SCIENCE AND SOCIETY

Quality control in molecular genetic testing

Elisabeth Dequeker, Simon Ramsden, Wayne W. Grody, Timothy T. Stenzel and David E. Barton

DNA-based testing for genetic diseases has developed from nothing into a principal part of laboratory medicine over the past 15 years. In the rush to bring these powerful new technologies into medical use, issues of quality have not always been given sufficient attention. Efforts are now being made to assess the quality of the output of genetic testing laboratories, and the results show that there is room for improvement.

Historically, molecular genetic testing developed in the mid-1980s in research laboratories where disease genes were being identified. Having identified a disease gene, it was a small step to proceed to test patients for mutations that might cause the disease. Although much diagnostic testing for rare disorders is still carried out in research laboratories, testing for more common monogenic genetic disorders, such as cystic fibrosis, the muscular dystrophies and fragile X syndrome, is now usually carried out in specialist diagnostic laboratories. Most of the main diagnostic molecular genetic laboratories are associated with clinical genetics departments and cytogenetics laboratories in medical genetics centres, and they still carry out research or are closely associated with research laboratories. However, as commercial testing kits become available, molecular genetic testing for inherited disorders is increasingly being carried out in general pathology laboratories and commercial centres.

Molecular genetic tests are extremely specific — they are usually designed to detect a single mutation in a specific gene. The results are for the most part qualitative (presence or absence of a mutation), although quantitative testing (number of repeat units, or number of copies of a gene sequence present) is also important. Testing laboratories use a wide variety of equipment and technologies, and new tests and technologies are continuously being introduced.

Data are sparse on the numbers of molecular genetic tests that are carried out, but the fact that more than 50,000 tests per year are done in the United Kingdom alone[1] indicates that hundreds of thousands of tests are probably carried out annually across the European Union (EU), and perhaps millions worldwide. The number of tests is likely to rise rapidly with the completion of the human genome sequence in the next few years. Furthermore, genetic tests will also be developed to determine susceptibility to

Box 1 | **Definitions**

Quality assurance (QA)
The policy, procedures and systematic actions established in a laboratory for the purpose of providing and maintaining a high degree of confidence in test integrity and accuracy, which includes sample input, handling, testing, result interpretation and report output.

External quality assessment (EQA)
Also known as proficiency testing. The process of determining whether a laboratory can produce results that are fit for the purpose intended. EQA schemes are run by organizations that are independent of the participating laboratories. EQA scheme participants are sent samples to test, with mock clinical referral reasons. Reports are sent back to a panel of experts for assessment (FIG. 1). Regular participation in EQA is a vital part of the QA process and is a requirement for accreditation.

Accreditation
A procedure by which an authoritative body gives formal recognition that a laboratory is competent to carry out specific tasks. Laboratories are assessed against a clearly defined set of standards that cover all aspects of the organization. Genetic testing laboratories might be accredited by specialist accreditation bodies (for example, the Clinical Laboratories Improvement Act, the College of American Pathologists and Clinical Pathology Accreditation in the United Kingdom), who develop their own standards. Alternatively, they might be accredited by national or international accreditation bodies — commercial or state sponsored — such as ILAC (International Laboratory Accreditation Cooperation) or EA (European Cooperation for Accreditation) against international standards, such as ISO 17025.

Reference materials
Reference materials (RMs) are materials (for example, DNA samples, cell lines and recombinant DNAs) that are known to have a defined property (such as a known genotype). RMs are used to calibrate and validate assays. Certified reference materials (CRMs) are RMs that have passed a process of examination, such as for their composition, content, stability and shelf life, and are certified to meet defined criteria for these properties.

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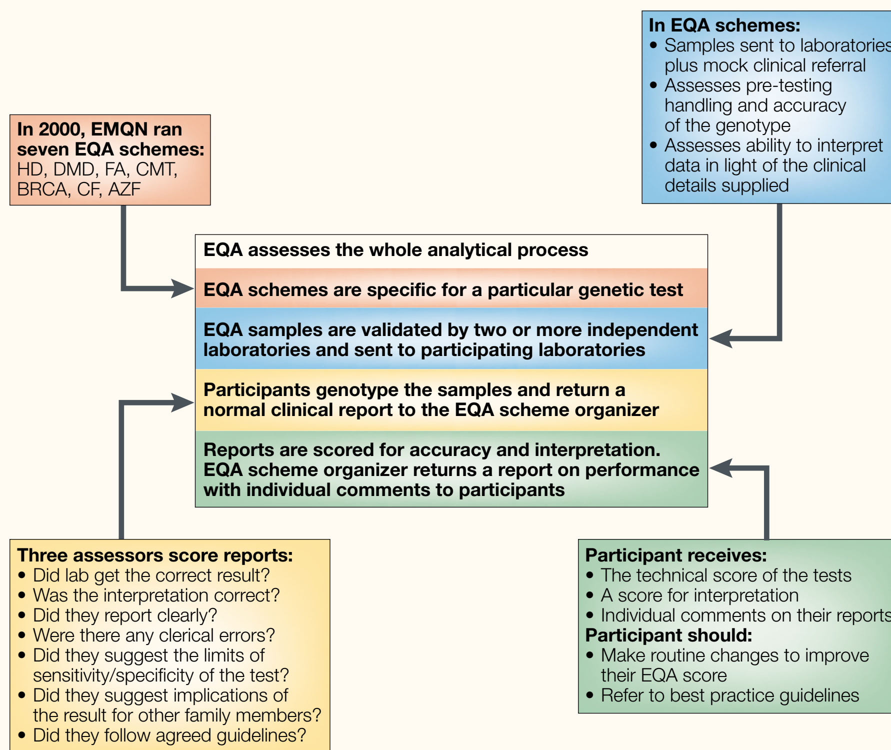

Figure 1 | **Anatomy of an EQA scheme.** The CF and AZF schemes were conducted in conjunction with European Concerted Action on Cystic Fibrosis and the European Academy for Andrology, respectively. AZF, azoospermia factor; BRCA, inherited breast cancer; CF, cystic fibrosis; CMT, Charcot–Marie–Tooth disease; DMD, Duchenne muscular dystrophy; EMQN, European Molecular Genetics Quality Network; EQA, external quality assessment; FA, Fanconi anaemia; HD, Huntington disease. (Adapted with permission from REF. 14 © (2000) Karger, Basel.)

common disorders, such as diabetes, heart disease, cancer and infectious disease, as well as for pharmacogenetic applications for predicting drug responses.

The regulatory machinery has had difficulty in keeping up with the pace of these advances. In many countries, anybody can set up a laboratory and start offering genetic tests. There is more regulation of tests offered as kits than of tests delivered as an in-house service, but only a small minority of tests is carried out using commercial kits. Most developed countries have laboratory accreditation systems (see below), but accreditation is usually voluntary and might not be available to specialist genetic testing laboratories.

Genetic tests are unlike other forms of laboratory diagnostic testing in several respects. First, because the genetic composition of an individual does not alter with time, patients are usually only tested once, and an incorrect result can stay with them for life. Second, the results of genetic tests can have profound implications for other family members. For example, prenatal tests that determine the genetic status of a fetus are often used to decide whether a pregnancy will be terminated. Third, the public and the medical profession have a high degree of confidence in the results of molecular genetic tests, perceiving them as 'state-of-the-art', and are thus perhaps less likely to question the validity of their results.

All of these factors contribute to the importance of maintaining the highest possible quality standards in molecular genetic testing laboratories, a requirement which is reinforced when one considers the growth in potential for testing in this discipline and the rate of development of new technologies. This article provides an overview of efforts to ensure quality in genetic test reporting in Europe and the United States.

Measuring quality

Quality (accuracy, reproducibility) and comparability of test results relies on laboratory accreditation, adherence to standard protocols, training of personnel, participation in external quality assessment schemes and the routine use of reference materials[2,3]. (See BOX 1 for definitions of relevant terms.) Although much attention has been focused on the appropriate qualification requirements for directors of genetic testing laboratories (for example, REF. 3) and on the steps required for test validation[4], it is only by studying the output of testing laboratories that a true measure of the reliability of test results can be obtained.

Many laboratories now participate in schemes in which identical samples are sent to all participants for testing, and the results compared to each other or to a recognized standard. This process is known in Europe as external quality assessment (EQA) and in the United States as proficiency testing. Three well-established schemes are described below. In the early days of EQA, schemes focused on accurate genotyping as the measure of quality. More recently, the scope of EQA schemes has broadened to include the entire analytical process, from sample receipt to final interpretative report. This approach, which is unusual in EQA, recognizes the special importance in genetic testing of interpreting each result in its particular context. A F508del heterozygous genotype means something quite different if the question is "Does this patient have cystic fibrosis (CF)?" rather than "Is this patient a CF carrier?" Grading or marking report content is much more subjective than scoring genotypes, and best practice guidelines (see below) provide an important framework for this process. The structure of the schemes run by the European Molecular Genetics Quality Network (EMQN) is typical of a modern EQA scheme in genetics (FIG. 1).

EQA schemes seek to educate — not to punish — the participants, so that they can achieve the highest possible quality standards. Participants whose returns do not meet the standards expected (especially in genotyping) are usually invited to explain to the scheme organizer(s) how the mistake occurred and what measures have been taken to prevent a recurrence. As these schemes mature, scheme organizers will inevitably have to develop procedures to deal with laboratories who consistently underperform, while maintaining a balance between their dual roles as educators and as protectors of patient safety.

Comparison of three EQA schemes

In this section, we describe EQA schemes based in the United Kingdom, Europe and the United States (TABLE 1), and then discuss the results obtained for CF testing in each scheme. There are, of course, well-established schemes in operation elsewhere, not covered in this article.

United Kingdom. The Clinical Molecular Genetics Society (CMGS), formed in the United Kingdom in 1988 to represent the new discipline of diagnostic molecular genetics, quickly recognized the need for external quality assessment to maintain

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Table 1 | **Comparison of the structure of UK, European and US EQA schemes**

	United Kingdom	Europe	United States
Number of participating countries	3	34	11
Average number of participating laboratories	28	160	45*
Average number of CF samples distributed each year	3	6	5
Data returned to the scheme organizer			
Genotype results in separate table	No	Yes	Yes
Written interpretative reports	Yes	Yes	Yes‡
Raw data (for example, copy of gels)	No	Yes	No
Details of mutation detection method(s) used	Yes	Yes	Yes
List of mutations that were tested	Yes	Yes	Yes
Returns accepted in several languages?	No	Yes	No
Individual evaluation reports	Yes	Yes	Yes
General evaluation reports	Yes	Yes	Yes
Certificate for successful participation	No	Yes	Yes

*This is the number of labaratories participating in the cystic fibrosis survey. See the text for the total number of laboratories participating in all the surveys. ‡Using ACMG/CAP report forms. ACMG, American College of Medical Genetics; CAP, College of American Pathologists; CF, cystic fibrosis; EQA, external quality assessment.

standards in this field. The CMGS set up quality assessment pilot schemes in 1991 and 1993 to include CF and Duchenne muscular dystrophy, respectively, the most commonly tested conditions at the time. From the outset, the scheme has required a full interpretation to accompany all genotype reports. Genotyping and interpretation are scored separately. After a transition period when the scheme was given seed funding by the UK Department of Health, it became, in 1999, a full member of the UK National External Quality Assessment Scheme (NEQAS) — an 'umbrella' organization that provides quality assessment schemes across the range of pathology disciplines. The UK NEQAS code of practice helps to ensure nationally agreed standards in quality assessment. The current scheme is self-financing and non-profit making and is administratively independent of the CMGS, although close links are maintained. The scheme distributes samples once a year for eight or nine diseases selected from a panel of 13. Diseases that have not given problems in one round of EQA are sometimes dropped for a year, so that new diseases can be included (TABLE 2). EQA schemes for haemophilia A and factor V Leiden are run under the auspices of the haematology branch of UK NEQAS, but these schemes only assess genotyping ability, not interpretation.

Europe. After a pilot study in 1994, as part of the EU-funded European Concerted Action on Cystic Fibrosis, annual EQA schemes for CF have been run since 1996 as part of the European Cystic Fibrosis Network. The set-up of the EQA was the same in each year's scheme: sets of six blinded DNA samples that carry common *CFTR* (cystic fibrosis transmembrane conductance regulator) mutations were sent to laboratories with the request to test for the presence of *CFTR* mutation(s) using their routine protocols. The samples were designated as "correctly genotyped" when the result was correct or when the laboratory did not test for the mutation in question and assigned it as wild-type. Most European laboratories that provide genetic services for CF take part and 104 laboratories have participated in all five EQA schemes between 1996 and 2000.

United States. Although there have been, and continue to be, several regional and informal interlaboratory specimen-exchange programmes, the predominant national proficiency testing programme for diagnostic molecular genetics in the United States is sponsored jointly by the College of American Pathologists (CAP) and the American College of Medical Genetics (ACMG). These molecular genetics programmes represent just a small part of CAP's overall quality assurance activities. At the present time, 203 laboratories subscribe to the molecular genetics proficiency surveys programme out of 878 US laboratories that undergo accreditation by the molecular pathology inspection checklist (encompassing DNA- and RNA-based tests applied to genetic diseases, cancer, infectious diseases, histocompatibility typing, forensic testing, marrow engraftment analysis and parentage analysis). These numbers might be contrasted with the total of 25,000 laboratories that participate in any of the 171 proficiency testing programmes offered, and 6,165 laboratories of all types that receive accreditation through the CAP programme. After several years of pilot development, the first formal CAP/ACMG molecular genetics proficiency survey was launched in 1995, with Duchenne muscular dystrophy, CF, fragile X syndrome and sickle-cell disease as the first tested disorders. The following year Huntington disease was added, followed in subsequent years by several more diseases, including factor V Leiden, myotonic dystrophy, Prader–Willi and Angelman syndromes, hereditary haemochromatosis and multiple endocrine neoplasia type 2. Some of these disease challenges have been offered twice a year and others once, each shipment containing either two or three unknown specimens to be tested.

Table 2 | **Diseases covered by the UK NEQAS for molecular genetics**

	1997	1998	1999	2000	2001
Cystic fibrosis	•	•	•	•	•
Duchenne and Becker muscular dystrophies	•	•	•	•	•
Familial adenomatous polyposis coli	•	•			
Familial breast and ovarian cancer			•	•	•
Fragile X syndrome	•	•	•	•	•
Friedreich ataxia					•
HMSN1/HNPP*	•	•		•	•
Huntington disease	•	•		•	
Mitochondrial diseases		•	•		
Myotonic dystrophy	•		•		•
Prader–Willi and Angelman syndromes	•	•		•	•
Spinal muscular atrophy	•		•		
Spino-cerebellar ataxias		•		•	

*HMSN1, hereditary and motor sensory neuropathy type 1; HNPP, hereditary neuropathy with liability to pressure palsies. UK NEQAS, UK National External Quality Assessment Scheme.

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Table 3 | **Genotypes of cystic fibrosis samples used in the three EQA schemes**

Genotype	European scheme	UK scheme	US scheme
Wild-type/wild-type		•	•
F508del/wild-type	•	•	•
I507del/wild-type	•		•
1717–1 G→A/wild-type	•		•
R553X/wild-type	•	•	
G551D/wild-type	•		
N1303K/wild-type	•		•
G542X/wild-type	•	•	•
W1282X/wild-type		•	•
621+1 G→T/wild-type		•	
R347H/wild-type		•	
F508del/F508del	•	•	•
F508del/N1303K	•		
F508del/G551D	•		•
F508del/621+1 G→T	•	•	•
F508del/R117H	•	•	•
F508del/G542X	•	•	
F508del/W1282X		•	
G551D/R553X	•		•
G542X/G542X			•
G85E/621+1 G→T			•

EQA, external quality assessment.

Results for cystic fibrosis. Three EQA schemes for CF have been organized for at least five years and can thus provide data for an overall comparison. These schemes include the UK NEQAS scheme (since 1995, on average 28 laboratories from the United Kingdom, Ireland and the Netherlands), the European scheme (since 1996, on average 160 laboratories from 34 countries) and the CAP scheme (since 1996, on average 45 laboratories mostly from the United States).

TABLE 3 summarizes the range of samples used in the different EQA schemes for CF. Although the design, the analysis and the data reporting of the schemes are different, the results of all three schemes illustrate that in successive QA schemes, the genotyping error rate declined. For the EU schemes between 1996 and 2000, errors have steadily decreased from 63 (3.8%) to 29 (1.3%). The proportion of laboratories making genotype errors decreased from 35 to 10% over the same period (FIG. 2). Similarly, the UK scheme observed a reduction from 0.9 to 0% of incorrect genotypes (15–0% of laboratories making errors) between 1995 and 1996, and there have been no genotyping errors since then. Genotyping error rates of 2–10% have been observed in EMQN schemes for other disorders (for example, REF. 5).

Although, for technical reasons, the figures might not be directly comparable to those above, the overall error rate for CF genotyping in the US scheme for the period 1995–2000 is estimated at 1.5%. Preliminary data indicate that ~50% of the errors observed are analytical/interpretative and 50% are due to clerical or sample mix-up problems. More data collection is needed to establish firm numbers for these categories. To this end, the scheme has recently initiated a programme to query the participating laboratories on the precise causes for each incorrect result. The two most significant analytical/interpretative errors seen so far include the failure to distinguish between two 3-nucleotide deletion mutations, F508del and I507del, and reporting a homozygous G542X sample as heterozygous, possibly because of the absence or misinterpretation of an internal wild-type control for this mutation.

The written reports, including the interpretation of the data (as normally sent to the referring medical doctor), were also evaluated in the most recent European and UK schemes, and showed considerably more variation between the participating laboratories. More than 30% of the laboratories that submitted reports to the European scheme in 1999 and/or 2000 made a mistake in at least one of the reports. The reports contained clerical errors (for example, typing errors, incorrect patient names or dates of birth), mistakes in risk calculations and incorrect interpretation of the (otherwise technically correct) results.

How can quality be improved?

The results from the EQA schemes presented above indicate a low but significant error rate in genotypes reported by molecular genetics diagnostic laboratories, and a higher rate of errors of interpretation. Although EQA provides an important measure of output quality, five principal elements of quality assurance must be in place for a laboratory to maintain a consistently high standard of performance[3,6].

Use of validated tests. As commercial kits are only available for a small number of tests, most laboratories use their own in-house (so-called 'home brew') test methodologies and reagents. Each laboratory must therefore show that its tests (both commercial and home brew) can consistently deliver the correct genotype under normal working conditions. The qualitative nature of most genetic tests (presence or absence of a mutation) means that the results are not susceptible to drift in the way that a quantitative test might be — the tests usually give the correct answer or fail completely. However, because most genetic tests are really two tests in one — looking for the mutant and wild-type alleles simultaneously — there is considerable scope for missing one allele if the methodology is suboptimal[7]. Collaborative studies to validate new methodologies (for example, REFS 8,9) before they are introduced into routine service are to be encouraged. Diagnostic scientists should make every effort to publish their validation work, as publications in this area are sparse, leading to unnecessary duplication of effort. New journals, such as *Genetic Testing* and *Genetics in Medicine*, as well as established titles, such as the *Journal*

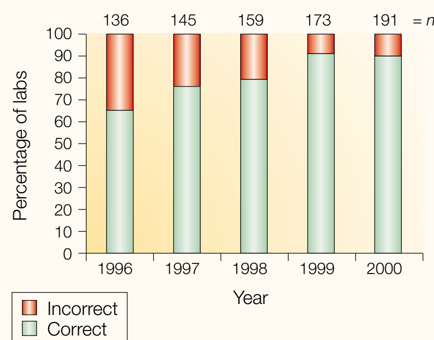

Figure 2 | **Incorrect genotypes in the European EQA scheme for cystic fibrosis.** The graph shows a progressive reduction in the percentage of laboratories making errors (red) during the five years of European external quality assessment (EQA) schemes for cystic fibrosis. Numbers across the top indicate the total number of participating laboratories (*n*).

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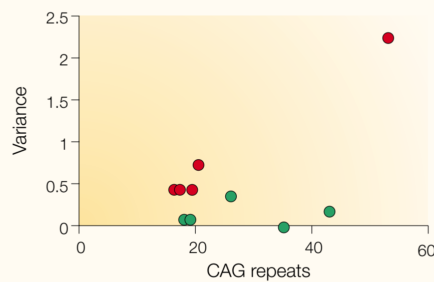

Figure 3 | **Effect of the introduction of a reference standard.** The plot illustrates the reduction in variance of Huntington disease repeat sizing in the UK National External Quality Assessment Scheme (UK NEQAS) when a 35-repeat standard was issued to all participants in 1998. Each point represents the variance of the results of all 11 participating laboratories, when testing for a Huntington disease gene allele of a specific size. The variance in repeat size measurement (green dots) using the reference standard is consistently lower than that seen before the standard was introduced (red dots), although some variance remains.

of Medical Genetics and *Clinical Chemistry* readily accept the results of relevant validation studies for publication.

Of course, developing a test that reliably produces the correct genotype is pointless if the consequences of having that genotype are uncertain. Tests must be validated clinically as well as technically. There are many pressures on laboratories to introduce new tests as quickly as possible, especially in the commercial sector. The most commercially attractive tests are those that are technically simple and for which there is a high volume of demand, such as testing for apolipoprotein E4 (*ApoE4*) (for Alzheimer disease susceptibility), factor V Leiden and hereditary haemochromatosis. Unfortunately, these are often the cases in which it is most difficult to draw definitive conclusions about the consequences for the patient of a particular test result. A system is required by which regulatory bodies at national and international level can cooperate to approve such tests in a timely and efficient manner. Furthermore, research should not stop when a test is first introduced — it is essential that the process of defining the phenotypic consequences of a particular genotype (or combination of genotypes) continues indefinitely. Where applicable, licence agreements that cover genetic tests should mandate (or at least not inhibit) the gathering and publication of such data.

Reference materials. To verify that an assay is giving the correct result, it is essential to run positive-control samples for the expected genotypes with each batch of test samples. But where do such positive controls come from? Reference samples are normally supplied with commercial diagnostic kits, whereas most diagnostic laboratories develop their own positive controls, verified by a second assay method, or obtain control samples from a research laboratory or another diagnostic laboratory. However, a recent EU Directive will outlaw such transfers unless the materials have passed the necessary tests to carry the 'CE' mark — the EU symbol for conformity to a defined standard[10]. There is a clear requirement, therefore, for independently validated reference samples, and moves are now being made to generate such materials.

The European Commission has recently funded a project to develop a range of certified reference materials for molecular genetic testing, and to develop general guidelines for the production of such materials. The Certified Reference Materials for genetics project, which will start in October 2001, involves ten centres from seven European countries. Such materials will be particularly useful in calibrating assays for disorders caused by the expansion of trinucleotide repeats[11]. Sizing of PCR products that consist entirely of CAG repeats against commercial marker ladders gives inconsistent and inaccurate results[12], so products must be sized against a series of known standards. In Huntington disease, there is no gap between the upper end of the normal range of repeat sizes and the lower end of pathogenic sizes (in fact a small overlap exists), so precision is essential. A marked reduction in variance of Huntington-disease-repeat sizing was achieved by the distribution of a sequenced 35-repeat standard (representing the interface between the normal and pathogenic ranges) to participants of the UK NEQAS scheme in 1998 (FIG. 3).

Staff training. The mix of clinicians, scientists and technicians in genetics laboratories varies considerably between different countries, but whatever the staff structure, qualifications must meet required standards and training must be given high priority. In hard-pressed laboratories, with burgeoning sample volumes, training can be forced to take a back seat, but it is incumbent on laboratory directors to insist that funding and resources are made available to ensure staff are adequately trained to carry out the work assigned to them, and that this training continues throughout their careers. The recent introduction in the United Kingdom of a register and legal protection for the title Clinical Scientist is a positive move in this direction (see link to The Council for Professions Supplementary to Medicine).

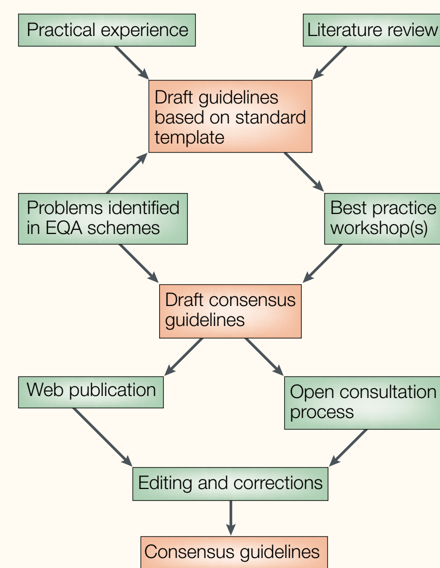

Figure 4 | **Model pathway to CMGS/EMQN best practice guidelines.** The flow diagram illustrates the flow of information through the guidelines development process. Procedures must be in place to update and correct the guidelines once published. CMGS, Clinical Molecular Genetics Society; EMQN, European Molecular Genetics Quality Network; EQA, external quality assessment.

Best practice guidelines. Early experience in EQA schemes for molecular genetics showed that a wide range of approaches, techniques and nomenclatures were in use in participating laboratories. In the United Kingdom, the CMGS addressed (and continues to address) this problem by organizing best practice workshops for each disease, for the purpose of sharing experience, identifying problems and developing consensus best practice guidelines. The EMQN has adapted this approach[13], and now organizes consensus workshops for laboratories across Europe (FIG. 4). Other organizations have taken similar approaches[14] or have developed complementary sets of guidelines, usually generic rather than disease specific. The American College of Medical Genetics has developed "Standards and Guidelines for Clinical Genetics Laboratories"[15], covering all aspects of the work of a molecular genetics diagnostic laboratory, and NCCLS (formerly the National Center for Clinical Laboratory Standards) has produced specific guidelines on "Molecular Diagnostic Methods for Genetic Diseases"[16]. Best practice guidelines are linked into the EQA process (and hence into accreditation) because the guidelines are used as a basis for marking returns in EQA schemes, and solutions to problems identified in EQA are incorporated into the guidelines (FIG. 4).

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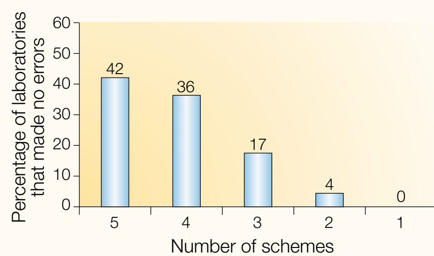

Figure 5 | **Distribution of errors in laboratories from 1996 to 2000.** A total of 104 laboratories participated in all five European external quality assessment (EQA) schemes for cystic fibrosis. The chart shows how the number of schemes completed without error is distributed among the laboratories, and shows that 58% of laboratories produced errors in at least one scheme. Therefore errors are not limited to only a small percentage of laboratories. Each scheme included six samples (12 alleles) to genotype.

Accreditation. The key to ensuring that all diagnostic laboratories implement the types of quality assurance programme described above is laboratory accreditation. Accreditation obliges laboratories to meet a pre-defined set of standards on laboratory management, facilities, staff qualifications and training, as well as EQA participation and performance. The process itself involves submission of documents for inspection and visits by a team of quality experts to the laboratory. Accreditation can be highly specific, certifying that a laboratory is competent to carry out a specific test, or it can be very broad, covering analytical testing in general. There is an important link between accreditation and EQA. Accreditation standards dictate that all laboratories must participate in appropriate EQA schemes, and must show satisfactory performance in such schemes. Therefore, laboratories that do not do well (or do not participate) in EQA risk losing their accredited status. Without this link, there is no incentive for laboratories to participate in EQA, and poor laboratories might simply shy away from such assessments. Once accreditation systems are in place, the next step is to make accreditation mandatory for all diagnostic laboratories. This is already the case in some countries such as the United States[17], but these are the exceptions. The United Kingdom has no specific restrictions on laboratories that offer diagnostic testing, and there is no common EU policy in this area.

Conclusion

The organization of EQA schemes has proven to be useful for several reasons. First, genotyping errors in the scheme allow the participating laboratories to identify shortcomings in their internal quality control and to detect the source of their mistake(s). Indeed, most laboratories that made a genotyping mistake in the European scheme could identify the source of error, and subsequently adapt their analytical protocol to prevent future mistakes.

Second, genotyping mistakes that are made by several laboratories might pinpoint shortcomings in the methods applied. As an example, both the European and the CAP schemes for CF noticed that several laboratories erroneously genotyped DNA samples that carry mutations F508del and/or I507del (both three-nucleotide deletions) in the *CFTR* gene, which cannot be distinguished by all routine diagnostic methods[18]. This problem has not been seen in the UK scheme because no DNA sample with mutation I507del has yet been distributed. Overall, most of the technical genotyping mistakes in EQA schemes could be explained because laboratories did not know the limits of the applied mutation detection technique[18,19]. It should, however, be noted that not all genotyping mistakes are due to technical errors or misinterpretation of technically correct data: clerical mistakes and sample mix-up were reported in all three EQA for CF.

Third, the data from more than 100 laboratories that participated in all five European EQA schemes for CF allow an overall analysis of the progress of genetic testing quality for CF. Published data from a three-year study period show that less than half of the laboratories that participated in three consecutive annual trials made no mistake in any of the three EQA[19]. FIG. 5 shows that the results, now extended to a five-year period, still point to the same conclusion. This observation challenges the idea that only a small number of laboratories are responsible for repeated mistakes in EQA schemes.

Fourth, EQA schemes might also be a good tool to follow up on the development of testing strategies used by diagnostic laboratories. Towards this aim, the European EQA scheme has, since 1996, included a request for information on the CF mutations tested and the technologies that have been applied to this. It seems that the selection of mutations to test, as well as the technology applied, is largely laboratory dependent but is also influenced by the geographical location of the laboratory (as an obvious consequence of the frequency variation of specific *CFTR* mutations between populations in Europe). Since 1996, there has been a clear tendency in molecular diagnostic laboratories to shift from home brew techniques and protocols to commercial kits, especially as the primary testing technique. As a consequence, the average number of mutations being tested has also increased[19]. For the United States, the diversity in size and nature of the *CFTR* mutation panels offered by 43 routine diagnostic laboratories was evaluated in 1997, and also showed significant variation between laboratories[20]. There was a wide range in the number of mutations tested, from just 1 to 70. Recently, specific recommendations for population-based CF carrier screening in the United States have been published by the ACMG[21], who proposed a minimal 25-mutation panel.

Although the results from individual laboratories in EQA schemes are confidential, the collation, comparison and publication of anonymous aggregate data from EQA schemes will be an important part of the continuing drive to improve quality.

It is clear that, although important progress has been made in the introduction and implementation of quality assurance measures in molecular genetic testing laboratories, much work remains to be done. This article has focused on the United Kingdom, the European Union and the United States as examples. Many other countries have quality systems in place, but vastly more have none. These countries can benefit from the experience of the others, through the work of international organizations, such as the Human Genome Organization (HUGO), the Human Genetics Societies (such as the American Society of Human Genetics and the European Society of Human Genetics) and the Organization for Economic Cooperation and Development (OECD).

The OECD has recently embarked on a worldwide study of quality standards in molecular genetic testing, modelled on a similar study of US laboratories[2]. The European Union funded the Concerted Action on Genetics Services in Europe (CAGSE), which surveyed the current services in 30 European countries and proposed standards for the organization of genetics services[1]. The results of such surveys, and the policy documents based on their results, can be a powerful influence on funding agencies and legislators in less-developed countries, as they establish standards for all to aim at. There is, at present, much overlap in the development work that is carried out by diverse organizations involved in EQA and in the development of guidelines. There are now excellent opportunities for co-operation between these organizations to share experience and expertise, and to harmonize methodology and standards.

Although the subject of this article has been the quality of the output of molecular genetic testing laboratories, there is clearly a requirement to ensure that the patient, and not just the physician, receives a correctly interpreted genotype. As the number and

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complexity of available tests increase inexorably, educating current and future medical staff in the interpretation of test results and how to transmit them to patients will be an important challenge. It is hoped that the lessons learned in developing quality systems in the laboratory setting will be of some help.

Elisabeth Dequeker is at the European Thematic Network for Cystic Fibrosis and European Molecular Genetics Quality Network (EMQN), Department for Human Genetics, University of Leuven, Campus Gasthuisberg, Herestraat 49, B-3000 Leuven, Belgium.

Simon Ramsden is at UK NEQAS for Molecular Genetics, Regional Molecular Genetics Laboratory, St Mary's Hospital, Hathersage Road, Manchester M13 0JH, UK.

Wayne W. Grody is at Divisions of Medical Genetics and Molecular Pathology, Departments of Pathology and Laboratory Medicine and Pediatrics, UCLA School of Medicine, Los Angeles, California 90095-1732, USA.

Timothy T. Stenzel is at Box 3712, Department of Pathology, Duke University Medical Center, Durham, North Carolina 27710, USA.

David E. Barton is at EMQN and UK NEQAS for Molecular Genetics, National Centre for Medical Genetics and Department of Medical Genetics, University College Dublin, Our Lady's Hospital for Sick Children, Crumlin, Dublin 12, Ireland. Correspondence to D.E.B. e-mail: david.barton@olhsc.ie

Links

DATABASE LINKS Cystic fibrosis | Haemophilia A | Factor V Leiden | *CFTR* | Duchenne muscular dystrophy | Fragile X syndrome | Sickle-cell disease | Huntington disease | Myotonic dystrophy | Prader–Willi | Angelman | Hereditary haemochromatosis | Multiple endocrine neoplasia type 2 | *ApoE4*

FURTHER INFORMATION European Molecular Genetics Quality Network | Clinical Molecular Genetics Society | UK National External Quality Assessment Scheme | European Cystic Fibrosis Network | College of American Pathologists | American College of Medical Genetics | The Council for Professions Supplementary to Medicine | National Center for Clinical Laboratory Standards | Human Genome Organization | American Society of Human Genetics | European Society of Human Genetics | Organization for Economic Cooperation and Development

1. Harris, R. & Reid, M. Medical genetic services in 31 countries: an overview. *Eur. J. Hum. Genet.* **5**, S3–S21 (1997).
2. McGovern, M., Benach, M. O., Wallenstein, S., Desnick, R. J. & Keenlyside, R. Quality assurance in molecular genetic testing laboratories. *J. Am. Med. Assoc.* **281**, 835–840 (1999).
3. Holtzman, N. A. & Watson, M. S. (eds) *Task Force on Genetic Testing: Promoting Safe and Effective Genetic Testing in the United States* (Johns Hopkins Univ. Press, Baltimore, Maryland, 1997).
4. Prence, E. M. A practical guide to the validation of genetic tests. *Genetic Testing* **3**, 201–206 (1999).
5. Losekoot, M., Bakker, B., Laccone, F., Stenhouse, S. & Elles, R. A. European pilot quality assessment scheme for molecular diagnosis of Huntington's disease. *Eur. J. Hum. Genet.* **7**, 217–222 (1999).
6. Holtzman, N. A. & Shapiro, D. Genetic testing and public policy. *Br. Med. J.* **14**, 852–856 (1998).
7. Jeffrey, G. P., Chakrabarti, S., Hegele, R. A. & Adams, P. C. Polymorphism in intron 4 of HFE may cause overestimation of C282Y homozygote prevalence in haemochromatosis. *Nature Genet.* **22**, 325–326 (1999).
8. Kubota, T. *et al.* Validation studies of SNRPN methylation as a diagnostic test for Prader–Willi syndrome. *Am. J. Med. Genet.* **66**, 77–80 (1996).
9. Rowland, J. S., Barton, D. E., Taylor, G. R. & the UK Clinical Molecular Genetics Society HMSN Project Group. A comparison of methods for gene dosage analysis in HMSN type 1. *J. Med. Genet.* **38**, 90–95 (2001).
10. Directive 98/79/EC of the European Parliament and of the Council of 27 October 1998 on *in vitro* diagnostic medical devices. *Official J.* L **331**, 1–37 (1998).
11. Cummings, C. J. & Zoghbi, H. Y. Fourteen and counting: unravelling trinucleotide repeat diseases. *Hum. Mol. Genet.* **9**, 909–916 (2000).
12. Bruland, O. *et al.* Accurate determination of the number of CAG repeats in the Huntington disease gene using a sequence-specific internal DNA standard. *Clin. Genet.* **55**, 198–202 (1999).
13. Patton, S., Barton, D. & Elles, R. Retaining the confidence of the public in molecular genetic testing — quality assurance. *Commun. Genet.* **3**, 164–169 (2000).
14. Dequeker, E. *et al.* Recommendations for quality improvement in genetic testing for cystic fibrosis. European Concerted Action on Cystic Fibrosis. *Eur. J. Hum. Genet.* **8**, S2–S24 (2000).
15. Watson, M. S. (ed.) *Standards and Guidelines: Clinical Genetics Laboratories* 2nd edn (American College of Medical Genetics, 1999). Available at http://www.faseborg/genetics/acmg/stds/copyrite.htm
16. NCCLS. *Molecular Diagnostic Methods for Genetic Diseases: Approved Guidelines.* NCCLS Document MM1-A (NCCLS, Wayne, Pennsylvania, 2000).
17. Schwartz, M. K. Genetic testing and the clinical laboratory improvement amendments of 1988: present and future. *Clin. Chem.* **45**, 739–745 (1999).
18. Dequeker, E. & Cassiman, J. J. Evaluation of *CFTR* gene mutation testing methods in 136 diagnostic laboratories: report of a large European external quality assessment. *Eur. J. Hum. Genet.* **6**, 165–175 (1998).
19. Dequeker, E. & Cassiman, J. J. Genetic testing and quality control in diagnostic laboratories. *Nature Genet*. **25**, 259–260 (2000).
20. Grody, W. W., Desnick, R. J., Carpenter, N. J. & Noll, W. W. Diversity of cystic fibrosis mutation — screening practices. *Am. J. Hum. Genet.* **62**, 1252–1254 (1998).
21. Grody, W. W. *et al.* Laboratory standards and guidelines for population-based cystic fibrosis carrier screening. *Genet. Med.* **3**, 149–154 (2001).

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TIMELINE

The consequences of political dictatorship for Russian science

Valery N. Soyfer

The Soviet communist regime had devastating consequences on the state of Russian twentieth century science. Country Communist leaders promoted Trofim Lysenko — an agronomist and keen supporter of the inheritance of acquired characters — and the Soviet government imposed a complete ban on the practice and teaching of genetics, which it condemned as a "bourgeois perversion". Russian science, which had previously flourished, rapidly declined, and many valuable scientific discoveries made by leading Russian geneticists were forgotten.

"We cannot wait for Nature's good graces — to take them from her is our goal."[1]

The Communist state that replaced the Russian Empire in 1917 was based on Marx and Engels' thesis that it was possible to quickly and successfully alter economic relationships and even change the nature of all organisms, including human beings. The belief that Nature is malleable and can be heritably altered by the environment contradicted the laws of genetics, which are incompatible with the theory of acquired characteristics proposed by Lamarck. The 1920s saw vicious debates between geneticists and Lamarkists. Although, initially, many Soviet geneticists argued that genetics was entirely compatible with Communist ideology[2], the Communists soon shifted towards supporting the inheritance of acquired characteristics. By the early 1920s, geneticists were being publicly attacked by Lamarkists[3], and by the end of the decade many of them were being condemned as "bourgeois scientists". It was also at that time that Trofim Denisovich Lysenko, an agronomist by training who sided with the Lamarkists, made his first claims of being able to create new wheat varieties by varying environmental conditions, so providing a much needed improvement in grain harvest in the USSR. Communist Party leaders wholeheartedly embraced Lysenko's promising claims. By 1934, Lysenko was proclaiming that genetics was a hostile science for those who supported communist ideology — a view that culminated in a ban on genetics[4]. Political dictatorship in science in the USSR led to the

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