



Capacity building improve Malaysia's inspection and monitoring system for aquaculture and fishery products

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Training Report



Wageningen UR Centre for Development Innovation (CDI) works on processes of innovation and change in the areas of secure and healthy food, adaptive agriculture, sustainable markets and ecosystem governance. It is an interdisciplinary and internationally focused unit of Wageningen University & Research centre within the Social Sciences Group.

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Project: Capacity building improve Malaysia's inspection and monitoring system for aquaculture and fishery products (BO-10-009-117)

This research project has been carried out within the Policy Supporting Research task for the Ministry of Economic Affairs, Agriculture and Innovation, Theme: Robust Systems, Cluster: International Cooperation.

The course was implemented in partnership by Wageningen UR Institute of Food Safety (RIKILT).



Capacity building improve Malaysia's inspection and monitoring system for aquaculture and fishery products

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The project aimed to help build a credible inspection and monitoring system that can guarantee safe quality products of Ministry of Health (MoH) and Department of Fisheries (DoF) by upgrading the analytical capacity of the laboratory staff directly involved in the analysis and detection of forbidden substances. Two training courses were implemented in 2011 in the Bio Security Centre in Kuantan, Malaysia. The first training course on 'Marine lipophilic toxins using LC-MS/MS has been implemented in June and the second training course on on Stilbenes and Nitroimidazoles sample preparation and analysis with LC-MS/MS equipment was implemented in November, 2011. Through this knowledge transfer and laboratory enhancement the project contributed the laboratory's process towards getting accreditation under ISO 17025. The courses were implemented in partnership by Wageningen UR Institute of Food Safety (RIKILT) and Wageningen UR Centre for Development Innovation (CDI).

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Training Course on Marine Lipophilic Toxins Using LC-MS/MS

20-24 June, 2011

Kuantan, Malaysia

Arjen Gerssen, Mirjam Klijnstra, Ingrid Gevers

Summary

The Department of Fisheries of Malaysia is recognised as a Competent Authority by the EU since 2010. It has issued tenders to purchase more advanced test equipment such as GC TOS MS, ICP MS and additional LC-MS/MS to enhance the number of samples and parameters to be analysed in the laboratories. Consequently there is a need to improve the capability and competence of the technicians and laboratory staff manning and operating these equipment's. There is not only a need to upgrade the analytical capacity of the laboratory staff directly involved in the analysis and detection of forbidden substances, but also to improve their skills and knowledge to analyse additional parameters. Through knowledge transfer and laboratory enhancement this BOCI project will support the laboratory's process towards getting accreditation under ISO 17025.

The short term objective of the BOCI project is to increase the knowledge and skills of Ministry of Health (MoH) and Department of Fisheries (DoF) as EU-accredited competent authority (CA) . The long term objective is to have a national body that provides good governance, effective control over the entire production chain from farm to table, with the ultimate goal to help build a credible inspection and monitoring system that can guarantee safe quality products.

The project follows a training of training approach that allows the Department of Fisheries in Malaysia to increase the capacity and competence of all relevant staff on methods of analyses over a wider spectrum of parameters. Subsequently this will result in improved Laboratory Services as an important component of the Food Safety and Quality Assurance System. Eventually the greater capacity and capability of the laboratories and their staff will result in accreditation for official analyses.

The first training course on 'Marine lipophilic toxins using LC-MS/MS has been implemented from 20-24 June 2011. The course was implemented in partnership by Wageningen UR Institute of Food Safety (RIKILT) and Wageningen UR Centre for Development Innovation (CDI). RIKILT provided the expertise on laboratory tests and - practices, and CDI was responsible for the overall design of the training curriculum, overall project management and facilitation of a dialogue around ISO certification during implementation of the training course.

A total of 10 participants working at different laboratory of the Biosecurity Centre under the Department of Fisheries in Malaysia participated in this course (see Annex I). The course focused marine biotoxins, sample preparation and analysis with LC-MS/MS equipment. The training activities of the Malaysian Department of Fisheries will follow an institutional capacity development strategy that is clearly linked to the residue monitoring plan and the EU food safety and quality requirements.

The training was guided by a dialogue on laboratory accreditation and identified gaps to achieve this. Recommendations were given to the Kuantan laboratory to improve procedures, infrastructures and operation to work towards this ISO 17025 accreditation. The course was highly interactive using a combination of theoretical interactive PP presentations and practical laboratory work aiming to improve the analytical skill of the participants. Theory and practice were alternated throughout the course.

The daily course programme is given in Annex II. The course topics were all in line with the course objectives described in the proposal of the course although the timing allowed for each sessions changed based on needs and expectations of the participants.

To assess if the training activities were indeed relevant for the participants and contributed to building a credible inspection and monitoring system that can guarantee safe quality products monitoring, the training course included an evaluation focusing on content and methodology used. At the end of the training a meeting was held to discuss the progress made so far and issues remaining with regard to the Laboratory Support Services. This information provide the basis for the further design of the capacity development activities for 2011 and beyond with the final aim for the Malaysian laboratories to become accredited for official analyses. This discussion was guided by the residue monitoring plan and the EU food safety and quality requirements.

Following you can find a short description of the different daily sessions included in the training course.

Day 1, June 20

Opening of the course

Mr Hamdan Jaafar, Head of the Fisheries Biosecurity Centre opened the course and welcomed the participants. Ingrid Gevers of Wageningen UR Centre for Development briefly introduced the objectives and the content of the course.

Introduction to Wageningen UR RIKILT Institute of Food Safety and Centre for Development Innovation

Introduction to the course

The first day started with a getting to know session. The participants were asked to work in pairs and get to know their colleagues by answering the following questions:

1. What is your name, what organisation do you work for and what is your position?
2. How much experience do you have with the operation of the LC-MS/MS and
3. How many samples have you analysed with the LC-MS/MS yet?

Each pair introduced each other. It became clear that very few participants of the course had actually worked with the LC-MS/MS. The total number of samples analysed was very limited and no marine biotoxins in fish were analysed yet in Malaysia.

After the getting to know the participants were asked to write on coloured cards what they hoped to learn from the training course. These learning objectives can be found in Annex III.

Introduction to marine toxins and RIKILT

After introducing the organisation RIKILT – Institute of Food Safety Arjen Gerssen of RIKILT gave a general introduction on marine biotoxins. Different types of toxin producing algae, their occurrence as well as intoxication syndromes that can be obtained after consumption were discussed.

Official methods and legislation

After discussing the various types of legislation such as EU, CODEX and FDA an assessment was done on which toxins should be monitored for export but also awareness was created that testing on these toxins is also important for the Malay shellfish consumers. An assessment was done on:

1. Which toxin group should be monitored as requirement by the EU, CODEX and FDA?
2. For which toxins methods of analysis are available in Malaysia?
3. For which toxin is a monitoring plan available?

Based on the outputs of the assessment, the different regulatory limits (EU, CODEX and FDA) were discussed. This assessment clearly makes visible where the gaps are and where method development and establishment of monitoring programs should take place.

ISO 17025 and working towards accreditation in Malaysia

Beside the training in the lipophilic marine biotoxin method the training also included the subject working towards ISO17025 laboratory accreditation. The session started with group work. The participants were divided into 2 groups and asked to discuss the following questions:

1. Define accreditation, what is it?
2. Why is accreditation important?
3. What is needed for a laboratory to get accredited/ What needs to be in place?
4. What does Malaysia already have in place? What has been done?

After 30 minutes discussion the results of each group were presented to each other. The different aspects in ISO17025 were then discussed guided by a PP presentation and the participants were asked to pay attention to the various technical accreditation aspects during the lab training sessions. Particular attention was given to the definition of accreditation in the context of ISO 17025; competence of testing and calibration of laboratories. The difference between accreditation, registration and certification was also discussed. The importance of accreditation is not only important for laboratories to be recognised but also it ensures that data has been collected according to set procedures.

Both the management and technical requirements for accreditation were discussed. From a management perspective it is for example important that the staff is properly supervised, all documents are controlled and a clear management system is present. And the technical aspects are more lab based and some examples were given such as identification/labelling of equipment, regularly check performance of all equipment. Of course it was pointed out that the importance of getting ISO17025 accreditation is clear as this will break down barriers for international trade.

The participants recognised that the accreditation process entails more than initially was thought about. They learned that Malaysia does not yet have all requirements in place and that all staff needs to be involved and informed. Equally important is the support of the top level management to ensure sufficient resources are made available to ensure the equipment in the laboratories can be maintained, sufficient samples can be analysed and budget is available for operation and capacity development of staff.

Note:

Malaysia aims to have the laboratories accredited by next year. However during the training it became evident that although laboratories are following procedures during analysis little has been documented and few monitoring plans are developed. The discussion around accreditation seems to be very much lead by a few persons. However the requirements of accreditation should be clear to all staff and management. Everybody should be aware on what it entails.

It is important that accreditation and capacity development of staff in methods of analysis is not only driven by the requirements set by the EU when exporting products. Ensuring safe products for the local market should have priority as well.

Day 2, June 21

Reflection

General introduction to MS and MS trouble shooting

At day two the lab session of the training started with a brief general introduction into liquid chromatography tandem mass spectrometry. The trainers found the laboratory very well equipped and a lot of investment was done over the past few years in the procurement of new equipment. For some of the participants the LC-MSMS introduction was a repetition of previously followed training courses but for others this was new information.

The first practical part the setting up the mass spectrometer for lipophilic marine biotoxins was done with standards prepared by the RIKILT trainers. Later in the course this step including calculations on the preparation of standard solutions were made by the participants themselves supported by the trainers. The preparation of standards should be general knowledge of laboratory staff which was not the case at the beginning of the training but the participants improved greatly on this aspect.

Aspects related with ISO17025 were directly communicated in an interactive manner during the laboratory sessions. Discussions were facilitated on proper labelling of chemicals (track and trace), safety issues (waste management etc.) but also the importance to know what your colleague is doing in the laboratory.

Day 3, June 22

Explanation of SOP and relation with ISO17025

Supported by a PP presentation with visuals the quantification and confirmation of lipophilic marine toxins in relation to ISO17025 was discussed. It was clearly outlined which steps are included in the Standard Operating Procedure (SOP) and details related to accreditation were highlighted. The participants followed used the RIKILT standard operating procedure (SOP) and were asked to keep in mind the accreditation lessons learned and take notes for points of improvement during laboratory work (observation).

Laboratory session

RIKILT brought samples to be analysed for lipophilic marine toxins. These samples were also used for the official method validation which was performed by RIKILT within Europe (RIKILT report 2011.008 available at <http://www.rikilt.wur.nl/NL/publicaties/Rapporten/>). The samples were analysed overnight and on day four data interpretation of these analysis were done.

Day 4, June 23

Integrating and calculating the results of wednesday

On day four the interpretation of the data analysed overnight was done. Quality control and statistical evaluation showed an excellent performance of the whole group which indicated that basic laboratory skills are present such as accurate weighing, pipetting. It was observed by the trainers that also the lessons learned with respect to labelling were applied.

Analysis of Malaysian shellfish for lipophilic and PSP toxins

In the afternoon of day four the whole lipophilic marine biotoxin procedure was repeated with Malay shellfish samples and the participants were requested to work more individually.

Day 5, June 24

Discussion of the lab results of Thursday

On the fifth day, Friday morning, results of the Malay shellfish samples were presented and again the quality control was excellent. In the afternoon during the reflection each participant was asked to write down his or her remarks with respect to accreditation and/or actions needed in the lab (Table 1). It was pointed out by the trainers that this exercise was not to criticise the laboratory facilities where the training was held but each participant should also do a critical evaluation at its own lab facilities. Overall the trainers advised the participants to start to work with the equipment and accreditation process at their lab as soon as possible.

Evaluation of the training

They made a comparison between what they learned in the past week and what their expectations (learning objectives) were at the start of the training. The participants reflected back what they have learned. They were asked:

- How confident are you to do the extraction procedure

They were then asked to list the positive points and the points that need improvement and give the reasons why for the following topics:

- Content of the training
- Training approach followed
- Trainers

The evaluation conducted at the end of the course showed that the participants felt the objectives of the course were met. They appreciated the interactive approach that was followed and the combination of theory with practical sessions in the laboratory. Most participants would have liked the training to last longer and felt that more hands on experience is needed to be confident to work with the LC –MS/MS. The results of the evaluation can be found in Annex IV.

After the evaluation the course was closed and certificates were handed out.

Findings and Recommendations

First of all, the participants were very eager to learn from the experts and they gained a lot of knowledge on both the trained method of analysis as well as the accreditation process.

On the remarks made by the trainers with respect to health and safety during laboratory sessions, direct action was taken by the responsible person. This shows the right attitude when aiming for accreditation under ISO17025.

Some other lessons learned during the training course on marine biotoxins were the following:

- Laboratory staff lack of skills for the calibration, operation and trouble shooting of the LC/MS-MS
- Transfer of staff to other positions in the Department of Fisheries complicates matters in building sufficient capacity for the laboratories to be operated effectively over the long run
- Maintenance of the LC/MS-MS equipment and is expensive
- The staff does not have enough expertise and confidence to do method validation
- Not all staff in all Biosecurity Centres are equally involved and informed about the on-going process towards lab accreditation (e.g. 2 labs are in the process of accreditation and quality manuals are written but the staff on the workfloor are not aware of this)

In response to these findings it is advised to conduct a more thorough gap/needs analysis before a training is requested for. From such assessment it should become clear which knowledge gaps exist and

what the specific needs in the laboratories are. This assessment will also provide a better insight on the knowledge and skills of the future course participants. Based on this the learning objectives of the courses can be formulated.

The current situation is that a specific method and knowledge gap is identified and a training is organised around this topic. It will be more effective to build capacity of the laboratory staff (from extraction to analysis on generic method development for various equipment (such as LC-MS/MS) than only on specific components so that the participants gain general knowledge and skills in method development and understand the theory behind the different approaches. In the end it should not matter to analytical chemists if they develop a method for veterinary drugs or marine biotoxins as the same principles and approaches apply.

It is therefore recommended to develop general knowledge and build capacity in extraction procedures, liquid chromatography and mass spectrometry in order to make the Malaysian Biosecurity Centres and other laboratories more independent and more flexible to setup their own methods and work towards the development of specific SOPs for the laboratories in Malaysia. From a capacity building point of view the same people should attend these training courses to facilitate their learning by following a stepwise approach to increase their knowledge and skills.

These activities will ensure that the lab staff will gain more hands on experience and confidence with the handling of the high tech equipment and will be able to analyse more components on 1 equipment. The knowledge and skills gained will be better institutionalised. It could also help facilitate the accreditation process so it is better communicated to all involved in it.

It is also recommended to let laboratory staff perform the whole procedure from extraction to MS analyses to data analysis individually and also be responsible for taking care of their (chemical) waste. This way the person involved knows exactly what happened with the samples from A-Z and can explain results much better than when different persons are involved in the procedures.

During all laboratory sessions the participants were asked to write down the points in the Kuantan laboratory that need improvement for the laboratory to become accredited under ISO17025. During and at the end of the training these points were listed and discussed with the trainers (see Table 1)

Table 1. Summary of the observation made by the participants during their laboratory work in relation to implementation of ISO17025. If (2x) is mentioned this issue is raised by 2 individual participants.

Remark by participants	Reaction of trainers
Management	
<ul style="list-style-type: none"> - Funding / Budget - Building personal qualification number of staff (2x) - Support from top management - Entrance to the building should be fully secured - Emergency exit plan (from each room) - Monitoring temperature for freezer and refrigerators + track and tracing - Authorization of reproduce SOP - The equipment must have flow chart - Log instrument numbers - Written instructions (technical SOP) for the equipment - No adequate instructions on the use of the equipment 	<ul style="list-style-type: none"> - Beside the number of staff it is also important to keep the staff for a longer period in the same function in order to build capacity and have some knowledge transfer. Enough funding and support is needed also for maintaining the accreditation (management) - Documents (and document control) on emergency plans, SOPs on both methods and equipment, monitoring of performance, instrument performance should all be present - Also the entrance of the building should be secured in such a way that nobody can enter unattended the building. So visitors should be guided by employees - Training of staff should take place when new procedures and/or equipment are going to be used (qualified people)

<p>Track and tracing</p> <ul style="list-style-type: none"> - Records, lab journal, logbook for maintenance equipment, visitor records, utilities, consumables, stocks, reagents and chemicals (3x) - Consumables should kept in a labelled cupboard - LC-MS/MS equipment should have a logbook (History of service, persons using LCMSMS) - Calibration pipette, balance, freezer / refrigerator - Labelling bottles all equipment (3x) - No unique identification of LCMSMS and compartment 	<ul style="list-style-type: none"> - In order to review results it is important that clear track and tracing of all important procedures are done - For example; <ul style="list-style-type: none"> - labelling of chemicals and standards to ensure that the correct non-expired chemicals are used - Logbooks, lab journals etc. where all important lab procedures are written down so that a supervisor can check the results - Also write down your mistakes as this will help to explain strange results or are a lesson for colleagues to pay extra attention to a certain procedure. For example if the same mistake happens more often by different people maybe the SOP should be revised - Identification and labelling of equipment (LC-MS/MS, freezers, pipettes etc.)
<p>Safety/Health</p> <ul style="list-style-type: none"> - Wash Hand before and after doing analysis, using gloves when handling solvents (6x) - All dangerous chemicals should put in fume hood example: ACN/MeOH used are put in the fume hood after used - Chemical must be located in the correct cabinet e.g. acid and strong alkaline cannot be placed together(air circulation) (7x) - Don't let solvents evaporated to air Working in fume hood / enclosed during pipetting and titrating (3x) - Fume hood fully utilised it (chemicals etc) - The space for doing lab activity must be comfortable - Environmental condition To hot invalid test results: temperature of the instruments will effects the results (3x) - Must be follow correct method / SOP - Make preparation before doing analysis 	<ul style="list-style-type: none"> - Of course personal hygiene is important but also wearing a lab coat, glasses and gloves when necessary. A safety officer should pay attention if these rules are followed and corrective actions should be taken - Waste management and correct storage of chemicals, acidic, alkaline, organic and halogenated chemicals should be stored in well ventilated special cupboards (and not together) furthermore waste of these solutions should be treated correctly and collected separately and not end up in the sink - Also these measures are management task for ISO 17025 accreditation, taking care of you employees do not expose them to harmful chemicals but also a nice working environment (i.e. air conditioning)
<p>Quality Control</p> <ul style="list-style-type: none"> - Arrangement of equipment according to quality manual / SOP. Water bath, balances, centrifuges storage facilities (5x) - Checklist during sample preparation - Put the pipettes in a proper position 	<ul style="list-style-type: none"> - Use the equipment for its purpose i.e. use the correct pipette for the volume you want to pipette - Regularly (weekly, monthly, yearly) check the performance of the equipment (i.e. check sensitivity and mass accuracy MS, if the pipette volume is correct, balance is correctly weighing, water bath temperature is correct, freezer temp is correct, etc.)
<p>Capacity of staff</p> <ul style="list-style-type: none"> - Skill technical pipette - Pipetting process must be done carefully 	<ul style="list-style-type: none"> - Technical skills are a requirement for ISO17025 so proper training in whatever procedure should be done (people at the entrance should be trained how to receive guests, packages etc, lab managers how to manage their people, quality managers how to perform an internal audit and lab technicians how to operate the equipment).
<p>Waste management</p> <ul style="list-style-type: none"> - Toxins, sharp object / glasses, wastage of chemicals should be thrown in proper place (3x) - Lab must be clean also when doing sample preparation 	<ul style="list-style-type: none"> - See the remarks made with the Safety and health

ANNEX I - LIST OF PARTICIPANTS

*Training on Marine Biotoxin Analyses
Fisheries Biosecurity Centre, Kuantan Pahang, Malaysia
20th – 24th Jun 2011*

No.	Names	Position and Office
1.	Azahari Bin Othman	Senior Fisheries Officer - G48 Fisheries Biosecurity Centre Kuantan, Pahang
2.	Ahmad Saifullah Bin Mohammad	Fisheries Officer - G41 Fisheries Biosecurity Centre Kuantan, Pahang
3.	Bakri Bin Saad	Assistant Science Officer C27 Fisheries Biosecurity Centre Kuantan, Pahang
4.	Maisarah Bt. Abdullah	Laboratory Assistant - C17 Fisheries Biosecurity Centre Kuantan, Pahang
5.	Azlan Bin Salleh	Laboratory Assistant - C17 Fisheries Biosecurity Centre Petaling Jaya
6.	Belayong Anak Nyuak	Senior Fisheries Officer - G48 Fisheries Biosecurity Centre Bintawa, Sarawak
7.	Siti Nadirah Bt. Abdullah	Assistant Science Officer - C27 Fisheries Biosecurity Centre Bintawa, Sarawak
8.	Ainah Bt. Puyong	Fisheries Officer - G41 Fisheries Research Centre Likas, Sabah
9.	Mohd. Nor Azman Bin Ayub	Senior Research Officer - Q44 Fisheries Research Institute (FRI) Batu Maung, Pulau Pinang
10.	Mohamd Shafie B. Buchik	Ministry of Health, Malaysia

ANNEX II - COURSE PROGRAMME

Course Programme from Day to Day

TRAINING COURSE ON MARINE LIPOPHILIC TOXINS USING LC-MS/MS

20 – 24 June 2011

Course Coördinator:	Hamdan Jaafar Ingrid Gevers
Resource persons	Arjen Gerssen Mirjam Klijnstra

MONDAY 20 JUNE

09.00 – 09.45	Azahari / Hamdan	Opening of the course
09.45 – 10.30	Gevers/Gerssen	Introduction Wagening UR- Centre for Development Innovation & RIKILT Institute of Food Safety
10.45 –12.15	Gevers	Getting to know each other Experience with LC-MS/MS and biotoxins Expectations and learning objectives Introduction to the programme House rules
12.15 – 13.00	Gerssen	Introduction to Marine toxins
14.30 – 15.30	Gerssen	Official methods and legislation (EU, CODEX)
15.30 – 17.30	Gerssen/Gevers	ISO17025 and working towards accreditation in Malaysia

TUESDAY 21 JUNE

09.00 – 09.15	Participants/Gevers	Reflection on the previous day on lessons learned
09.00 – 10.00	Gerssen/Gevers	Requirements for accreditation
10.00 – 11.00	Klijnstra	General introduction to MS
11.00 – 12.00	Klijnstra/Gerssen	MS trouble shooting in the laboratory
12.00 – 13.00	Klijnstra/Gerssen	Lab session, MS infusion experiments
14.30 – 17.30	Klijnstra/Gerssen	Lab session, MS infusion of standards
20.00	Azizi/Rummenie/Chong/ Hamdan	Opening dinner & official opening of the course

TRAINING COURSE ON MARINE LIPOPHILIC TOXINS USING LC-MS/MS

20 – 24 June 2011

Course Coördinator:	Hamdan Jaafar Ingrid Gevers
Resource persons	Arjen Gerssen Mirjam Klijnstra

WEDNESDAY 22 JUNE

09.00 – 09.15	Participants/Gevers	Reflection on the previous day on lessons learned
09.15 – 10.30	Gerssen/Klijnstra	Discussion and reflection of results of Tuesday
10.45 – 11.45	Gerssen	Explanation of SOP and the relation with ISO17025
11.45 – 13.00	Gerssen/Klijnstra	Lab session: setting up the chromatography (lipophilic marine toxins and PSPs)
14.30 – 17.30	Gerssen/Klijnstra	Lab session: Extraction and analyzing validation samples (lipophilics) and PSPs

THURSDAY 23 JUNE

09.00 – 09.15	Participants/Gevers	Reflection on the previous day on lessons learned
09.15 – 11.15	Gerssen/Klijnstra	Integrating and calculation results Wednesday
11.30 – 12.00	Gerssen	Quality assurance and comparing results with RIKILT Study
12.00 – 13.00	Gerssen/Klijnstra	Lab session, the analysis of Malaysian shellfish for lipophilic and PSP toxins
14.30 – 17.30	Gerssen/Klijnstra	Lab session, the analysis of Malaysian shellfish for lipophilic and PSP toxins

FRIDAY 24 JUNE

09.00 – 11.00	Gerssen/Klijnstra	Discussion lab results of Thursday
11.00 – 12.15	Gevers/Gerssen/ Klijnstra	Technical part of the ISO17025 and discussion on steps to take towards accreditation
12.15 – 14.30		Time for prayer & Lunch
14.30 – 15.15	Gevers	Evaluation of the training'
15.15 – 15.30	Hamdan/Gerssen/ Klijnstra/Gevers	Closing ceremony and handing out certificates

ANNEX III - LEARNING OBJECTIVES

Legislation

- EU legislations

Methods to analyse biotoxins

- Methods of marine biotoxins analysis
- To know the 'official' method for analyzing the toxins
- Learn new method to analyse marine biotoxin + extraction procedure
- Clearly know about biotoxin analysis (why and how)
- Biotoxin sample analysis from preparation to instrument (method, steps)

Sample preparation

- Sample preparation recognized method by EU
- Learn about extraction and the concept on biotoxins analysis
- Know how to extract the sample (method)
- Sample preparation for marine toxin sample, using LC-MS/MS
- Learn on sample preparation in marine biotoxins sample

LC-MS/MS operation, maintenance & trouble shooting

- Analysis of biotoxin by using LC-MS/MS
- To learn to use LC-MS/MS in biotoxins
- LC/MS-MS handling
- Troubleshooting LC-MS/MS
- LC-MS/MS operating system (learn to operate and troubleshooting)
- Principal & theory LC-MS/MS (operate & maintenance)
- Hands on LC-MS/MS
- Operational use with LC-MS/MS

Data Analysis

- Learn more in analysing & interpreting data using LC-MS/MS
- Interpret LC-MS/MS result, reporting result

Accreditation (ISO17025)

- To know about the ISO17025 implementation
- Accreditation of labs
- Requirements of lab accreditation
- Learn about how to get working toward accreditation ISO17025
- Accreditation of ISO17025
- Procedure of accreditations
- Accreditation of ISO17025 (task/role, process)

SOP

- Proper write up of SOP

ANNEX IV - EVALUATION

Training method/approach

Positive

- Well suited to course contents but time a bit short
- Hands on training make every participant understand and “feel” the test
- Interactive approach give all participants involved in discussion of any problems can be raised-up before complicated
- Training method samples preparation = excellent. LCMSMS calculation & interpretation data
- Good to have introduction and briefing before doing the lab work / discussion of results after doing lab work
- Hands-on / participation / demonstration
- Easy to understand / follow
- The method make it easy to understand / the method is simple but must follow the correct procedure
- Very good content, just need more time

Negative

- Give more video presentation: video on accredited lab and sample preparation

Trainers / facilitators

Positive

- Articulate the topics very well & eager to help & explained to us
- Transparent & really willing to help give a clear explanation to many questions
- Very good coordination / everybody get involved / time is fully optimised
- Very friendly / learning process that is easy to understand
- Friendly / easy for me to understand
- Friendly / understanding / sharing
- Excellent / easy to understand / methods of teaching attitude.
- Explanation from trainer very clearly easy to understand
- Friendly / good attitude
- Eager to teach us

Negative

- Time management, sometimes time deviation happens

Content of course

Positive

- Good because all aspects of marine toxins lipophilic are covered / Theory –principle – methods of extraction + analysis / The course also taught us about accreditation, it's definition , what are the requirements & things to do in order to be accredited.
- Comprehensive course for lipophilic biotoxin given - sample preparation till using LCMSMS / new method for lipophilic extraction / demo for data analysis
- Learning new toxin extraction (DSP) and analysing with LCMSMS
- Good / learn a new method of DSP / Learn preparation of sample / Have hand on experience with LCMS/MS
- Learn a new thing about using LCMS/MS and preparation of standards and samples
- Can learn about sample preparation / Using LCMS/MS,how to lay in data in sequence

- Satisfied = covered all syllabus / informative / data analysis (calculation) 2 week course.
- Data interpretation still needs to improve – test for handling LCMSMS (included)
- Yes: get many new knowledge: many useful information
- Everything that we should know is inside the training

Negative

- Time short: 5 days is still not enough to learn every content, especially the 1st time course
- Expected to learn about PSP&TTX using LCMSMS
- Not enough time to practise data analysis

How confident are you now to use the LCMS/MS?

Scoring / Why

Low			High	
1	2	3	4	5
1 dot	2 dots	2 dots	3 dots	No dots

- 3 / Because I never having basics using LCMS-MS. Before this I only run HPLC instrument
- 3 / Need more practice using LCMS/MS
- 4 / Still need to practise using it / hand on training
- 4 / But trouble shooting = 2 / Identifying problems = 2
- 1 / Miss 1 day training and need more training to use LCMS/MS
- 3 / Need more practice with the LCMS/MS
- 4 / Hands-on more on LCMSMS – selection of the mass
- 2 / Still new with the LCMS/MS but as lab assistant my job more focus about sample preparation

How confident are you now to do the extraction procedure?

Scoring / Why

Low			High	
1	2	3	4	5
No dots	No dots	1 dot	3 dots	3 dots

- 4 / More practice
- 3 / Need more practice – test run
- 5 / Because it's simple comparing to another analysis method – Have done many extraction before
- 4 / Need more practice
- 4 / To maintain the score commitment and practice is needed
- 5 / Quite confident – SOP very clear

Which skills or knowledge are still needed?

- Understanding the result / calculation / reports
- Method validation, procedure & report
- Principle calculation of CC α & CC β
- Operation of trouble shooting / analysis using software
- Results interpretation (analysis) / more extraction method practice

- Learn more about sample / standard preparation
- Need more practice about LCMS/MS software / trouble shooting / interpret the data
- LCMS/MS / the time is too short
- It's new for me
- Want to know the basics for using LCMS/MS from first until final – trouble shooting
- More detail about LC-MS/MS from 1st step until interpretation data (more practice)

Training Course Sample Preparation and LC-MS/MS Analysis for Stilbenes and Nitroimidazoles

14-18 November, 2011

Kuantan, Malaysia

Paul Zootjes, Martien Essers, Ingrid Gevers

Summary

The Department of Fisheries of Malaysia is recognised as a Competent Authority by the EU since 2010. It has issued tenders to purchase more advanced test equipment such as GC TOS MS, ICP MS and additional LC-MS/MS to enhance the number of samples and parameters to be analysed in the laboratories. Consequently there is a need to improve the capability and competence of the technicians and laboratory staff manning and operating these equipment's. There is not only a need to upgrade the analytical capacity of the laboratory staff directly involved in the analysis and detection of forbidden substances, but also to improve their skills and knowledge to analyse additional parameters. Through knowledge transfer and laboratory enhancement this BOCI project will support the laboratory's process towards getting accreditation under ISO 17025.

The short term objective of the BOCI project is to increase the knowledge and skills of Ministry of Health (MoH) and Department of Fisheries (DoF) as EU-accredited competent authority (CA) . The long term objective is to have a national body that provides good governance, effective control over the entire production chain from farm to table, with the ultimate goal to help build a credible inspection and monitoring system that can guarantee safe quality products.

The project follows a training of training approach that allows the Department of Fisheries in Malaysia to increase the capacity and competence of all relevant staff on methods of analyses over a wider spectrum of parameters. Subsequently this will result in improved Laboratory Services as an important component of the Food Safety and Quality Assurance System. Eventually the greater capacity and capability of the laboratories and their staff will result in accreditation for official analyses.

This training course on sample preparation and LC-MS/MS Analysis for Stilbenes and Nitroimidazoles contributed to the achievement of the long and short term objective. The course was implemented from 14-18 November, 2011 in Kuantan Malaysia. The course was implemented in partnership by Wageningen UR - Institute of Food Safety (RIKILT) and Wageningen UR Centre for Development Innovation (CDI). RIKILT provided the expertise on laboratory tests and - practices, and CDI was responsible for the overall design of the training curriculum, overall project management and facilitation.

The course focused on Stilbenes and Nitroimidazoles sample preparation and analysis with LC-MS/MS equipment. The training was guided by a dialogue on laboratory accreditation and identified gaps to achieve this. Recommendations were given to the Kuantan laboratory to improve procedures, infrastructures and operation to work towards this ISO 17025 accreditation. The course was highly interactive using a combination of theoretical interactive PP presentations and practical laboratory work aiming to improve the analytical skill of the participants. Theory and practice were alternated throughout the course.

A total of 9 participants working at different laboratories of the Biosecurity Centre under the Department of Fisheries in Malaysia, the National Public Health Laboratory of the Ministry of Health and the Veterinary

Public health Laboratory under the Department and the Veterinary Services participated in this course (see Annex I).

The daily course programme is given in Annex II. The course topics were all in line with the course objectives described in the proposal of the course. The content and modules of the course were the same as given in the original proposal but allowed for changes based on the expectations of the participants. The course facilitators followed a flexible program that allows adaptation to the specific needs of individuals and the group. Each day the group reflected on the lessons learned the day before. The course included an 'end-of-course' evaluation in which the participants gave their feedback about the content of the course, the modes of instruction and the quality of the resource persons and facilitators. Additionally at the end of the course the trainers reflected on what they learned during the course and gave feedback for improvement.

Following you can find a short description of the different daily sessions included in the training course.

Day 1, November 14

Introduction to the course

The first day started with a getting to know session. The participants were asked to work in pairs and get to know their colleagues by answering the following questions:

1. What is your name, what organisation do you work for and what is your position?
2. What do you do in the laboratory?
3. Which problems in residue analysis do you encounter at the lab?

After a brief discussion the participants introduced each other. After this session the participants were asked what they hope to learn during this week. The learning objectives can be found in ANNEX III. After agreeing on house rules the experts from RIKILT introduced themselves and provided an overview of the programme of the training.

The curve of DEMING was discussed to remind the participants that quality is a process that's always on the move. Some main rules in quality are explained:

- Describe what you do
- Do what you have described
- Prove that you have done what has been described

Presentation about Quality control Samples

This presentation gave an overview about quality control samples when using a LS-MSMS system: matrix matched standards (MMS), matrix matched recovery standards (MMRS), 1st line control, standard addition and the use of the Shewhart control chart .

Opening of the course

Mr Ahmed Hazizi Bin Azziz, the Director of the Fisheries Biosecurity Division of the Department of Fisheries in Malaysia, Mr Hamdan Jaafar, Head of the Fisheries Biosecurity Centre and Mr Adnan, Director of Pahang State Fisheries opened the course and welcomed the participants. Ingrid Gevers of Wageningen UR Centre for Development briefly introduced the objectives and the content of the course.

Presentation From theory to practice

Explanation about sample receipt, registration and traceability of data in relation to SOP and the sample's in relation to the ISO 17025. In the presentation was an explanation about the SOP for analysing Stilbenes in fish step by step. The same procedure was followed for analyzing Nitroimidazoles in fish. The

participants were divided in groups and where ask how to preparing standard mixes. Each group had to provide a calculation and explanation.

Practical Training preparing standards

Under the supervision of the trainers the participants where ask to prepare stock solutions by calculation the amount, weighted the amount of standard and dissolve. After preparation the stock solution the participants prepared the mix spike solutions

Day 2, November 15

Reflection

The participants reflected back on what they learned the day before. They all shared their most important lesson learned which ranged from appreciating the mix of participant of different ministries and laboratories to the more technical aspects of sample preparation. The following lessons learned were mentioned:

- All stages of extraction of stilbenes
- Better understanding of the SOP; clear instruction were provided
- To remove the heptane layer using the pump
- How to work with the Ultra-Turex
- How to mix the sample using head over head at speed 5
- Understand better what we are doing in order to avoid mistakes
- If we find some problems we have to be more creative & innovative to solve them (example trap made by experts)
- Before starting you have to make a good plan to do your analysis
- Conversion of RPM to G
- To include purity and correct molecular weight in calculation standard concentration
- Preparation of mobile phase for stilbenes
- RIKILT procedure in doing MV&MU
- Chemical principles
- Learned how to siphon/draw the upper layer using a pump (we usually use a pipette)

The participants were also asked on Monday to write down during the laboratory work which practices in laboratory are already complying with ISO17025 and which practices still need to be improved.

Practical laboratory training Stilbenes

The participants were divided in 4 groups, each group prepared 4 sample's following a check list provided by the trainers. During the sample preparation the trainers answered questions and gave information and tips how to get the best results.

Day 3, November 16

Reflection

The participants reflected back on what they learned the day before. They all shared their most important lesson learned which ranged from appreciating the mix of participant of different ministries and laboratories to the more technical aspects of sample preparation. The good practices in the laboratory already complying with ISO17025 and the points for improvement that were written down asked on Monday during the laboratory work were listed. The points of discussion can be found in Annex IV. On Friday additional points will be discussed further in a plenary session and the experts will give recommendations on how to improve certain practices for better compliance.

Practical laboratory training Nitroimidazoles

The participants were divided in the same 4 groups as the day before, each group prepared 4 samples following a check list provided by the trainers. During the sample preparation the trainers answered questions and gave information and tips how to get the best results

Day 4, November 17

Reflection

The participants reflected back on what they learned the day before. They were asked to answer 3 questions individually:

1. What was the most interesting lesson learned?
2. How will you transfer knowledge and skills to your colleagues?
3. How will you continue to collaborate with each other (between Ministries and between laboratories).

The participants provided the following feedback in response to the questions

Most interesting individual lesson learned:

- Learned a shorter extraction procedure for nitroimidazoles from what we are doing now in my lab
- The new method of standard preparation
- Always to label before starting
- Knowledge about complete procedure on nitroimidazole sample extraction
- For the evaporator step we need to add MeOH for dry up the sample if the sample is not dry enough (2)
- For SPE step we need to dry the sample with vacuum correctly. It must be dried properly
- Sample preparation for stilbenes and nitroimidazoles. The SOP is easy to understand
- On SPE washing step: column should be dry
- Evaporator: how to dry the sample quickly

Transfer of knowledge and skills

- We will have internal/echo training of the colleagues within 1 month after the training
- Would like to try this method together with my colleagues
- Share all new 'tricks' taught by the expert
- I will conduct an in house training for my staff
- Pass on the training notes and presentation to my colleagues
- As soon as possible I will practice together with my colleague (every month a in house training)
- I will teach my colleagues the technique to draw/siphon the upper layer using the pump
- I will tell my colleagues about the importance of taking care of the quality of the chemicals that we are using: 'What goes out must stay out'
- Make the training & practice with the colleagues and share the knowledge that I had about the SPE step and evaporation step
- Make a proper plan to prepare for a training. Then order chemicals which are not available (can approach other labs for this). Set up the methods and organise an in-house training

Collaboration

- Keep in touch with each other
- Conduct interlaboratory testing
- We will keep in touch via e-mail, phone, facebook to share information (2)
- Keep in touch with each other (through phone, e-mail) if we face any problem or better way to perform the analysis
- Always communicate through e-mail, phone (2)
- Have meetings to discuss

- Invite another lab for in-house training to improve technical skills
- We can do more training together at different locations
- Through a community of practice or a platform where information can be exchanged.

A small discussion followed about the importance to share knowledge and information internally. But also to stay in touch to keep on learning from and with each other. Already a committee is established in Malaysia for information sharing on veterinary drugs. Ministry of Health, Department of Fisheries, Chemical department and 2 universities are members. The participants of this course could participate in this committee.

Note:

It was mentioned that ordering the materials (standards etc) take a long time (up to 3 months) and is costly since procedures are put in place to order it through a company. This makes it difficult to quickly do an analysis or conduct a follow-up of this training at other labs. For now the labs agreed to share standards needed for stilbenes. But this dependence of the company makes it hard to act quickly.

The experts of RIKILT are very willing to have a look at the worksheets that are generated once the analysis of stilbenes and nitroimidazoles are replicated in the other labs. Feedback will be given for improvement.

The experts also advised that it is possible to participate in the ring tests that the EU organises. This way more experience with the analysis of samples can be gained. It is anonymous and feedback will be given to the results

Practical laboratory training LCMSMS

The participants got LC-MSMS introduction by interaction between the participants, the trainers and trainers of Thermo. Information about Ionisation technique, Tune parameters (Cone voltage, Cone gas flow, Capillary voltage, Source temperature) were directly carry out on the apparatus. Second part of the LC-MSMS training was the interpretation of data from all measured Stilbenes and Nitroimidazoles samples. This was done by the use of an excel spread sheet provided by the trainers. This spread sheet is based on the criteria in the EU document 657.

Day 5, November 18

Evaluation trainers day by day

The trainers gave their feedback from day to day in the form of a PP presentation. In this presentation also the results were presented, explained and discussed. On request of the participants there was an additional explanation about the calculations in the spread sheet provided by the trainers on Thursday. As a reminder a enumeration of ISO 17025 management requirements, technical requirements and examples of content of a SOP and working documents.

Reflection/Evaluation participants

The participants reflected back what they have learned. They made a comparison between what they learned in the past week and what their expectations were at the start of the training. The participants were divided in groups and discussed the training, learning targets, approach and where the training could be improved. The conclusion was that a lot of questions were handled but some have not been answered.

After the evaluation the course was closed and certificated were handed out.

Recommendations

To support the Kuantan laboratory in its process towards getting accreditation under ISO 17025 the trainers provided recommendations for improving the laboratory facility and personal safety. Besides the list given in Annex IV the following points also need attention:

1. Under the fume cabinet and laboratory cupboards no storage of chemicals
2. Chemicals stock in cold room, with sufficient ventilation
3. Special Chemical cupboard should have direct ventilation, also in the laboratory's
4. Storage of standard in marked racks
5. Storage of standards and samples in separate refrigerators
6. Purchase special waste bottles for different fluids and market stickers: Halogen pore, acids etc
7. All the bottles with chemicals should be in leak reservoir (Tray).
8. Do not wear open shoes in the laboratory
9. Introduction of journals and logbooks
10. Equipment have to be validated or calibrated (MS, pipets, balances)
11. Temperature registration of coolers and refrigerators
12. Purchase of non-return bottle for SPE reservoir
13. Purchase of column oven for the LC-MSMS
14. Use UPLC columns on the LC-MSMS (better performance and sensitivity)

Throughout the course the trainers have noted down points that need additional attention.

Recommendations for improvement:

1. The labs are well equipped. It is now of utmost important to keep developing the knowledge and practical skills of the laboratory staff that have to operate them. To not only build individual capacity of the staff but also strengthen organisational capacity it is important to think about how to follow up on the training courses given by Wageningen UR.
2. After the first training on marine biotoxins (June 20-24, 2011) very few samples were analysed after finalising this course. To really build the required expertise and capacity in the laboratories it is crucial that the laboratory staff keep practicing the newly learned methodology and knowledge. Especially since the experience with operating the LC-MS/MS equipment is still very limited. Only through learning by doing the participants will master how to operate the equipment and use the software. It is therefore advised that the laboratories will send each other spiked samples for analysis. RIKILT is also willing to support this process. Alternatively the laboratories could get involved in ring tests. It is equally important to budget for the standards and other materials that are needed to run such analyses.
3. It is essential to become more efficient in the exchange of knowledge and experience between the different laboratories in the country and to continue a learning process at organisational level. Suggestions on how this could be approached are:
 - a. The different labs in Malaysia (DOF) should support each other in capacity development. Some staff have more experience than others. Let them function as resource persons during internal training course to exchange knowledge. This is especially important since some of the staff have a background in fisheries management and lack relevant expertise in chemistry.
 - b. Allow staff from Kuantan to work at Kuala Lumpur laboratory for a week (or more) to gain hands on experience with LCMSMS and other laboratory equipment. Let them analyse samples (e.g spiked ones) by going through the steps of sample preparation and let them practice with the operation of the LCMSMS and the data analysis (software).
 - c. Share methods developed for the analysis of a group of compounds between laboratories in Malaysia. If one lab has already developed a certain method other labs can learn from them and benefit.
 - d. In some areas it became evident that the staff of the MOH have a better chemical background. It is advised to strengthen the collaboration between the Ministry of Fisheries

and the Ministry of Health and explore if it is possible for staff of the labs of the Biosecurity Centre to be trained in house at the Ministry of Health

4. Although the lab in Kuantan is not yet analysing real samples from the field, they could be given spiked samples so they can continue to gain experience while analysing them. This way the lab staff will gain experience and confidence to do the analyses. Management needs to strongly supervise and encourage this.
5. The laboratory in Kuantan is equipped with 2 LCMSMS with the reason to measure different groups of compounds with 1 machine. Although this is a good idea once sufficient numbers of samples will be measured. However to be more efficient and cost effective it is advised to analyse more groups of compounds on 1 machine. The seems to be excessive to have more than 1 when capacity of staff in its operation still needs to be build. The budget spend for this equipment could have been used for other purposes.
6. Instead of equipping the new lab in Kuala Lumpur eventually with a new LCMSMS (the present one is from 2004) one LCMSMS unit of Kuantan could be transferred to Kuala Lumpur. Budget saved this way could then be allocated to support capacity development of staff. This way exchange of LCMSMS knowledge and problem shooting experience can be shared and one can support each other more efficiently.

ANNEX I - LIST OF PARTICIPANTS

*List of Participants for Training Analyses on Stilbenes & Nitroimidazole
14th – 18th November 2011
Fisheries Biosecurity Centre, Kuantan, Pahang Malaysia.*

No.	Name	Designation and Office	E-mail
1.	Ahmad Saifullah Bin Mohammad	Fisheries Officer - G41 Fisheries Biosecurity Centre, Kuantan, Pahang	saif_entra@yahoo.com
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3.	Noor Aishah Binti Wahab	Assistant Science Officer - C27 Fisheries Biosecurity Centre, Kuantan, Pahang	echa_30may@yahoo.com
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5.	Abu Yazidyusnisab Bin Muhammad	Assistant Research Officer - Q27 FRI Batu Maung, Pulau Pinang	yazid_remora@yahoo.com
6.	Tan Cheng Keng	Food Technologist - C41 National Public Health Laboratory, Ministry of Health Malaysia, Sg, Buloh, Selangor	cktan@moh.gov.my
7.	Tosiah Abdullah	Senior Food Technologist – C52 National Public Health Laboratory, Ministry of Health Malaysia, Sg, Buloh, Selangor	tosiahabdullah@moh.gov.my
8.	Eddy Afandi Bin Abdullah	Assistant Science Officer – C32 Veterinary Public Health Laboratory Department of Veterinary Services Bandar Baru Salak Tinggi Sepang, Selangor	d_bosn@yahoo.com
9.	Marni Sapar	Senior Research Officer – Q48 Veterinary Public Health Laboratory Department of Veterinary Services Bandar Baru Salak Tinggi Sepang, Selangor	marni@dus.gov.my msapar65@yahoo.co.uk

ANNEX II - COURSE PROGRAMME

TRAINING COURSE ON SAMPLE PREPARATION AND LC-MS/MS ANALYSIS FOR STILBENES AND NITROIMIDAZOLES

14 – 18 November 2011

Course Coordinators:	Hamdan Jaafar & Azahari Othman Ingrid Gevers
Resource persons	Paul Zoontjes Martien Essers

MONDAY 14 NOVEMBER

09.00 – 9.30	Gevers	Getting to know each other & learning objectives
09.30 – 11.15	Zoontjes/Essers	Aim of the training
11.15 – 12.00	Hazizi/Jaafar/Azahari	Opening of the course
12.00 – 13.30		Lunch
13.30 – 14.00	Zoontjes/Essers	Quality samples
14.00 – 15.15	Zoontjes/Essers	From theory to practice <ul style="list-style-type: none">• Stilbenes conform ISO 17025• Nitroimidazoles conform ISO 17025
15.15 – 17.30	Zoontjes/Essers	Practical training preparing standards <ul style="list-style-type: none">• Direct infusion LC-MSMS

TUESDAY 15 NOVEMBER

09.00 – 09.15	Participants/Gevers	Reflection on the previous day on lessons learned
09.15 – 12.30	Zoontjes/Essers	Practical laboratory training Stilbenes <ul style="list-style-type: none">• Calculation and preparing standard mix and calibration curve (MMS, MMRS and 1st line control)• Spiking samples, explanation, decision on spiking levels
14.00 – 17.30	Zoontjes/Essers	Continuation of practical laboratory training Stilbenes <ul style="list-style-type: none">• Sample preparation conform method

TRAINING COURSE ON SAMPLE PREPARATION AND LC-MS/MS ANALYSIS FOR STILBENES AND NITROIMIDAZOLES

14 – 18 November 2011

Course Coordinators:	Hamdan Jaafar & Azahari Othman Ingrid Gevers
Resource persons	Paul Zoontjes Martien Essers

WEDNESDAY 16 NOVEMBER

09.00 – 09.15	Participants/Gevers	Reflection on the previous day on lessons learned
09.15 – 12.30	Zoontjes/Essers	Practical laboratory training Nitroimidazoles <ul style="list-style-type: none">• Calculation and preparing standard mix and calibration curve (MMS, MMRS and 1st line control)• Spiking samples, explanation, decision on spiking levels
14.00 – 17.30	Zoontjes/Essers	Continuation of practical laboratory training <ul style="list-style-type: none">• Sample preparation conform method• Direct infusion Nitroimidazoles• Setting up a LC-MSMS method for measuring the samples• Measuring prepared samples

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THURSDAY 17 NOVEMBER

09.00 – 09.15	Participants/Gevers	Reflection on the previous day on lessons learned
09.15 – 12.30	Zoontjes/Essers	Practical laboratory training LC-MSMS <ul style="list-style-type: none">• Setting up LC methods for Stilbenes• Direct infusion Stilbenes
14.00 – 17.30	Zoontjes/Essers	Continuation of practical laboratory training LC-MSMS <ul style="list-style-type: none">• Setting up MS method, troubleshooting, optimisation• Measuring prepared samples

FRIDAY 18 NOVEMBER

09.00 – 11.15	Zoontjes/Essers	Discussion lab results of Thursday <ul style="list-style-type: none">• Interpretation of the measured data• Getting data from the LC-MSMS system• Result excel spreadsheet• 1st line control spreadsheet
12.15 – 14.30		Time for prayer & Lunch
14.30 – 15.15	Zoontjes/Essers	Evaluation of the training/Questions & Answers
15.15 – 15.30	Azahari/Zoontjes/ Essers	Closing ceremony and handing out certificates

ANNEX III - LEARNING OBJECTIVES

Sample preparation

- To know sample preparation/extraction of nitroimidazoles and stilbenes
- Sample extraction & analysis of stilbenes
- Sample extraction & analysis of nitroimidazoles
- Nitroimidazoles analysis: sample preparation
- Preparation of QC sample
- Sample preparation for nitroimidazoles and stilbenes (2)
- Stilbenes analysis: sample preparation
- Sample preparation for nitroimidazoles and stilbenes according to ISO 17025
- Stilbenes extraction and inst. optimization

Data analysis

- Nitroimidazoles analysis: instrumentation
- Stilbenes analysis: instrumentation
- To understand the software for LCMSMS (2)

Validation

- How to investigate PT outlier for c (proficiency testing)
- Method of stilbenes & nitroimidazoles
- Best technique to do LOD, LOQ
- Method validation: How to do it? Especially for non CRM sample
- Method validation
- Data interpretation especially sample with outlier result ($cc\alpha$ and $cc\beta$)
- Confirmation criteria of veterinary drug (acceptance criteria according to EU rules)
- The use of Measurement Uncertainty (MU) in the result
- Measurement Uncertainty

MS Analysis

- To familiarise with LC/MS-MS system
- To know the basic parts of LCMSMS
- Instrumentation part (LCMSMS). Optimising quantifier ion & qualifier ion
- LC-MS/MS analysis for nitroimidazoles

General

- To understand & know what stilbenes & nitroimidazoles are
- What is stilbenes & nitroimidazoles?

Other

- Guidance from RIKILT in finding blank samples for analysis of crystal violet in fish/prawn

ANNEX IV - GOOD AND BAD PRACTICES RELATED TO ISO17025

Implementation of ISO17025 in the laboratories of Kuantan Biosecurity Centre

The participants were asked to write down while working in the laboratories what practice in the lab where already implemented according to ISO17025 and what practices need to be improved.

Good points	To be improved
Everybody wears lab coat and gloves (6)	The solvent label is not complete. There is no preparation and expiry date and the name of the person that prepared it is missing (2)
The lab is clean	Lab does not provide waste bottle
The use of fume hood	Use nitrile instead of latex gloves
Equipment and material (beaker, spatula etc.) is easy to find in the lab and labelled (2)	Certain lab equipment is missing (Weighing cups for scale, small spatula etc.) (3)
	Not enough was bottle with ethanol spray available
	No log book is kept for the preparation of standards
	The lab did not provide any suitable shoes
	Materials

This report describes the content, the approach used and lessons learned during the implementation of a capacity development programme to build the analytical capacity of laboratory staff of the Department of Fisheries (DoF) and the Ministry of Health (MoH) who are directly involved in the analysis and detection of forbidden substances in fish and fisheries products. Two training courses were implemented in 2011 in the Bio Security Centre in Kuantan, Malaysia. The first training course on 'Marine lipophilic toxins using LC-MS/MS has been implemented in June and the second training course on on Stilbenes and Nitroimidazoles sample preparation and analysis with LC-MS/MS equipment was implemented in November, 2011. Through this knowledge transfer and laboratory enhancement the project contributed the laboratory's process towards getting accreditation under ISO 17025. The courses were implemented in partnership by Wageningen UR Institute of Food Safety (RIKILT) and Wageningen UR Centre for Development Innovation (CDI).

More information: www.cdi.wur.nl

