test exercises should attempt to **identify all specimens to species and complete the 'confidence level' section of their ring test datasheets** to enable additional information to be gathered regarding the difficulty of ring test specimens.
- 6. Since the beginning of the scheme, continual improvement to the learning structure of the Scheme reports has been crucial. For the FRRT and FRT detailed results have been forwarded as **individual exercise interim reports** to each participating laboratory as soon after the exercise deadlines as practicable. The results and subsequent differences raised in both exercises should **benefit all scheme participants**. A bulletin was circulated after each exercise, reviewing the literature used, detailing the accepted identification of the taxa received or circulated, and including images of relevant specimens and discussing problematic species. Participants are encouraged to review all exercise reports and **provide feedback concerning content and format** wherever appropriate.

- 7. Despite being raised as a problematic group in the past gobies and grey mullet continued to be groups with a high number of differences recorded. Future Fish Ring Test exercises are expected to target taxa that were highlighted as potentially problematic in FRT14 and FRRT12. Participants are encouraged to provide feedback on problem taxa that could be included in future exercises and are invited to submit specimens for use in future exercises (approximately 20 specimens of similar size and condition).
- 8. The distribution and analysis of an 'Image only' FRT in scheme year 2019/2020 provided lots of feedback and helped raise potential difficulties that would need to be overcome for the use of images to replace specimens in an exercise. However, the use of 'image only' specimens remain a potentially useful option for the inclusion of species of conservation interest, larger-bodied species, or scarce species that would otherwise be impractical to circulate. **Participants are encouraged to provide feedback on the use of 'image only' specimens in future exercises.**
- 9. After the distribution of preserved specimens in the previous Fish Ring Test (FRT13) some participants requested fresh specimens. For FRT14 all specimens were distributed frozen. Once thawed some of the smaller specimens were very fragile and easily damaged, potentially by larger still frozen specimens. Any relatively small or fragile specimens distributed in future exercise will be packaged separately to avoid damage in transit.
- 10. One of the laboratories submitted multiple data sets for the Fish Ring Test. Participants are encouraged to submit multiple data sets for sub-teams and individual analyst where possible to improve the training aspect of the exercise.
- 11. Protocol documents for each exercise of the Fish Component have now been produced. Protocols for the Reverse Ring Test can be found <u>here</u> and for the Ring Test <u>here</u>. **Participants are encouraged to review the protocols and provide feedback and suggestion to improve exercises.**
- 12. APEM Ltd. always strives to ensure smooth running and **transparency of the Scheme**. APEM Ltd. log and make available all correspondence to the Fish Component Contract Manager (Jim Ellis, CEFAS). Participants can be assured that their anonymity will be protected if this correspondence is required to be shared with the Committee.
- 4.4 Reports

Fish Component Annual Report 2020/2021 (Year 27)

Duncombe-Smith, S., and Hall, D., 2021. Fish component - Report from the contractor. Scheme Operation - 2020/2021. A report to the NMBAQC Scheme coordinating committee. 16pp, April 2021.

FRT 14 – March 2021

Duncombe-Smith, S., Hall, D., and Pears, D. 2021. NE Atlantic Marine Biological Analytical Quality Control Scheme. Fish Ring Test Bulletin: FRT#14. Report to the NMBAQC Scheme participants. APEM Report NMBAQC FRT#14, 24pp, March 2021.

FRRT12 – January 2021

Duncombe-Smith, S., and Hall, D., 2021. National Marine Biological Analytical Quality Control Scheme. Fish Reverse Ring Test: FRRT12. Final report to the NMBAQC Scheme participants. APEM Report NMBAQC FRRT12, 25pp, January 2021.

5 Phytoplankton component

Scheme Coordinator: Rafael Salas, Observatorio Canario de Algas Nocivas (OCHABs)

5.1 Summary of activities

IPI Phytoplankton Report

The phytoplankton component is now administered from the Canary Islands Harmful Algal Bloom Observatory (OCHAB), University of Las Palmas de Gran Canaria, Spain in collaboration with the IOC Science and Communication Centre on Harmful Algae, Denmark (and in association with the NMBAQC, UK). The change in location from the Marine Institute, Galway, Ireland to the Canary Islands, Spain was due to the host organiser and Phytoplankton Technical Manage for the NMBAQC scheme, Rafael Salas relocating to undertake his PhD. Previously, this component undertook intercomparison exercises under the BEQUALM banner. However, as the BEQUALM programme closed in 2014, these exercises were renamed in 2016 as IPI (International Phytoplankton Intercomparison).

Due to the global pandemic of Covid-19, the IPI 2020 exercise was cancelled. All participants were advised of this cancellation, once all partners had agreed this was the best course of action. For 2021, the IPI will be hosted at the University of Las Palmas de Gran Canarias in the OCHABs centre, the Canary Islands Harmful Algal Bloom Observatory, for the next four years from 2021 to 2025. A partnership agreement was signed by the University of Las Palmas and the IOC, and dates for the new 2021 IPI exercises were announced early 2021, with registration from March 2021 (deadline for submission of samples and online test results November 2021).

The IPI exercise will continue as it was before with the full collaboration of the IOC Science and Communication Centre on Harmful Algae, OTGA (OceanTeacher Global Academy) within the IODE office in Ostend, Belgium and in association with the NMBAQC in the UK. The only change is in relation to the host institution and administration centre for the IPI, which now will be based in Las Palmas, Gran Canaria, Spain.

Details of the next 2021 IPI exercise will be provided under the 2021/2022 NMBAQC Annual Report.

6 Macroalgae component

Contract Manager: Claire Young, DAERA-NI. Component Administrator: Emma Wells, Wells Marine.

This is the fifteenth year of the Macroalgae Component.

6.1 Summary of activities

The format for 2020 - 21 followed that of the previous year.

The component consisted of three modules:

1. **Opportunistic Macroalgae Biomass Ring Test (OMB - RT):** - synthetic samples of different weights for washing and drying to both wet and dry weights.

2. **Opportunistic Macroalgae/Seagrass Cover Ring Test (OMC - RT)**:- estimation of percentage cover of opportunistic macroalgae and seagrass based on photographs of field quadrats.

3. Rocky Shore Macroalgae Ring Test (RM - RT): - Identification of twenty macroalgae species based on a series of images.

The analytical procedures of all modules were the same as for the previous year of the Scheme. There were 4 laboratories participating in the OMB-RT, 7 laboratories in the OMC-RT and 4 laboratories in the RM-RT.

6.2 Summary of results

Biomass of macroalgae (<u>OMB-RT12</u>)

This is the twelfth year in which biomass of macroalgae has been included as a module of the NMBAQC scheme and was included as a single exercise. The format followed that of previous years of the module (OMB RT01 – RT11 - see <u>NMBAQC website</u>). Test material was distributed to participating laboratories from which data forms were completed with algal biomass results and returned for analysis.

Four laboratories were issued with test material. All four laboratories completed the macroalgae biomass module of the NMBAQC scheme. All of the participating laboratories were government; no other organisations took part in this component of the macroalgae exercises.

Results for wet weight of biomass varied between laboratories with some laboratories producing high measures of biomass compared against the average biomass and actual/expected biomass, particularly for the larger sample. The dry weights also showed a high degree of variability between laboratories. All laboratories remained within the Z-score limit of +/- 2.0 for both the dry weight and wet weight against the mean, which may have been due to the high standard deviation caused by the high range of results.

All four laboratories showed significant deviation from the actual dry weight of sample A with a further two 'Fails' against both wet and dry weight from one laboratory. It is worth noting that this means of assessment (against actual weight) is not as accommodating towards outliers hence the higher number of 'Fails'. There was a total of six 'Fails' across all assessments of which five could be attributed to dry weight comparisons. Three laboratories had dry weights lower than that of the actual dry weight for sample B, suggesting minor losses of material during the rinsing process.

Cover of macroalgae & seagrass (OMC-RT12)

This is the twelfth year in which % cover estimations of macroalgae have been included as a module of the NMBAQC scheme and the tenth year for which seagrass has been assessed as a separate exercise. This module included one exercise for macroalgae and one for seagrass both of which were split into three additional tests based on methodology. The format followed that of previous years (RT03 – RT11).

Seven laboratories were issued test material. All seven laboratories completed the % cover macroalgae/seagrass module with a total of 24 participants. Of those laboratories submitting results, all ten were government organisations.

Results for % cover of both opportunist macroalgae and seagrass varied between participants and between the different methods used. Several results deviated from the sample mean and from the % cover as calculated by image analysis. Deviation from the latter was more noticeable and this has also been reported in previous years. There was a considerable lack of consistency between the three methods in terms of the degree of continuity between participants as well as how the data compared with the image analysis % cover. There was greater preference for methods A and C for both macroalgae and seagrass and, as seen in previous years, method B had far fewer participants. The number of 'Fails' between test methods and comparison against mean or image analysis varied considerably with no apparent trend. The overall number of 'Fails' was similar for macroalgae and seagrass particularly when compared against ImageJ. The tests continue to produce a broad range of results thereby increasing the standard deviation, this results in the Z-scores being unable to pick up slight deviations from mean or ImageJ analysis % cover.

Rocky shore Macroalgae (<u>RM-RT15</u>)

This is the fifteenth year in which the identification of intertidal macroalgae has been included as a module of the NMBAQC scheme, with the format following that of previous years. Test material was distributed to participating laboratories using file transfer, from which species identification forms were completed and returned for analysis.

Four laboratories subscribed to the Rocky Shore macroalgae ring test module with all four laboratories submitting results with a total of four participants. Three of the subscribing laboratories were government organisations and one was an independent consultancy.

Round fifteen of the ring test produced a good degree of agreement between identifications made by participating laboratories and initial identification as made by Wells Marine. The ring test tried to incorporate a variety of common and more challenging species including some microscopic and epiphytic species.

The level of performance between laboratories and participants varied, with scores ranging from 29, with 4 incorrect genus names and 7 incorrect species names, to 38, with one incorrect genus and species name. No one participants correctly identified all species correctly. All participants correctly identified ten species. Most incorrect species

identification were made at the species level with only one species showing considerably difficulty at both genus and species levels. Overall, the level of identification was relatively consistent with the previous year with a high level of knowledge of the common species and increased knowledge of the more challenging and unusual species. Although the results were broadly comparable with those of previous years (RT01 through RT14) there was a noticeable decrease in the number of participants, making it difficult to make direct comparisons.

6.3 Reports

OMB RT12 Final Report 2021

Wells, E., 2021. National Marine Biological Analytical Quality Control Scheme-Macroalgae Biomass Component Report - OMB RT12 2021. Report to the NMBAQC Scheme participants. Wells Marine Surveys.

OMC RT12 Final Report 2021

Wells, E., 2021. National Marine Biological Analytical Quality Control Scheme-Macroalgae/Angiosperms % Cover Component Report - OMC RT12 2021. Report to the NMBAQC Scheme participants. Wells Marine Surveys.

RM RT15 Final Report 2021

Wells, E., 2021. National Marine Biological Analytical Quality Control Scheme-Macroalgae Component – Algal Identification Module Report - RM RT15 2021. Report to the NMBAQC Scheme participants. Wells Marine Surveys.

7 Epibiota component

Component Administrator: Joey O'Connor, JNCC.

7.1 Summary of activities

External quality assurance processes

JNCC, Cefas and Marine Scotland Science continued to include external quality assurance processes for further quality assuring results of imagery analyses undertaken in-house and sub-contracted for offshore Marine Protected Area monitoring. These include a full reanalysis of a subset of 10% of the imagery data by an independent analyst, a subsequent comparison of the two analyses to check for differences and remediation where necessary before the imagery analysis is deemed complete. The processes run alongside internal quality assurance checks undertaken by the primary analysts. The protocols are set out in each project specification with a summary of the protocol followed and results/remedial action undertaken captured in each project report. The protocols have been refined on a project-by-project basis with JNCC, Cefas, Marine Scotland Science, Envision Ltd, Seastar Survey Ltd, MarineSpace Ltd, Ocean Ecology Ltd and Galathea Ltd participating in 2020/2021. Costs relating to these new external quality assurance processes have been absorbed into the imagery analysis costs of each project on a per project basis.

Big Picture Group: Terms of Reference, Big Picture II workshop, Quality Assurance Framework and Epifaunal Identification Protocol

The work of the Big Picture Group was coordinated by JNCC in 2020/2021. JNCC drafted Terms of Reference for the Big Picture Group in August 2020, which have been agreed by all members, and held an online <u>Benthic Imagery Action Plan workshop</u> in November 2020 to identify Project Working Groups to take Actions identified in the <u>Benthic Imagery Action Plan</u> forward.

JNCC hosted a second benthic imagery analysis workshop ("THE BIG PICTURE II") via online video conferencing in March 2021. The three-day virtual event was a success with 85 attendees from the UK, Spain, Belgium, Netherlands, USA and Canada discussing the successes, funding routes, global outreach opportunities and future challenges of and for the Big Picture Group in implementing the UK Benthic Imagery Action Plan (https://jncc.gov.uk/news/big-picture-ii/).

JNCC have worked with the Marine Biological Association and Big Picture Project Steering Groups to develop an end-to-end benthic imagery Quality Assurance Framework (QAF), which includes an online tool for automating quality checks on MEDIN-compliant imagery purpose-specific data recording proformas. Taxonomic identification formed one of the themes at the first Big Picture Workshop and the development of an epibiota taxonomic discrimination protocol to inform taxonomic identification from imagery has continued through the QAF work. As part of this work JNCC held a series of mini workshops with imagery taxonomy experts between December and February 2021 to further develop the Epibiota Identification Protocol (EIP), which now contains 800 taxa. The outputs of this work were added to the NMBAQC website April 2021 (more information to follow in the next NMBAQC Annual Report) and the QAF process will be trialled by NMBAQC members throughout 2021/2022.

Priorities for 2021/2022 are to further develop the EIP and to host a Big Picture Group workshop on standards for sharing and archiving marine image annotations.

8 Zooplankton component

Component Administrators: David Johns & Marianne Wootton, the Marine Biological Association.

8.1 Summary of activities

Following a successful trial in 2014/2015, the first official zooplankton ring-test took place in 2016/17. The test is sent out biennially and details below describe the third official zooplankton ring-test representing the 2020/2021 term.

Sent out to participants in Dec 2020, the 2020/21 ring-test included a specimen identification (ID) element, a written quiz and an enumeration exercise, based upon developmental stages of copepods. An online results and discussion workshop was held

in September 2021 to evaluate results, to help taxonomists with any ID issues they may have and to exchange best practice within and between institutions.

Following the results discussion, participants were offered the opportunity to stay online to listen to expert guest lectures. Lectures included: "Introduction to copepod nauplii identification" by Dave Conway, Marine Biological Association; "Paracalanidae: characteristics and phylogenetic relationships. Biological and ecological information; morphological and molecular taxonomy" by Astrid Cornils, Alfred Wegener Institute and an "introduction to decapod larvae identification" by Antonina dos Santos, Instituto Português do Mar e da Atmosfera.

8.2 How Many Participants and From Where?

The 2020/21 ring-test saw 21 participants, from 14 different laboratories, from seven different countries take part in the exercise. As in previous years, most participants are from European laboratories; in addition, we welcome back a laboratory from the United Arab Emirates. There was an even mix between consultancies and governmental organisations taking part in the test.

8.3 Results

The results of the three-stage assessment are as follows:

The average result for the specimen ID was 74.73% with results ranging between 45% and 100%; the average score for the written exercise was 81.6%, with marks ranging from 58.3% to 100%. The enumeration section was coupled with basic copepod identification (separation of *Calanus* from other similar co-occurring genera) and an assessment of ability to separate different copepods life stages. For the non-expert reader, *Calanus* is a very common, abundant, and important copepod in the NE Atlantic. Levels of accuracy varied between 0% and 100% for the various life stages, with numbers of total *Calanus* showing a more reassuring % relative error ranging between a perfect 0% to 23.9%. Clearly, some participants struggle to separate different life stages and sexes of *Calanus*, which is of somewhat concern.

A full report detailing each section of the test is in the process of being compiled and will be completed during 2022.

Appendix 1 - NMBAQC Co-ordinating Committee – 2020/2021

Name	Organisation	Position /Role
David Johns	The Marine Biological Association (MBA)	Chair and Zooplankton Technical Manager
Tim Mackie	Department of Agriculture, Environment and Rural Affairs, Northern Ireland (DAERA)	
Graham Phillips	Environment Agency (EA)	Finance Manager and CMA representative
Myles O'Reilly	Scottish Environment Protection Agency (SEPA)	Invertebrate Technical Manager and CMA representative
Rafael Salas	Marine Institute, Ireland (MI)	Phytoplankton Technical Manager
Claire Young	Department of Agriculture, Environment and Rural Affairs, Northern Ireland (DAERA)	Macroalgae Technical Manager
Ross Griffin	Ocean Ecology Ltd	Contractors' Representative
Joey O'Connor	Joint Nature Conservation Committee (JNCC)	Epibiota Component Administrators
Jim Ellis	Centre for Environment, Fisheries & Aquaculture Science (Cefas)	Fish Technical Manager
Claire Mason	Cefas	PSA Technical Manager
Paul McIlwaine	Cefas	CMA Representative
Matthew Green	Natural Resources Wales (NRW)	CMA Representative
Adele Boyd/Alex Callaway	Agri-Food Biosciences Institute, Northern Ireland (AFBI)	CMA Representative
Claire Taylor	The Marine Biological Association (MBA)	Technical Secretary

Appendix 2 - NMBAQC Scheme – Component Participation for 2020/2021

(Participants from UK unless otherwise stated)

Invertebrates 2020-2021 Participants:

	Ring Test (RT) Module	Laboratory Reference (LR) Module	Own Sample (OS) Module (audit)
	(intercalibration / training)	(intercalibration / training)	
Agri Food Biosciences Institute (AFBI) NI	-	-	\checkmark
APEM	Administrator	Administrator	Administrator
Benthic Solutions Limited	-	-	\checkmark
Biofar	\checkmark	\checkmark	-
Biotikos Limited	-	-	✓
Cefas Lowestoft Benthic Laboratory	\checkmark	-	-
Cyfoeth Naturiol Cymru / Natural Resources Wales	-	-	√(x6)
DAERA Environment, Fisheries and Marine Group Laboratory	\checkmark	✓	\checkmark
Eco marine Consultants Ltd	-	-	\checkmark
Ecospan Environmental Ltd	\checkmark	\checkmark	✓
Environment Agency, Kingfisher House	-	-	√(x2)
Eurofins Hydrobiologie France	-	-	\checkmark
Eurofins Omegam BV	\checkmark	\checkmark	-
Fishlab, Viby, c/o Orbicon, Denmark	\checkmark	-	-
Fugro GB Marine Limited (Edinburgh)	\checkmark	-	-
Fugro GB Marine Limited (Gt. Yarmouth)	\checkmark	-	-
Fugro GB Marine Limited (Portsmouth)	\checkmark	-	\checkmark
HEBOG Environmental Limited	\checkmark	-	\checkmark
Hull Marine Laboratory (formerly IECS)	\checkmark	-	\checkmark
ILVO (Institute for Agricultural and Fisheries Research) -	\checkmark	-	\checkmark
Magnus Axelsson	\checkmark	-	-
Marine Invertebrate Ecological Services	-	-	\checkmark
Marinescope Taxonomy Ltd	\checkmark	-	-
MBM Benthic Identification Services (The Lab Shed)	\checkmark	-	-
Myriad Taxonomy	-	-	\checkmark
Natural England	-	-	\checkmark
Ocean Ecology	\checkmark	-	\checkmark
Pelagia Nature & Environment AB, Sweden	\checkmark	-	-
Pharmaq Analytic Limited (formerly Fish Vet Group)	-	-	\checkmark
	1		

Precision Marine Survey Ltd	\checkmark	-	-
Rijkswaterstaat CIV	\checkmark	\checkmark	-
Seastar Survey Ltd	-	-	\checkmark
Scottish Environment Protection Agency	\checkmark	-	\checkmark
Shalla Benthic Indentification Services	\checkmark	-	-
Thomson Ecology Ltd	-	-	\checkmark
WMR (Wageningen Marine Research)	\checkmark	\checkmark	-

PSA 2020-2021 Participants:

	Particle Size (PS) Module (intercalibration / training)	Particle Size Own Sample (PS-OS) Module (audit)
ABPmer	-	\checkmark
Agri Food Biosciences Institute (AFBI) NI	\checkmark	\checkmark
APEM	Administrator	Administrator
Benthic Solutions Limited	\checkmark	-
Biotikos Limited	-	\checkmark
Cefas Lowestoft Benthic Laboratory	\checkmark	\checkmark
Cyfoeth Naturiol Cymru / Natural Resources Wales	\checkmark	√(x4)
DAERA Environment, Fisheries and Marine Group Laboratory	\checkmark	\checkmark
Ecospan Environmental Ltd.	\checkmark	-
Fugro GB Marine Limited (Portsmouth)	\checkmark	-
Hull Marine Laboratory(formerly IECS)	\checkmark	\checkmark
Kenneth Pye Associates Ltd	\checkmark	\checkmark
Marine Scotland Laboratory	\checkmark	-
National Laboratory Services (EA)	\checkmark	\checkmark
Ocean Ecology	\checkmark	-
Precision Marine Survey Ltd	\checkmark	-
Rijkswaterstaat CIV	\checkmark	-
RPS	\checkmark	-
Scottish Environment Protection Agency	\checkmark	\checkmark
Thomson Ecology Ltd	\checkmark	-

Fish 2020-2021 Participants:

	Fish - Reverse Ring Test (FRRT10)	Fish - Test Ring (FRT12)
Agri Food Biosciences Institute	\checkmark	\checkmark
APEM Limited	Administrator	Administrator
Cyfoeth Naturiol Cymru/Natural Resources Wales	√ (x2)	-
Department of Agriculture, Environment and Rural Affairs (DAERA)	\checkmark	\checkmark
Environment Agency (ECMAS)	✓	√(x6)
Fugro GB Marine Limited	\checkmark	-
Hull Marine Laboratory (formerly IECS)	\checkmark	-
Ocean Ecology ltd.	\checkmark	-
The Marine Biological Association of the United Kingdom	-	\checkmark