Molecular genetic testing is a fast-growing diagnostic discipline. Only a few decades have passed since Kan and Dozy (1) described the first DNA test, which used a restriction enzyme digestion to test for sickle cell disease, and my group (2) reported the first use of linked DNA markers for first-trimester prenatal diagnosis of Duchenne muscular dystrophy. Since then, molecular genetic testing with direct or indirect mutation detection has been widely applied to confirm the clinical diagnosis for many monogenetic disorders and to determine carrier status. It also has been applied to predict presymptomatic cases and to predict, early in pregnancy, whether a fetus will be affected with a severe disorder.

Because the complete human genome (sequence) became available in 2003, molecular genetic testing will be applied more widely every day. Risk assessments for multifactorial disorders will gain more attention, as will predictions of genotypes that increase the possibility of adverse outcomes when certain drugs are used (pharmacogenomics).

Genetic information usually does not change throughout an individual’s life; therefore, the sequence of a gene need be determined only once in a person’s lifetime. For this reason, the outcome of a genetic test should be unambiguous, time resistant, and universal in nomenclature. The “DNA sequence of a gene” or “of a gene region” is often perceived as the ultimate result of a molecular genetic test. If we consider the nucleotide (A, T, G, or C) as the “SI unit” for DNA, then a nucleotide sequence comes as close to a gold standard as one can get in terms of a genetic test result. This is particularly true for monogenetic disorders, in which the gene sequence, disease-causing mutations, and a reference sequence usually are available in the public domain. The reference sequence can be used to highlight possible pitfalls in sequencing, such as a polymorphism under the primer sequence, the possible heterozygous deletion of an exon, or a whole gene that might be missed by sequencing. For the nomenclature, an internationally accepted recommendation for reporting sequence variations (mutations) is available at the Human Genome Variation Society web site http://www.hgvs.org (3). Additionally, molecular genetic technology has improved dramatically in recent years, and high-throughput DNA-sequencing instrumentation is widely used.

One fact that remains, however, is that molecular tests, including highly standardized methods such as DNA sequencing, are as prone to laboratory errors (technical or human) and/or interpretation errors as any other laboratory test. In this issue of Clinical Chemistry, the authors of two Europe-wide studies on external quality assessment (EQA) for DNA sequencing report their findings (4,5), and both show considerable variations in expected outcomes and some wrong genotyping results (diagnostic errors). In fact, in one of the studies (4), for 30% of the cases, no interpretation of the obtained result was provided. Although the two groups [the EQUALseq and the European Molecular Genetics Quality Network (EMQN)] come from opposite sides of laboratory medicine—the fields of clinical chemistry and clinical molecular genetics, respectively—they come to an overlapping conclusion: EQA is essential, even for a routine technology such as DNA sequencing. Moreover, the EQUALseq group declares that EQA should be mandatory, whereas EMQN proposes that EQA should be used as a benchmarking tool to rank individual laboratory performance.

These statements are understandable reactions to the problems encountered. However, in both clinical chemistry and clinical molecular genetics, much attention has already been given to the quality of laboratory results and the quality of the professionals, both in the United States and in Europe. Clinical chemists on both sides of the Atlantic regularly publish on these topics, as evidenced by the recent publication of “Essential Criteria for Quality Systems in Medical Laboratories” (6) and “Thoughts on Quality-Control Systems: A Laboratorian’s Perspective” (7). In addition, molecular geneticists have actively embraced quality assurance programs in molecular diagnostics. In the United Kingdom, EQA schemes for 3 disorders have been introduced since 1991 (8), and in Belgium, an EQA program for cystic fibrosis was organized in 1992 by Cuppens and Cassiman (9). Over the past few years, the European Union (EU) has funded several initiatives on quality assessment for molecular genetics, including the EQALseq and EMQN programs. For example, in January 2005, in a major effort to improve and harmonize the performance and quality of genetic testing in Europe, an EU-funded Network of Excellence in molecular genetic testing (www.EuroGentest.org) was started by Professor Jean-Jacques Cassiman (University of Leuven Center for Human Genetics, Leuven, Belgium). This network will bring together all stakeholders involved to enhance the awareness of quality in genetic testing. EuroGentest will assist laboratories in setting up a total quality management system, including EQA. The involvement of all laboratory staff is essential to successfully implement and maintain a quality system. The initial major investments by institutions of time and money will be worthwhile because the system will ultimately lead to improved traceability, accountability, and efficiency.

In several European countries, professional groups [e.g., Clinical Pathology Accreditation (UK), Ltd. (CPA); available at http://www.cpa-uk.co.uk] have endorsed the establishment of EQA systems, and Switzerland recently expressed its endorsement by passing legislation [Law on the Genetic Testing of Humans (GUMG); available at http://www.admin.ch/ch/d.ff/2004/5483.pdf]. In these countries, laboratories are complying with CPA guidelines or with international quality standards (ISO) and are seeking accreditation from their professional systems.
most early accredited laboratories, such as our laboratory in Leiden, voluntarily put quality systems in place. We began in 1994 by implementing ISO standard 17025. Although this is a guide for general testing laboratories, it helped us enormously in establishing the continual improvement cycle and standardization of processes. In 1998, our laboratory was the first molecular genetic testing laboratory in The Netherlands to be accredited by the Dutch Board of Accreditation (RvA). Later this year, we will switch to the new ISO standard 15189, which recently became available and has been adopted by international bodies for accreditation of medical testing laboratories. These guidelines also address particular requirements for quality and competence in the medical field.

In my opinion, laboratory directors must shoulder the responsibility to put in place rigorous total-quality systems in their laboratories, with or without the help of EuroGentest. As heads of clinical laboratories, they are solely responsible for the output of those laboratories. The fact that maintaining a quality system is not yet a common practice is solely because there is no formal obligation to do so. In most European countries, the governments have delegated this responsibility to the professionals or to national professional bodies.

In 1998, the In Vitro Diagnostic Directive (97/98/EC) was announced by the EU, and the directive became active in all member states of the EU as of December 8, 2003. This directive is applicable to all genetic tests marketed for diagnostic use, which should comply with CE Marking (Molecular Device Safety Service, In-Vitro Directives Division; available at http://mdss.com/IVDD/ivddtoc.htm). In-house tests, the so-called “home-brew kits”, do not need to comply with CE Marking, but these tests should be properly tested and validated in house before diagnostic use. This should also be a trigger for the management of a medical laboratory to put into place a rigorous quality system, because the requested validation should be demonstrable at all times.

In their respective reports, both EQUALseq (4) and EMQN (5) clearly demonstrate that there is room for improvement and that EQA is essential for assessing the quality of the molecular genetic tests being performed and the test reports being issued. However, laboratories need to embed total quality management systems in their basic operations. Mistakes will occur in any laboratory because of technical or human errors, errors in interpretation of test results, and other factors, but a chain of traceability should be in place to pinpoint occurrences, put corrective measures in place, and prevent recurrences.

References


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