

FAO Reference Centre - Annual report for the FAO Reference Centre for Bivalve Mollusc Sanitation Calendar 2019-20

(Thematic areas¹)

Title of FAO Reference Centre	FAO Reference Centre for Bivalve Mollusc
	Sanitation
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above)	
Reporting period	January 2019 – December 2019
Data of reporting	May 2020
Date of reporting	iviay 2020

1. Introduction

The Centre for Environment, Fisheries and Aquaculture Science (Cefas), was designated by the Food and Agriculture Organization of the United Nations (FAO) as an FAO Reference Centre for Bivalve Mollusc Sanitation in 2018. The Reference Centre's mandate is to contribute to the implementation of FAO's scientific, technical and economic programme priorities and to provide specific, independent technical/scientific advice aimed at strengthening capacities in countries and regions. According to the 'Terms and Conditions of the Designation' agreed between FAO and the United Kingdom's Department of Environment, Food and Rural Affairs (Defra), the duration of the FAO Reference Centre for Bivalve Mollusc Sanitation is 4-years. This annual report describes the technical and scientific activities delivered via the Reference Centre to support the FAO Department of Fisheries and Aquaculture, and the associated costs for calendar year 2019. Financial support for operation of the Reference Centre was afforded by Defra and the United Kingdom's Food Standards Agency (FSA), with additional contribution specifically for the Bivalve Mollusc Sanitation workshop, Nairobi, Kenya, November 2019 from FAO Department of Fisheries and Aquaculture. Details of finances are provided in Annex I.

2. Areas of Collaboration

The annual work programme of the FAO Reference Centre for Bivalve Mollusc Sanitation was agreed in December 2018 [https://www.cefas.co.uk/faobivalves/]. Five main areas of collaboration were agreed for 2019.

2.1 Provision of support for the development and maintenance of FAO eLearning material and resources on bivalve mollusc sanitation.

In 2018, as part of the FAO/WHO Food Safety and Quality Series, Technical Guidance for the Development of the Growing Area Aspects of Bivalve Mollusc Sanitation Programmes was published². This technical guidance was produced by a group of expert contributors from Canada, Chile, Italy, Japan, Mexico, Morocco, Namibia, New Zealand, Peru, Spain, United Kingdom, United States and Viet Nam, including significant input from Cefas. One of the primary outputs for the FAO Reference Centre, in collaboration with FAO Department of Fisheries and Aquaculture and FAO eLearning group for the period of the designation, is the translation of this technical guidance to an eLearning format. In 2019 the framework, format and content of three eLearning modules was agreed completed, published (Figure Module one was reviewed and [https://www.cefas.co.uk/international-centres-of-excellence/seafood-safety/our-services/trainingand-workshops/] in 2019. Plans for the development of modules 2 and 3 are underway. FAO eLearning user statistics indicated over 600,000 active learners regularly access eLearning globally. This demonstrates the power and reach of this approach, which also reduces the necessity for incountry visits reducing Carbon emissions and helps target capacity building needs in Low- and Middle-Income Countries (LMICs).

² FAO and WHO (2018) *Technical guidance for the development of the growing area aspects of Bivalve Mollusc Sanitation Programmes.* Food Safety and Quality Series No. 5 Rome, 292 pp Licence: CC BY-NC-SA 3.0 IGO.



Figure 1. Framework for the development of eLearning based upon Technical Guidance for the Development of the Growing Area Aspects of Bivalve Mollusc Sanitation Programmes



Figure 2. eLearning Module 1 – Bivalve Mollusc Sanitation Growing Area Risk Profile published in 2019 [https://www.cefas.co.uk/international-centres-of-excellence/seafood-safety/our-services/training-and-workshops]

2.2 Provide guidance on relevant laboratory protocols, accreditation and use of methods for bivalve mollusc testing

A website hosted by Cefas [https://www.cefas.co.uk/faobivalves] was established enabling interested parties to access relevant information, download laboratory method protocols, troubleshooting guidance, technical reports, eLearning and register for proficiency/comparative testing (Figure 3).

The website forms the primary method of dissemination for the FAO Reference Centre community and will be continuously improved based upon stakeholder feedback and user statistics.

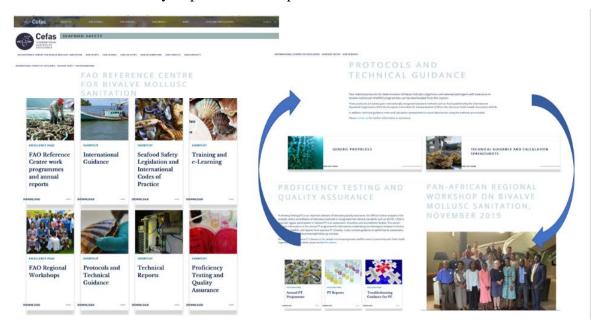


Figure 3. Newly established website www.cefas.co.uk/faobivalves

2.2.1 Laboratory protocols (accredited ISO/IEC 17025 and non-accredited) available from the Reference Centre

The methods listed in Table 1 were undertaken at the Reference Centre with applicability to bivalve molluscs and were made available to interested parties via the website www.cefas.co.uk/faobivalves. Reference materials can be provided on request to laboratories wishing to develop and implement methods for determination of bivalve mollusc or growing area quality.

Methods accredited to ISO/IEC 17025 with applicability for bivalve mollusc sanitation

Enumeration of *Escherichia coli* in bivalve molluscan shellfish by the most probable number (MPN) technique (based on ISO 16649-3)

Detection of potentially pathogenic Vibrio spp. in bivalve shellfish

Detection of Salmonella spp. in bivalve molluscs (based on ISO 6579-1)

Quantitative determination of norovirus and hepatitis A virus in bivalve molluscs (using ISO 15216-1)

Non-accredited methods and guidance with applicability for bivalve mollusc sanitation

Protocol for the enumeration of FRNA bacteriophage in bivalve shellfish

Guidance for derivation of MPN results for official control testing of bivalve molluscs

Non-accredited methods and guidance with applicability for bivalve mollusc sanitation (cont.)

Guidance for determining uncertainty of measurement for the enumeration of E. coli in bivalve molluscs

Guidance for the use of proprietary Salmonella detection kits for detection and identification of *Salmonella* spp. in bivalve molluscs

Guidance for providing supplementary data on the performance of the vvHA real time PCR assays for the detection of *V. vulnificus*

Determination of limits of detection and quantification for determination of viruses in bivalve shellfish

Example datasets for determination of limits of detection and quantification for determination of viruses in bivalve shellfish

Calculation spreadsheet for quantification of viruses in bivalve shellfish using ISO 15216-1

Guidance for best practice for norovirus testing in shellfish

Table 1. ISO/IEC 17025 accredited and non-accredited methods with applicability for bivalve mollusc sanitation

2.2.2 Method Standardisation Activities and Representation

Personnel at the Reference Centre led or contributed to activities supporting method development, validation and standardisation with relevance to bivalve molluscs, in 2019 these included;

ISO Ad Hoc Group "ISO 15216-1:2017/AMD-1" – Preparing the technical amendment to ISO 15216-1:2017 Microbiology of the food chain — Horizontal method for determination of hepatitis A virus and norovirus using real-time RT-PCR —Part 1: Method for quantification

ISO/TC34/SC9/WG3 "Method Validation" – Preparing the series IOS 16140 setting out requirement for method validation

ISO/TC34/SC9/W27 "Vibrios" – Developing methods (quantitative and qualitative) for determination of human pathogenic *Vibrio* spp. (*V. parahaemolyticus*, *V. vulnificus* and *V. cholerae*) in seafoods (including bivalve molluscs.

CEN/TC463/WG1 "General requirements relating to PCR methods"

CEN/TC275/WG6/TAG4 "Viruses in foods" – development and validation of ISO 15216-1:2017 Microbiology of the food chain — Horizontal method for determination of hepatitis A virus and norovirus using real-time RT-PCR —Part 1: Method for quantification and Part 2: Method for Qualitative Detection

ISO Ad Hoc Group "ISO 6887-3/AMD1" - Preparing the technical amendment to ISO 6887-3 Microbiology of food and feed — Preparation of test samples, initial suspension and decimal dilutions for microbiological examination —Part 3: Specific rules for the preparation of fish and fishery products.

2.3 Prepare and deliver an International/regional workshop on bivalve mollusc sanitation

The Reference Centre supported FAO's scientific, technical and economic programme priorities in Africa via the delivery of a Pan-African regional workshop on Bivalve Mollusc Sanitation in Nairobi, Kenya on the 11th - 12th November 2019. The aim of the workshop was to begin to build targeted programmes of capability building and networks in sub-Saharan African Responsible Authorities and Official Laboratories. Invitation of national Responsible Authorities and others was preceded by a selection process considering previous interest in bivalve production (received via FAO or Cefas), previous involvement with FAO in projects developing bivalve mollusc production (including the FAO technical guidance pilot programmes in Angola, Madagascar, Namibia and Mozambique 2015/16), FAO production statistics for bivalves between 2011 and 2015, and existing international trade in fish, fisheries products and bivalve molluscs. The overarching aim of the workshop was to provide support to countries wishing to sustainably develop or expand their production of bivalve molluscs (oysters, mussels, clams etc.) for domestic consumption or Interregional and International trade. National Competent Authority delegates from Angola, Cameroon, Djibouti, Gambia, Ghana, Kenya, Madagascar, Mauritius, Mozambique, Senegal, South Africa and Sudan attended the two-day workshop (Figure 4), together with representatives from FAO Kenya, the Reference Centre and Health Canada. The workshop comprised presentation of existing capability and capacity data gathered via a pre-workshop questionnaire (Figure 5), dissemination of aspects of the technical guidance for growing areas aspects of bivalve mollusc production areas, and several interactive and scenario planning sessions identifying growing area hazards and constructing risk profiles to assist in decision making with respect to either establishing new (aquaculture) or exploiting natural bivalve resources (Figure 6). The workshop agreed 6 Resolutions identifying the benefits and priorities of further technical capacity building in the future (Figure 7). A list of the workshop Resolutions is Annex II. A full report of the workshop can be [https://www.cefas.co.uk/media/wscdwpui/1st-faorc-africa-workshop-final-report.pdf].



Figure 4. Delegates of the Pan-African regional workshop on Bivalve Mollusc Sanitation in Nairobi, Kenya on the 11th - 12th November 2019 attended by National Competent Authority delegates from Angola, Cameroon, Djibouti, Gambia, Ghana, Kenya, Madagascar, Mauritius, Mozambique,

Senegal, South Africa and Sudan

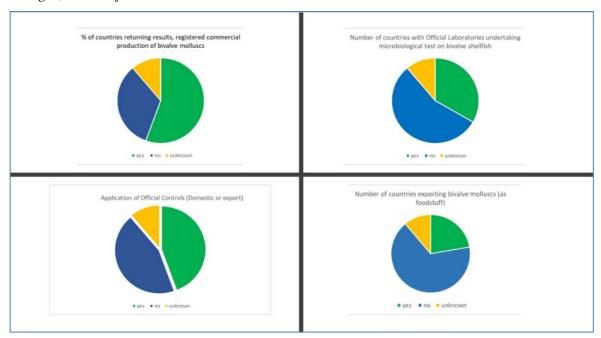


Figure 5. Summary extracted pre-workshop questionnaire responses – assessing existing capacity and capabilities amongst countries attending the Pan-African regional workshop on Bivalve Mollusc Sanitation



Figure 6. Scenario planning sessions at the FAO Pan-African regional workshop on Bivalve Mollusc Sanitation identifying growing area hazards and constructing risk profiles to assist in decision making with respect to either establishing new (aquaculture) or exploiting natural bivalve resources

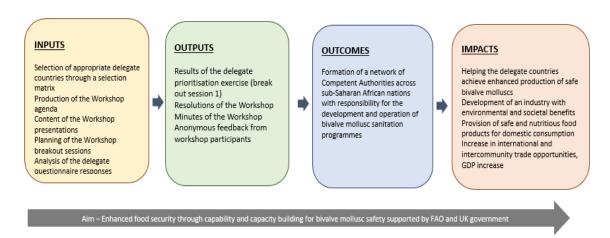


Figure 7. Benefits and impact assessment of the Pan-African regional workshop of the FAO Reference Centre for Bivalve Mollusc Sanitation

2.4 On request deliver training and support to include provision of protocols, reference materials and access to proficiency testing programmes on microbiological testing and assessment of bivalve growing areas.

Protocols, reference materials and proficiency testing distributions were made available via the website (see 2.2). A signposting demonstration of how to access materials was provided to delegates at the Pan-African workshop. In addition, the schedule for distributions of proficiency testing material in 2019 (available to all Member Countries, cost recovery basis) is provided as Annex III. Proficiency testing is provided for *E. coli*, *Salmonella* spp. and viruses with relevance to bivalve shellfish in both matrix and non-matrix samples, the latter in collaboration with the UK's Public Health England.

The Reference Centre provided training support, inclusive of the Pan-African regional workshop on Bivalve Mollusc Sanitation and outreach activities to 17 countries during 2019 (Figure 9). In addition to the main workshop output a session on the Technical Guidance for the Development of the Growing Area Aspects of Bivalve Mollusc Sanitation Programmes at a Newton Researcher Links programme in Florianopolis, Brazil focused on tools to assess water quality, public health and risk The aim, in part, of the programme was to introduce and foster collaborations between early career researchers from academia, delegates from the Brazilian Competent Authority and other government institutes with responsibility for bivalve shellfish production and public health.



Figure 8. FAO Reference Centre for Bivalve Mollusc Sanitation, countries to which training support and outreach activities were delivered during 2019



Figure 9. FAO Reference Centre for Bivalve Mollusc Sanitation supporting outreach and training in Florianopolis, Brazil, February 2019.

The expert group developing the Technical Guidance co-authored a paper presented by a member of staff from the Reference Centre at the International Conference on Molluscan Shellfish Safety, Mexico in September 2019. 'A new FAO Reference Centre for bivalve shellfish sanitation – global food in a global world'. The conference abstract is included as Annex IV.

2.5 Additional activities

The Reference Centre contributed to several initiatives in the FAO's mandate with respect to supporting the development of safe bivalve production in 2019.

2.5.1. Global Repository of Vibrio Genomes

Reference Centre funding was used to partially support the continued development of the global repository of vibrio genomes. This work supports the development of an online tool for molecular characterisation and deposition of vibrio genomes. The aim is to link this tool with the pre-existing remote vibrio risk mapping tool [https://www.ecdc.europa.eu/en/publications-data/vibrio-suitability-tool] produced previously in a collaboration with the European Centre for Disease Control (ECDC), which identifies permissive conditions for vibrio proliferation based upon global surface seawater temperature and salinities. On completion the Global Repositories of Vibrio will work alongside the risk mapping tool creating a biorepository of the Earth's vibrio genomes, providing tools that describe not only where potentially pathogenic organisms are likely to be found, but when they are found how they get there, how related to each other they are and what risks they pose to health, and to supply chains.

To date several thousand strains of *Vibrio parahaemolyticus* have been added to the database. The application of bioinformatic tools and imaging software enables real-time visualisation of complex genomic data within an epidemiological and ecological context (Figure 10).

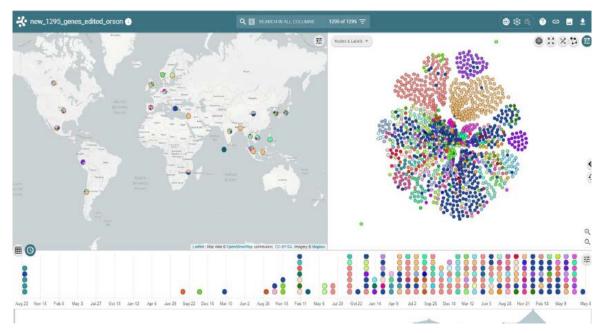


Figure 10. Global Phylogeny of Vibrios. Interactive visualisation of the phylogenetic relationships of strains collected across the world

2.5.2. WHO/FAO workshop

Over the last year, we have worked closely with the FAO, World Health Organisation (WHO) and the global community of vibrio experts on the development of global FAO/WHO microbiological risk assessments. In May 2019, the Reference Centre hosted the Joint FAO/WHO Expert Meeting on Microbiological Risk Assessment to update FAO/WHO guidance to reduce public health risks from pathogenic marine vibrios. Nineteen experts in the fields of genomics, epidemiology, risk assessment, pathogen detection, method standardisation and remote sensing from 13 countries, including the U.S. (CDC), Canada, India, Chile, New Zealand and Europe focused on how to improve existing risk assessments. The aim of the meeting was to establish the scientific basis for the development of future international seafood safety standards within Codex Alimentarius.

2.5.3 Peer review publications written by staff associated with the FAO Reference Centre for Bivalve Mollusc Sanitation

- 1. **Martinez-Urtaza Jaime** *et al.* The 2019 report of The *Lancet* Countdown on health and climate change: ensuring that the health of a child born today is not defined by a changing climate, The Lancet, <u>Volume 394, ISSUE 10211</u>, P1836-1878, November 16, 2019.
- 2. Yang, C., Pei, X., Wu, Y. **Martinez-Urtaza J.** *et al.* Recent mixing of *Vibrio parahaemolyticus* populations. *ISME J* **13,** 2578–2588 (2019). https://doi.org/10.1038/s41396-019-0461-5
- 3. Lozano-Leon A, Garcia-Omil C, Dalama J, Rodriguez-Souto R, **Martinez-Urtaza J**, Gonzalez-Escalona N. Detection of colistin resistance *mcr*-1 gene in *Salmonella enterica* serovar Rissen isolated from mussels, Spain, 2012- to 2016. *Euro Surveill*. 2019;24(16):1900200. doi:10.2807/1560-7917.ES.2019.24.16.1900200.
- 4. Núñez-Montero, K., Lamilla, C., Abanto, M. **Martinez-Urtaza J** *et al.* Antarctic *Streptomyces fildesensis* So13.3 strain as a promising source for antimicrobials discovery. *Sci Rep* **9**, 7488 (2019). https://doi.org/10.1038/s41598-019-43960-7

- 5. Yang, C., Pei, X., Wu, Y. **Martinez-Urtaza J** *et al.* Recent mixing of *Vibrio parahaemolyticus* populations. *ISME J* **13,** 2578–2588 (2019). https://doi.org/10.1038/s41396-019-0461-5
- 6. **Walker, D.I., Cross, L.J.**, **Stapleton, T.A.** *et al.* Assessment of the Applicability of Capsid-Integrity Assays for Detecting Infectious Norovirus Inactivated by Heat or UV Irradiation. *Food Environ Virol* **11**, 229–237 (2019). https://doi.org/10.1007/s12560-019-09390-4
- 7. **Lowther, J.A., Cross, L., Stapleton, T.** *et al.* Use of F-Specific RNA Bacteriophage to Estimate Infectious Norovirus Levels in Oysters. *Food Environ Virol* **11,** 247–258 (2019). https://doi.org/10.1007/s12560-019-09383-3
- 8. Hardstaff, J., Clough, H., Harris, J., **Lowther, J.**, Lees, D., & O'Brien, S. (2019). The use of capture-recapture methods to provide better estimates of the burden of norovirus outbreaks from seafood in England, 2004–2011. *Epidemiology and Infection*, 147, E65. doi:10.1017/S0950268818003217
- 9. Robson V. de Souza, **Andrew D. Younger, Mickael Teixeira Alves**, Carlos J.A. Campos, The influence of the number of *Escherichia coli* results on the classification status and assessment of microbiological risk of shellfish production areas, Food Control, Volume 103, 2019, Pages 86-90.

Note:

For country names, please refer to the Names of Countries system (NOCS): http://termportal.fao.org/faonocs/appl/

Annex I. Financial support for the FAO Reference Centre for Bivalve Mollusc Sanitation received at Cefas (Financial year 2019/20)

Funding Body	Amounts
Department for Environment, Food and Rural Affairs (Defra)	£95,000
Food Standards Agency (FSA)	£125,000
Other (FAO)*	£8,202
Total	£228,202
Staff Pay	£179,765
Non-Pay (T&S / Consumables etc)	£48,437

^{*}Contribution to delegate expenses for regional workshop on bivalve mollusc sanitation Nairobi, Kenya, November 2019

Annex II.





World Class Science for the Marine and Freshwater Environment

Resolutions of the 1st Workshop of the FAO Reference Centre for Bivalve Mollusc Sanitation, Ole Sereni Hotel, Nairobi, Kenya, 11-12th November 2019

- 1. The FAO/Cefas Reference Centre team thanked the representatives from the Member Countries of Angola, Cameroon, Ghana, Gambia, Djibouti, Kenya, Madagascar, Mauritius, Mozambique, Senegal, South Africa, and Sudan for their participation in the workshop and for their high level of engagement throughout the two days.
- 2. The representatives of the Member Countries agreed that they **shared the goal** of the **development and enhancement of microbiologically safe bivalve molluscs production**, and that this goal would be best achieved through the building of **scientific collaborations** and supporting **networks**.
- 3. Further to the above, the FAO/Cefas Reference Centre team confirmed the intent to support the development of scientific collaborations and supporting networks, and that this workshop and the formation of this group was a first, important step in developing these collaborations and networks.
- 4. The representatives from Member Countries identified their key challenges with respect to the further development and enhancement of safe bivalve shellfish following **priorities for further support and development**:
 - a. **Training of Competent Authorities** in all aspects of bivalve mollusc sanitation programmes.
 - b. **Training of laboratories** in testing of microbiological (and other) determinants as used in bivalve mollusc sanitation programmes.
 - c. Access to **project funding** in order to further development of bivalve sanitation programmes to achieve the goal identified in Resolution 2.
- 5. The representatives from Member Countries agreed the **FAO Reference Centre for bivalve mollusc sanitation** could play an **important role in the provision of training and further development of programmes**, but it was acknowledged that to maximise this additional project funding would be required.

The representatives from Member Countries also identified a need for information, **technical assistance** and **training** in other aspects of bivalve mollusc safety such as **marine biotoxins**.

Annex III

Cefas bivalve shellfish proficiency testing programme 2019

Reference No.	Description	Distribution date
NHV005 a	Norovirus and Hepatitis A virus examination in LENTICULES™ (quantitative and/or qualitative) (Cefas / FEPTU PHE collaboration)	February 2019
SF062 a	E. coli and Salmonella spp. examination in LENTICULES™ (Cefas / FEPTU PHE collaboration)	February 2019
PT 79 ^b	Matrix samples for examination for norovirus and hepatitis A (quantitative and/or qualitative)	June 2019
SF063ª	E. coli and Salmonella spp. examination in LENTICULES™ (Cefas / FEPTU PHE collaboration)	June 2019
NHV006 a	Norovirus and Hepatitis A virus examination in LENTICULES™ (quantitative and/or qualitative) (Cefas / FEPTU PHE collaboration)	October 2019
SF064 a	E. coli and Salmonella spp. examination in LENTICULES™ (Cefas / FEPTU PHE collaboration)	October 2019
PT 80 b	Whole animal distribution of live bivalve molluscs for examination for <i>E. coli</i> and <i>Salmonella</i> spp.	November 2019

^a For registration in the Cefas / FEPTU PHE collaborative scheme, please contact the PHE via email at foodeqa@phe.gov.uk

^b For registration in the Cefas Centre of Excellence scheme, please contact Cefas via email at fsq@cefas.co.uk or louise.stockley@cefas.co.uk

Annex IV

A new FAO Reference Centre for bivalve shellfish sanitation – global food in a global world

Rachel Hartnell, Jose Alejandro Barreiro Isabel, Yhony Omar Flores, Enrico Buenaventura, Covadonga Salgado, Paloma Ellitson, Btissam Ennaffah, Gregory Goblick, Suwimon Keerativiriyaporn, Mario Latini, Dorothy-Jean McCoubrey, Tran Bich Nga, Brian Roughan, Claudia Rozas Masataka Satomi and Esther GarridoGamarro.

Globally, bivalve molluscs comprise an important proportion of seafood. Increases in production will contribute to feeding the world population as this expands however bivalves, especially those consumed live or raw, are recognised as high-risk foods capable of concentrating contaminants and thereby mediating infectious diseases and intoxications. Food safety programmes directed at bivalve molluscs are intended to address these risks but have tended to be confined to developed countries. Application of such programmes in developing countries is necessary both to protect local consumers and to facilitate international trade, thus raising people from food poverty and increasing economic benefits to the community or nation. However, of the approximately 16 million tonnes of bivalves produced annually across the world only around 3% is traded outside of the country of production. One of the reasons for this restricted trade is the requirements for exporting nations to satisfy variable, and often very different, food safety legislation of importing countries. In 2018 FAO and WHO, working through a group of international experts, produced technical guidance to assist countries in developing and applying growing area sanitation programmes to help countries step through the complexity of developing bivalve shellfish programmes www.fao.org/3/CA1213EN/ca1213en.pdf... The guidance, which is based on Chapter 7 of the Codex Code of Practice for Fish and Fishery Products has been welcomed by the Codex Committee on Food Hygiene and recently a new FAO Reference Centre for bivalve shellfish sanitation has been established to further promote the development of bivalve shellfish programmes globally. One of the ambitions of the Reference Centre, which is funded by UK government, is to assist countries wishing to develop, enhance or expand their production of safe bivalve molluscs through providing scientific technical advice and training through, in particular, supporting the application of the FAO/WHO Technical Guidance.

In this paper we describe the development, risk-based philosophy and application of the technical guidance, and further set out the work programme for the FAO Reference Centre to achieve its aim in assisting nations to achieve their bivalve shellfish food security goals.