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Special report

Advancing the education in molecular diagnostics: The IFCC-Initiative "Clinical Molecular Biology Curriculum" (C-CMBC); A ten-year experience

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ABSTRACT

Molecular techniques are becoming commonplace in the diagnostic laboratory. Their applications influence all major phases of laboratory medicine including predisposition/genetic risk, primary diagnosis, therapy stratification and prognosis. Readily available laboratory hardware and wetware (i.e. consumables and reagents) foster rapid dissemination to countries that are just establishing molecular testing programs. Appropriate skill levels extending beyond the technical procedure are required for analytical and diagnostic proficiency that is mandatory in molecular genetic testing. An international committee (C-CMBC) of the International Federation for Clinical Chemistry (IFCC) was established to disseminate skills in molecular genetic testing in member countries embarking on the respective techniques.
We report the ten-year experience with different teaching and workshop formats for beginners in molecular diagnostics.

1. Introduction

Molecular diagnostics is becoming a standard analytical modality in many clinical diagnostic laboratories worldwide. To appreciate the increasing importance of molecular genetic testing, it is necessary to specify the areas of application in the field of laboratory medicine. Indeed, these range from the diagnosis of various infectious diseases, the quantitative assessment of viral load e.g. in chronic infectious diseases, advanced prenatal diagnosis, the detection of disease-causing mutations in rare disorders, the analysis of genetic susceptibility loci, molecular profiling and grading of malignant tumors or the detection of minimal residual disease in cancer. Also, molecular genetic testing is increasingly being used for therapy stratification using pharmacogenetics with an increasing number of gene loci associated with prodrug activation, drug metabolism and excretion. Finally, a few years ago molecular testing of tumors was based on the availability of solid tissues. In contrast, it is now widely accepted that nucleic acids circulate in the blood stream at any given time. This will allow performing non-invasive prenatal diagnostics as well as "liquid profiling" of the genetic and epigenetic make-up of tumors and their metastases.

To arrive at a result, molecular testing employs methods including nucleic acis preparation, amplification and various technologies for sequence-specific hybridization and detection. Many of the techniques, on which these sophisticated tests rely, can be traced back many years to when molecular biology methods were still new. Early on, the IFCC has provided recommendations on the professional laboratory setup and quality measures needed for molecular diagnostic applications [1]. Meanwhile, many of the basic methods are available as kit systems to accommodate robustness and to warrant compatibility with the hardware platforms. Still, the basic knowledge and understanding of the underlying general principles of molecular biology analytics is important for beginners for a number of reasons.

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For example, it has been shown in the context with external quality assurance (EQA) programs that laboratories acquainted with disposables and "black box" kit reagents perform significantly poorer in quality assurance tests than laboratories, which are used to adjust their methods by following primary literature and adjusting/optimizing their test protocols [2,3]. Consistently, European EQA schemes have shown that poor performers in EQA will improve significantly in subsequent EQAs after specific training in expert teaching laboratories [4,5].

Finally, the costs for molecular testing are hard to afford for many laboratories outside highly industrialized western Countries. Aside from the hardware investments, consumables are the major cost factor and limit the dissemination of molecular testing in the clinical diagnostic laboratory.

The dissemination of molecular techniques in laboratory medicine has been a goal of the International Federation of Clinical Chemistry (IFCC) for some years. Within the IFCC, the Education and Management Division (EMD) (http://www.ifcc.org/ifcc-education-division/) runs projects to provide IFCC member societies and their national healthcare communities with educational issues relevant to Clinical Chemistry and Laboratory Medicine. Within EMD, the Committee on Clinical Molecular Biology Curriculum (C-CMBC) has developed curricula and comprehensive hands-on laboratory training courses in basic diagnostic Molecular Biology techniques.

The dissemination of nucleic acids testing in the field of laboratory medicine, aims to improve skills and to tie IFCC members into molecular EQA systems in order to warrant a high level of analytical quality and diagnostic proficiency. Accordingly, the main course objectives are: i) to implement basic principles of molecular techniques or improve existing skills in a sustainable way rather than to exercise with prefabricated reagents and consumables, ii) to introduce laboratories to the principles of internal as well as external quality assessments (EQA), iii) to initiate networking amongst the participants of the C-CMBC courses assuming that they will benefit from each other's experiences using molecular genetic testing and iv) to improve the cooperation between the national societies and the IFCC in molecular diagnostic issues.

C-CMBC committee activities can be divided into two phases representing two different teaching concepts. During the first phase (2003–2008), the students travelled to a central teaching laboratory facility to participate in the C-CMBC courses. In the second phase (2008–2013), the C-CMBC committee travelled to the countries that had issued an invitation to the course program.

During the first phase, the annual courses took place in a state-of-the-art laboratory. Approximately twenty participants from various IFCC member countries convened with a team of 4–6 lab tutors and 5–7 lecturers. The course included both workshop seminars reviewing basic concepts of molecular biology and practical bench training in fundamental methodologies for DNA diagnostics. The practical parts were focused on DNA extraction, PCR and electrophoresis. At the end of each session results were discussed and further comments were driven from questions and exercises. During the theoretical part the scientific principles of molecular biology and their translational application in diagnostics were covered. Also, workflow aspects and infrastructural requirements for setting-up a new molecular diagnostics laboratory were discussed. In addition to the practical courses in the teaching laboratory, three seminar meetings were organized as excursions to Tunis, North Africa (2005 and 2007) as well as to the Fiji Islands (2008) utilizing facilities provided by the respective national hosts.

For monitoring sustainability, the C-CMBC made efforts to survey the success of the participants to implement newly acquired skills after returning home. Working and laboratory conditions are very heterogeneous in the participants' home countries and were generally very different from the technical capabilities in the C-CMBC teaching laboratory. Return from surveys and individual sustained contacts indicated that transfer of course experiences and skills to their home surroundings was difficult if not totally impossible for many students. Therefore, C-CMBC decided to support participants' activities locally and to tie in the national societies more closely.

Accordingly, a new C-CMBC course structure was developed around a "lab-in-a-suitcase" approach that was finally implemented in 2009.

This concept aimed at: i) organizing all courses through the national laboratory societies of the IFCC member countries, ii) leaving the selection of participants to the national societies to maximize dissemination, iii) establishing a beachhead in the hosting country through a native-tongue trainee prior to the course, iv) bringing the molecular techniques to the participants' countries and perform them in their every-day working environments, v) providing extensive documentation of all teaching materials for further dissemination, vi) allowing free-of-charge access to an international external quality assessment (EQA) program on molecular diagnostics (DNA-isolation, genotyping and DNA sequencing) for all registered participants and vii) establishing a junior member structure using the best students as continued liaisons between the IFCC and the hosting member countries.

This concept has several key elements described below in context with the course's organization and workflow.

2. Carry-on equipment

The lab-in-a-suitcase consists of a custom-made padded aluminum roller case (Fig. 1) and features a complete basic molecular biology laboratory equipment fit to develop and perform basic molecular genetic testing (Table 1). The suitcase was shipped by airfreight well in advance of the beginning of the course and shipped back to the course coordinator (Mannheim, Germany) after the end of the course activities.

Fig. 1. Lab-in-a-suitcase with contents prior to shipment to course venue.

Table 1
Contents carried along in the C-CMBC „Lab-in-a-suitcase“(weight 58 kg).

Assay step	Item	Category	Numbers included
NA preparation	Microcentrifuge	Instruments	1
	Nanofuge	Instruments	1
	Vortex mixer	Instruments	1
	Handheld pH meter	Instruments	2
NA amplification	Thermocycler	Instruments	1
	Pipettor (0.5–10 μl)	Instruments	5
	Pipettor (10–100 μl)	Instruments	5
	Pipettor (100–1000 μl)	Instruments	5
NA detection	Power supply	Instruments	1
	Electrophoresis chamber	Instruments	1
	UV-lamp	Instruments	1
	Camera	Instruments	1
	Incubation trays	Instruments	3
	Benchtop freezer	Instruments	1
	Goggles	Labware	2
Hard-/wetware	Glass- and plasticware	Labware	Various
	Racks	Labware	Various
	Pipette tips (yellow, blue)	Consumables	1000
	Disposable tubes	Consumables	
	Disposable pipettes	Consumables	
	General reagents	Consumables	
	Molecular biology reagents	Consumables	
	Oligonucleotides (lyophilized)	Consumables	
	Gloves	Consumables	

3. Precourse activities

As outlined in Fig. 2, a trainee travelled to the C-CMBC coordinator for a one week stay to be briefed on the course program. The trainee was appointed by the hosting society, usually had some prior experience in molecular techniques or was even well acquainted with molecular diagnostics. He/She ran through the one-week lab program once using the suitcase equipment and was familiarized with the tutorial videos as well as with all other documentation to be provided to the participants later. Where necessary, adjustments were made to the final course agendas on respective suggestions. For some courses, the tutorial videos were synchronized by the trainee in the native tongue. The trainee travelled back well in advance of the course and prepared the host-lab for the arrival of the suitcase and set up the course venue. At home, the trainee organized the support with perishable reagents that otherwise would have been difficult to transport or ship.

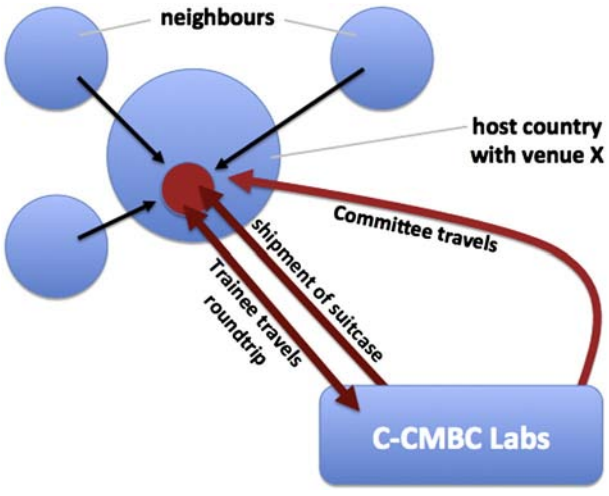

Fig. 2. Organizational procedure for the C-CMBC courses. Host country defines a venue, invites neighbors and sends a trainee. Trainee travels to the coordinating C-CMBC laboratory to get acquainted with the course before returning. C-CMBC ships suitcase to the venue. The trainee receives the lab-in-a-suitcase and sets it up prior to the committee traveling to the venue.

Each national hosting society had successfully advertised the course to sister societies in neighboring countries in order to disseminate the course program and contents beforehand and to invite participants from abroad. Since 2009, the C-CMBC was organized in five different countries: in Syria (2009), Uruguay (2010), Guatemala (2011), Malaysia (2012) and South Africa (2013). Taken together, these courses were attended by 81 participants from 9 countries (Fig. 3). To warrant a high teacher-to-student ratio, registrations were limited to a maximum of 16–20 students. Most participants are healthcare professionals, PhD and MSc holders, postgraduate students and lab technicians. The selection of course participants was mainly based on their potential to serve as future disseminators of the course contents (teach the teachers).

4. Course activities

The C-CMBC on-site course currently features intensive hands-on laboratory exercises in a standardized scheme of 6 days starting with the one-day precourse followed by the structured 5-day course program. Specifically, the pre-course lecture program addressed the basic principles of DNA chemistry, molecular hybridization principles and PCR methodology. During the next five days, nineteen 90 min lectures are held in total. In general, there are four types of lectures covering: i) molecular diagnostic techniques in the clinical lab with standard and real-time quantitative PCR and RT-qPCR, ii) detailed application of these techniques in various areas of molecular genetic testing like rare genetic disorders, pharmacogenetics, molecular virology and microbiology and molecular oncology, iii) molecular diagnostic strategies using epigenetics, DNA methylation, microRNAs, circulating tumor cells and companion diagnostics are highlighted and iv) state-of-the-art technologies and future prospects like multiplex testing or Next Generation Sequencing (NGS). To allow the students to prepare for the next day's schedule, a 30 min “preflight seminar” is scheduled at the end of the lab day one as a fixed part of the program.

Students are being encouraged to rate the day's program, and it is important to emphasize that the course program has continuously evolved based on the feed-backs of the respective previous years. For example, carrying out the one-day precourse introduction to molecular biology had been suggested in 2009 and was included into the program since. Also, the 2012 course in Malaysia had brought up the issues of interpretation of complex molecular data. Consequently, a bioinformatician was invited by the hosting society to give a lecture and a practical exercise in the 2013 course held in Capetown, South Africa.

5. Post-course activities

The course ends with a written multiple choice exam and distribution of a C-CMBC DVD containing the protocols, literature, tutorial videos and all lectures to the participants. IFCC certificates of attendance are issued to each participant. Evaluation of the exam and notes taken by the C-CMBC members leads to the identification of the best student by the end of the course. He/She is then invited to join the committee on next year's excursion as a guest of C-CMBC. In addition, these students are official junior C-CMBC members for a running period of three years by appointment of the IFCC. In their term, they keep contact with their fellow junior members, attend IFCC general conferences as invited C-CMBC members, establish/join young scientist networks and try to foster molecular diagnostics in their countries supported by their national societies. The expenses are generously being covered by the IFCC and supplemented by their societies in form of bursaries or additional support.

6. Results

During the course, many participants have gained their first hands-on experience on practical work in molecular diagnostics starting with preparation of buffers and reagents, inexpensive and robust DNA

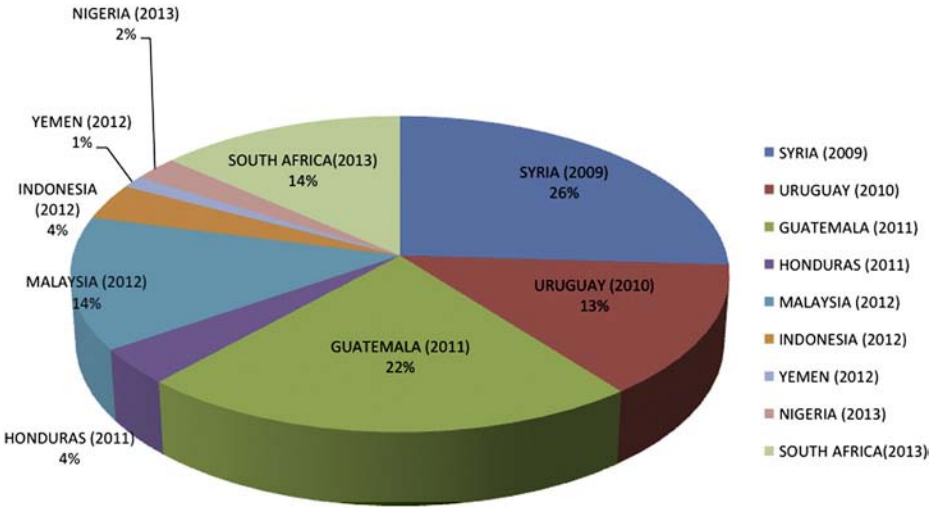

Fig. 3. The C-CMBC course during the last 5 years. Total number of participants: a) Syria, 2009: $n = 21$, b) Uruguay 2010: $n = 11$, c) Guatemala 2011: $n = 18$ from Guatemala, $n = 3$ from Honduras, d) Malaysia 2012: $n = 11$ from Malaysia, $n = 3$ from Indonesia and $n = 1$ from Yemen, e) South Africa 2013: $n = 11$ from South Africa and $n = 2$ from Nigeria.

isolation from whole blood, performance of a PCR reaction, ARMS-PCR, multiplex PCR, preparation of agarose gels, electrophoresis, dot blots and genotyping. We have noted that all participants needed training on genotyping issues, data interpretation and data navigation. Accordingly, much emphasis was put on *in-silico* work using public domain Internet sources for primer design, NCBI gene database work e.g. OMIM, assay design and prediction of PCR conditions, etc. These demonstrations and workshops were among the most appreciated ones of the entire program and have been given increasing room in the program as demanded.

One problem identified during the C-CMBC activities was that participants were insecure about the quality of their technical skills. The C-CMBC has placed much emphasis to strengthening the self confidence by addressing pre-analytical and quality aspects for the future clinical laboratory. Specifically, the participants were familiarized with international EQA programs. For example, all participants were registered to a three year – free of charge – access to the molecular diagnostics EQA schemes of the Reference Institute for Bioanalytics run by the German Society for Clinical Chemistry and Laboratory Medicine (www.DGKL-RfB.de). This allowed to establish a sustainable link to quality assessment in DNA isolation, genotyping of more than 57 different genetic risk loci and DNA sequencing in order to reference the analytical quality to the community of the labs participating in these schemes on a regular basis.

The activities of the C-CMBC committee have found the enthusiastic support of the EMD division chair, the IFCC president and the IFCC executive board and also have unanimously been appreciated by the local national societies hosting the courses.

Taken together, we believe that there are several strategic advantages of our current system to teach basic molecular techniques: i) as the trainee is a native speaker in the host country, communication is greatly facilitated and more substantial with the tutors, ii) more people from an area can be simultaneously trained allowing better contacts for subsequent local and regional collaborations, iii) by encouraging registrations from neighboring countries dissemination (teach the teachers) is thought to be more effective, iv) feedback allows to adapt the program to the needs of the attendants, v) granting easy and free-of-charge access to molecular EQA schemes should improve confidence in diagnostic results, vi) junior members that are supported by their national societies have a more sustained access to their peers allowing networking in the areas the course had been held, vii) finally, as a bottom-up approach, sustainable success may be more likely—indeed, suggestions of hosting a "second-stage" advanced course program have been suggested from participating countries. This feed-back encourages the C-CMBC to provide courses supporting National IFCC member Societies embarking on molecular diagnostics in the future.

Looking back at our ten year experience, we believe that this program represents an extraordinary support of young and enthusiastic laboratory scientists and will help young professionals to get started in analytical areas that can be foreseen to be of utmost importance to laboratory medicine. It should also provide between IFCC members in the area of molecular diagnostics and could be a role model for other innovative technologies to be spread in the professional community.

A full description of the course is provided on the IFCC webpage: http://www.ifcc.org/ifcc-education-division/emd-committees/c-cmbc/

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