Clinical Laboratory Reports in Molecular Pathology

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● ***Context.*—Molecular pathology is a rapidly growing area of laboratory medicine in which DNA and RNA are analyzed. The recent introduction of array technology has added another layer of complexity involving massive parallel analysis of multiple genes, transcripts, or proteins.**

***Objective.*—As molecular technologies are increasingly implemented in clinical settings, it is important to bring uniformity to the way that test results are reported.**

***Data Sources.*—The College of American Pathologists Molecular Pathology Resource Committee members summarize elements that are already common to virtually all molecular pathology reports, as set forth in the College of American Pathologists checklists used in the laboratory accreditation process. Consensus recommendations are proposed to improve report format and content, and areas of controversy are discussed. Resources are cited that promote use of proper gene nomenclature and that describe methods for reporting mutations, translocations, microsatellite instability, and other genetic alterations related to inherited disease, cancer, identity testing, microbiology, and pharmacogenetics.**

***Conclusions.*—These resources and recommendations provide a framework for composing patient reports to convey molecular test results and their clinical significance to members of the health care team.**

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The physician who orders a molecular test expects the laboratory report to state what test was done and the result that was obtained. Many clinicians find it helpful if the report further describes what the result means for their patient based on analytic and clinical performance characteristics of the test in correlation with the clinical setting in which the test was done.[1–3] This is true regardless of whether the report is a stand-alone molecular pathology report or part of a larger surgical pathology or hematopathology report in which molecular data is integrated with histologic examination and other tests done on the same specimen.[4,5]

The College of American Pathologists (CAP) has developed recommendations for composing laboratory reports. Many of these recommendations are outlined in the checklists that are used by inspectors to evaluate compliance as part of the laboratory accreditation process. The Laboratory General checklist summarizes guidelines that are common to every laboratory report, whereas the subspecialty checklists such as those for molecular pathology, microbiology, cytogenetics, anatomic pathology, and human leukocyte antigen (HLA) contain additional elements specific to the molecular testing done in specialized areas of the clinical laboratory. These checklists may be downloaded from the CAP Web site (www.cap.org). Some report components are federal requirements for tests performed in the United States, whereas others are recommendations or requirements for accreditation by the CAP. In this article, current reporting guidelines are summarized and additional recommendations are made to improve content and format of clinical laboratory reports.

CONTENT OF MOLECULAR PATHOLOGY REPORTS

The recommended elements of a molecular pathology report are shown in Table 1. The report should include fundamental information about the patient, the specimen, the ordering physician, and the laboratory where testing was done. Reports from an outside laboratory must be made available to the clinician, and they may also be transcribed into the local information system either verbatim or as summarized by a laboratory professional who is knowledgeable in the field.

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Table 1. Recommended Molecular Pathology Report Content

Laboratory/patient/sample identifiers
- *Name and address of reporting laboratory (optional: phone, FAX, e-mail, Web site)
- *Patient's name (first and last with middle initial or middle name)
- *ID No. and/or date of birth
- *Date of specimen collection (and time, if appropriate)
- *Date of receipt or accession in laboratory, with accession number
- *Specimen source (even if only 1 sample type is accepted) and how tissue was received (fresh is assumed unless designated as frozen, paraffin-embedded, etc)
- *Ordering physician

Results
- *List results by test name; use standardized gene nomenclature and standard units of measure
- *Reference range; or normal versus abnormal

Interpretation
- Analytic and clinical interpretation of results: Analytic interpretation involves synthesizing raw data to produce a reportable result. Clinical interpretation involves synthesizing analytic and clinical information to describe what the result means for the patient

Comments
- Significance of the result in general or in relation to this patient
- Correlate with prior test results
- Recommend additional measures (eg, further testing, genetic counseling)
- Condition of specimen that may limit adequacy of testing (eg, sample received thawed, partially degraded DNA)
- Pertinent assay performance characteristics or interfering substances
- Residual risk of disease (or carrier status) by Bayesian analysis
- Control test results, if unusual or especially pertinent
- Cite peer-reviewed medical literature or reliable Web sites on the assay and its clinical utility (eg, educational materials on genetests.org)
- Document intradepartmental consultation
- Document to whom preliminary results, verbal results, or critical values were reported and when
- Incorporate information specifically requested on the requisition (eg, ethnicity)
- Answer specific questions posed by the requesting clinician (eg, rule out CML)
- Reason specimen rejected or not processed to completion
- Disposition of residual sample (eg, sample repository)
- Chain of custody documentation, if needed
- *If the report is an amended or addendum report, describe the changes or updates
- *Describe discrepancies between preliminary and final reports
- *Name of testing laboratory, if transmitting or summarizing a referral laboratory's results

Procedure
- *Type of procedure (eg, Southern blot, PCR, RT-PCR of RNA, Q-PCR, in situ hybridization, gene dosage array, RNA expression array, protein expression array, sequencing, protein truncation test)
- *Defined target (ie, name of analyte tested such as gene, locus, or genetic defect; use HUGO-approved gene nomenclature)
- Pertinent details of procedure, for example analyte-specific reagent or kit version and manufacturer, instrument type
- Disclaimer on non-FDA approved tests in which a commercial analyte-specific reagent was used
- Signature and printed name of reporting physician, for any test having a physician interpretation
- Signature of lab director or designee when interpretation is performed (Reports may be signed electronically.)
- *Date of report (and time, if appropriate)

Demographic information
- Accession number, and specimen number from referring laboratory
- Genetic counselor, when appropriate
- Clinic/inpatient location; or name/address/phone of outside facility
- Indications for testing (reason for referral)
- Pertinent clinical history (clinical situation, ethnicity/race, pedigree diagram and/or family history, previous molecular/genetic studies, and other relevant clinicopathologic findings)

Billing information
- *ICD-9* code (clinical indication for test)
- *CPT* codes (laboratory procedures performed)

* At minimum, all reports should contain the elements designated by an asterisk, as delineated in College of American Pathologists checklists. The sequence of elements need not follow the order shown in this table. ID indicates identification; CML, chronic myelogenous leukemia; PCR, polymerase chain reaction; RT-PCR, reverse transcription PCR; Q-PCR, quantitative PCR; HUGO, Human Genome Organisation; FDA, US Food and Drug Administration; *ICD-9, International Classification of Diseases, Ninth Revision*; and *CPT, Current Procedural Terminology.*

Methodology

It is important to specify the gene or locus tested and the method used to analyze it, particularly because there are so many different methods that can be used, and each method has different performance characteristics. One should describe which commercial kit or analyte-specific reagent was used or specify the identification number of the probe or primer sequence if it is listed in the National Center for Biotechnology Information (NCBI) probe database (www.ncbi.nlm.nih.gov/projects/genome/probe/doc/Overview.shtml). For microarray testing in which it is impractical to list dozens to thousands of analytes, one should cite a publication or Web site detailing the pertinent methodologic parameters. It is also useful to cite a literature reference or Web site that provides analytic and clinical performance data relevant to the test procedure.

When serial testing is done, for example to measure viral load or to monitor tumor burden, it is useful to report which assay was used so that the same assay can be applied at future timepoints to more accurately track the disease. Regardless of which technology is used, human immunodeficiency virus, hepatitis C virus, and hepatitis B virus viral load testing benefit from international standards that are available and agreed on as a reference for quantitative measurement. The specimen types and units of measurement also tend to be universally consistent. For quantitative assays that are not calibrated to an internationally agreed-on standard, a consequence may be the need for a given patient to be serially monitored by the same laboratory. Clinicians might benefit from a report that graphs or lists results over time.

To help ensure quality, laboratories in the United States are required by law to validate assays and to participate in proficiency testing at least semiannually. It can be assumed that every Clinical Laboratory Improvement Amendments of 1988 (CLIA)–approved or CAP-accredited testing laboratory meets these requirements, and therefore it is not necessary to include a statement to this effect in each patient report. However, for any test using an analyte-specific reagent as defined by the US Food and Drug Administration (FDA), the US government requires a disclaimer on each report, as explained at www.advamed.org/publicdocs/reagents.htm. Any laboratory using an FDA-approved kit in unmodified form should so state in the patient report because this information conveys precisely which method was used. A list of FDA-approved molecular test kits is found on the Association for Molecular Pathology Web site (www.amp.org).

Abbreviations

It is recommended that abbreviations not be used in clinical reports. However, abbreviations are sometimes required because of limitations in the number of characters allowed in a computerized database. Furthermore, certain abbreviations are acceptable because they are universally understood, such as deoxyribonucleic acid (DNA), complementary DNA (cDNA), ribonucleic acid (RNA), messenger RNA (mRNA), polymerase chain reaction (PCR), base pair (bp), and kilobase (kb). Because confusion exists about whether RT-PCR stands for real-time PCR or reverse transcription PCR, either spell out the words or define ''Q-PCR'' to designate quantitative PCR and ''Q-RT-PCR of RNA'' to designate quantitative reverse transcription PCR. Once defined on first use in a report, the abbreviation may be used in the rest of the report.

Results

The result is the crux of every report. The result should be stated clearly and concisely and should be based on objective criteria that are described in the laboratory's procedure manual. Many molecular tests yield raw data, which must be interpreted before the result is evident. For example, bands on a gel must be evaluated visually or scanned to yield an interpretable electropherogram. Quantitative amplification assays may require extrapolating from a standard curve and interpreting that result in the context of the amount of control sequence that was amplified from the same sample. Interpretation criteria may depend on whether the control sequence was an exogenous DNA or RNA that was spiked in known amount versus an endogenous control sequence that is expected to vary (eg, when a patient's white blood cell count is low then the level of DNA or housekeeping RNA in the blood might also be low).

The expected result in a normal individual should be stated. This is expressed as a normal range for a quantitative test, along with the units of measurement. Interassay reproducibility is an important parameter that allows a clinician to determine whether 2 sequential test results are significantly different from each other, and information about assay reproducibility (and other performance characteristics) can be conveyed in the individual report or in a document that is readily accessible to the clinician.

Most qualitative assays have a clearly defined normal and abnormal outcome, although further explanation is sometimes required to interpret whether a base change is a disease-related mutation, a benign polymorphism, or a variant of undetermined significance. (Guidelines entitled ''Interpretation of Sequence Variants'' are published by the American College of Medical Genetics, www.acmg.net/resources/policy-list.asp.) Likewise, it is useful to specify whether detection of a microbe is interpreted as pathologic or normal flora. Some tests have no normal range, such as parentage testing or HLA-based organ matching in which the result is interpreted in concert with results on another person's sample.

Most laboratory tests have limitations that make them subject to false-positive or false-negative results, thus imparting a probabilistic risk of disease rather than an absolute answer. Depending on the performance characteristics of each assay and the clinical impact of an incorrect result, it is worth describing these limitations in the report. Thus, some molecular pathology reports may include verbiage on assay sensitivity, specificity, and/or precision. Analytic sensitivity refers to how good the assay is at detecting a specific molecular target or defect, whereas clinical sensitivity refers to how good the assay is at detecting the associated disease. When multiple genotypes can cause the same phenotype, or when the assay is not 100% sensitive for detecting the disease, it is important to describe the residual risk of disease. As an example, validation studies may show that a PCR assay is 100% sensitive for detecting a translocation involving the *CCND1* region on chromosome 11q13 and the *IGH* gene joining region on chromosome 14q32, as long as the tumor clone composes at least .001% of the cells in the specimen. Yet the PCR assay is only about 50% sensitive for detecting mantle cell lymphoma because about half of all mantle cell lymphomas have breakpoints outside of the major translocation cluster region targeted by this assay. When reporting negative molecular results in a marrow with a morphologic differential diagnosis of mantle cell lymphoma, the report might state that ''no *CCND1/IGHJ* was detected to a sensitivity of approximately 1 in 100 000 marrow cells; although no *CCND1/IGHJ* translocation was detected, this does not exclude mantle cell lymphoma because only about half of all mantle cell lymphomas have a detectable translocation.'' When testing for minimal residual disease in a patient whose original tumor was never assayed for this translocation, the report of a negative test might add that ''these results should be interpreted with caution because our laboratory has no record that this patient's original tumor was characterized by a detectable translocation.''

The British Clinical Molecular Genetics Society and the Organisation for Economic Cooperation and Development

Table 2. Partial Report of a Negative Cystic Fibrosis Screen

Test Name: Cystic Fibrosis Carrier screen
Result: Negative for the 32 *CFTR* gene mutations listed below.
Interpretation: There is no detectable *CFTR* gene mutation, decreasing the probability that this patient is a carrier of cystic fibrosis.
Comment: This negative result does not eliminate the possibility of a *CFTR* mutation not tested for in this panel. In the table below are the estimated detection rates for this test panel in various ethnic groups expressed as pretest and posttest carrier risks in individuals with a negative family history of cystic fibrosis.

Ethnic Group	Detection Rate, %	Pretest Risk	Posttest Risk
Ashkenazi Jewish	97	1/29	1/930
European Caucasian	80	1/29	1/140
African American	69	1/65	1/207
Hispanic American	57	1/46	1/105
Asian	Unknown	1/90	Unknown

References: *Genet Med*. 2004;6:387–391; www. genetests.org; www.acmg.org.

Procedure: The *CFTR* gene was analyzed for the 32 mutations listed below by polymerase chain reaction and allele specific oligonucleotide ligation (Abbott Laboratories CF v3.0). [Each laboratory should list the specific mutations tested for here.[23]]

have drafted Best Practice Guidelines for performing and reporting molecular genetic test results (www.cmgs.org and www.oecd.org). The American College of Medical Genetics has also devised guidelines for clinical genetics laboratories (www.acmg.net). For example, it is recommended that a report of a negative screening test for cystic fibrosis carrier status describe the residual risk in relation to ethnicity, and example wording of such a report is shown in Table 2. The Clinical and Laboratory Standards Institute (formerly NCCLS, www.nccls.org) has published several documents that include recommendations for molecular report content: *MM1, Molecular Diagnostic Methods for Genetic Diseases; MM2, Immunoglobulin and T-Cell Receptor Gene Rearrangement Assays; MM3, Molecular Diagnostic Methods for Infectious Diseases; MM5, Nucleic Acid Amplification Assays for Molecular Hematopathology; MM6, Quantitative Molecular Methods for Infectious Diseases; MM7 Fluorescence In Situ Hybridization (FISH) Methods for Medical Genetics; MM9, Nucleic Acid Sequencing Methods in Diagnostic Laboratory Medicine;* and *MM14, Proficiency Testing (External Quality Assessment for Molecular Methods).*

Every laboratory must do validation studies to show how well each assay performs in its facility.[6] Information on assay performance characteristics should be made available on request but need not be included in every patient report.

Interpretation

There are 2 kinds of interpretation, analytic interpretation and clinical interpretation. Analytic interpretation involves examining the raw data and forming a conclusion about the quality or quantity of the analyte, in other words producing a reportable result. Clinical interpretation involves describing what the result means for the patient, either in general or based on specific knowledge of that patient's situation. An example of the difference between analytic and clinical interpretation is shown in the Table 3.

Because molecular tests are relatively complex and may be done using any variety of methods, interpretation helps ensure that the clinical significance of the result is apparent and that the analytic limitations of the assay are disclosed in relation to the findings of that case. Interpretations are especially welcome by busy clinicians who are expected to synthesize an overwhelming amount of information to practice evidence-based medicine, and yet they often lack sufficient time and resources for optimal assessment of the current medical literature with regard to use of laboratory tests in clinical decision making.[7] Laboratory professionals can help fill the gap by composing reports that not only provide an analytic interpretation of the raw data but also provide guidance on the impact of the result for patient management.[8] The principles of evidence-based laboratory medicine should be followed as summarized at the International Federation of Clinical Chemistry and Laboratory Medicine Web site (www.ifcc.org).

Assays should be interpreted by individuals who are competent in the analytic and clinical aspects of the test.[9] A lesson regarding interpretation of complex laboratory data comes from colleagues in coagulation laboratories who recently showed that most ordering clinicians perceive that a pathologist-generated test interpretation saves them time, assists with differential diagnosis, and helps prevent misdiagnosis.[10] Clinicians who were surveyed about cystic fibrosis and factor V Leiden reports preferred comprehensive molecular reports that helped guide clinical decision making.[3] Composing such reports requires expert technical knowledge as well as medical expertise, sometimes indicating other laboratory or clinical parameters that should be investigated. Clinical interpretation further offers patient-specific analysis and understanding of the laboratory findings.

If general comments are routinely inserted into every report, they should be brief and educational with regard to test limitations or clinical significance. Alternatively, one should reference a reliable Web site or publication at which generic test information is found. Too much verbiage is burdensome to the clinician who wants to quickly evaluate laboratory findings and their significance, and it is a liability when actionable findings are hidden in a sea of words. Clinicians seem to prefer a summary of the findings at the top of the report.[1] The report should be limited to 1 printed page except in rare circumstances.

It is important to communicate with clinicians who read laboratory reports to get feedback on content and utility. The cautious terminology of pathologists often contrasts with the preferred terms of surgeons.[11] However, clinicians must keep in mind that laboratorians typically report what is known to be true based on objective data and expert technical knowledge, and beyond that the laboratorian may use his or her judgment to interpret the findings along with terms such as ''consistent with.''

Table 3. A Sample Molecular Pathology Report*

Patient Name: John Doe
Date of Birth: 1/2/2003
Testing Laboratory: (laboratory name and address, phone number, Web site)
Date of Collection: 7/10/06
Date Received: 7/10/06
Ordering Physician: John Smith, MD
Sample Type: Blood
Clinical Indication: Developmental delay
Test Name: Fragile X genotyping
Result: Abnormally large methylated *FMR1* gene segment, the size of which is in the ''full mutation'' range estimated as more than 200 trinucleotide repeats.
Analytic Interpretation: Fragile X genotype†
Clinical Interpretation: The findings are consistent with a diagnosis of fragile X syndrome of mental retardation.†
Comment: Genetic counseling is recommended. For further information see www.genetests.org and www.acmg.net.
Procedure: Southern blot analysis was performed using *Eco*RI and methylation-sensitive *Eag*I restriction enzymes followed by hybridization with the StB12.3 probe‡ targeting the *FMR1* gene on chromosome Xq27. This region contains a trinucleotide CGG repeat sequence with a range of 5 to approximately 44 repeats expected in the normal population. An allele of 45–58 repeats, called a gray zone allele, may slightly expand or contract in size but does not expand to a full mutation in the next generation. An allele of 59–200 repeats, termed a premutation allele, may expand to a full mutation in offspring; the risk of expansion is related to increasing repeat size. A full mutation allele contains >200 repeats and is generally associated with inappropriate methylation, lack of expression of FMR1 protein, and fragile X syndrome. Rare individuals with incomplete methylation of a full mutation allele have milder forms of disease.
Disclaimer: This test was developed and its performance characteristics determined by the Molecular Genetics Laboratory. It has not been cleared by the US Food and Drug Administration. However, such approval is not required for clinical implementation, and test results have been shown to be clinically useful. This laboratory is CAP accredited and CLIA certified to perform high complexity testing.
Signature: Electronically signed by (signature of pathologist or designee)
Date of Report: 7/17/06
***ICD-9* Code:** 315.2
***CPT* Codes:** 83891, 83896, 83892x2, 83894, 83897, 82397, 83912

* CAP indicates College of American Pathologists; CLIA, Clinical Laboratory Improvement Amendments of 1988; *ICD-9, International Classification of Diseases, Ninth Revision*; and *CPT, Current Procedural Terminology.*

† The interpretation section is divided into analytic and clinical portions for purposes of illustration; actual clinical reports need not distinguish results from analytic interpretation, or analytic from clinical interpretation.

‡ The ''National Center for Biotechnology Information (NCBI) ID No.'' of the probe could have been listed if the probe sequence had been previously deposited into the NCBI probe database.

The difficulty of composing a comprehensible report was evident in a study entitled ''Clinicians are from Mars and Pathologists are from Venus'' in which surgeons misunderstood anatomic pathologist's reports 30% of the time.[12] The gap can be bridged by personal communication, education of both the report writer and the reader, and attention to details. More experienced or subspecialist clinicians may have different perceptions of report content than do generalists or junior colleagues. In general, it is advisable to write each report so that it is easily understood by a nonspecialist physician. One should keep in mind that the report may be read by nonphysicians ranging from nurses to genetic counselors, epidemiologists, researchers, trainees, and even patients. It is reasonable to include references to published literature, to the testing laboratory's Web site, or to reliable external Web sites where further information is available. For inherited disease testing, one should consider referencing www.genetests.org, www.ncbi.nlm.nih.gov/omim, or www.acmg.org. For cancer testing, useful information is often found at www.infobiogen.fr/services/chromcancer and http://cgap.nci.nih.gov. For many types of laboratory tests, technical and clinical information may be available at www.labtestsonline.org.

REPORTING CONTROL TEST RESULTS

Nearly every clinical laboratory test is run alongside external and internal controls. External controls are used to ensure that the assay performed as expected, whereas internal controls are used to assess quality or quantity of the patient sample. Because controls are required by US federal law[13,14] and their outcomes are assumed to be within required limits in order for the result to be considered valid, the results of controls are not generally reported. However, in some cases a control result is considered worthy of mention if it is unusual or pertinent to interpretation of the findings. For example, if morphologic examination shows that the proportion of tumor is low but not so low as to be unacceptable for analysis, it is appropriate to comment on marginal sample adequacy. Likewise if a control RNA is shown to be partially degraded by Q-RT-PCR but is still within acceptable limits, then it is appropriate to mention that assay sensitivity for detecting a low-level transcript may be reduced. In DNA fingerprinting assays in which donor and recipient controls are normally used to assess the degree of engraftment in an allogeneic transplant patient sample, if only 1 of these 2 controls is available for comparison then a cautionary comment is warranted regarding the limitations of the assay. As a final example, genetic testing of a fetal sample is often accompanied by an identity test to show what proportion of the sample is of fetal versus maternal origin, with additional genetic testing of 1 or both parents to help interpret the significance of the fetal test results.

CHANGES TO REPORTS

An ''addendum'' report generally adds new information, whereas an ''amended'' report changes information that was previously reported. Addendum reports often contain a phrase such as ''this report was updated to in-

clude. . . . '' Amended reports should describe the change and, if appropriate, the reason for the change. Likewise, discrepancies between a preliminary report and a final report must be documented. It is advisable to personally contact the ordering physician if the change is likely to affect clinical management.

The introduction of a new test, or major changes in the way that an existing test is done or reported, should be explained to clinicians. It is advisable to get feedback from key clinicians prior to making major changes. In some cases, it is appropriate to emphasize the change within the text of the patient report.

INTEGRATION OF MOLECULAR RESULTS INTO ANATOMIC PATHOLOGY CONSULTATIONS

When molecular testing is performed on a sample that is also undergoing routine anatomic pathology workup, such as a biopsy or a bone marrow aspirate, the option exists to report the molecular findings in a freestanding report or to integrate them with other clinicopathologic data into a single anatomic pathology report.[4] Regardless of whether the molecular result is reported separately or not, it is important that a physician review the overall findings. In general, it seems to be the primary responsibility of the physician who selects and orders an ancillary test, whether it be the consulting anatomic pathologist or the clinician, to synthesize the test results with other pertinent clinicopathologic information. This synthesis is above and beyond any work performed by the molecular pathologist in the analytic and clinical interpretation of the molecular test.

The CAP recommends that patient-specific clinical laboratory records be retained for at least 2 years unless they are considered anatomic pathology reports (10 years) or cytogenetic reports (20 years). Guidelines for producing anatomic pathology reports, including ancillary test components, are found on the CAP Web site (www.cap.org) and on the Web site of the Association of Directors of Anatomic and Surgical Pathology (www.adasp.org). Recently, efforts have been made to improve the consistency of content and format of anatomic pathology reports. The process of standardizing reports can be surprisingly difficult, as was evident when the American College of Surgeons Commission on Cancer recently adopted reporting guidelines created by the CAP.[2] These guidelines, which represent a list of essential elements in pathology reports for cancer-directed surgical resection specimens, were initially perceived by some pathologists to be confusing until the recommendations were refined and the flexibility of reporting styles was ensured.[2] Many institutions are trending toward synoptic cancer reports in which checklists and templates are used to facilitate reporting of the essential elements and their sequence. Thomas McGowan recently showed the feasibility of adopting a formatted, synoptic anatomic pathology report (www.cancercare.on.ca/QualityIndex2006/measurement/pathologyReports/index.html). Similar principles and benefits are pertinent to molecular pathology reports.

CODING AND BILLING DOCUMENTATION

The pathology report is often used by administrators and third-party payors to justify payment for services rendered. The *ICD-9* (*International Classification of Diseases, Ninth Revision*) code is used to document the reason(s) for testing. It is the responsibility of the ordering physician to use sound judgment in deciding which laboratory tests to order on the patient. Laboratory professionals often provide crucial advice on which test(s) to perform in a given situation. Payors, in turn, may use the *ICD-9* code to determine if a test request is medically necessary. It is acceptable for a pathologist to alter an *ICD-9* code once the test result reveals a more specific diagnosis. For example, fragile X genotyping may be ordered because of *ICD-9* code 315.2 (developmental learning difficulty), but the code may be revised to 759.83 (Fragile X syndrome) once the positive test result is correlated with the findings on history and physical examination to establish a diagnosis.

Current Procedural Terminology (*CPT*) codes document the analytic and interpretive procedures that are performed in the laboratory. Clinical interpretation of a molecular test result is generally billable by a pathologist (or other physician) using *CPT* code 83912-26 (interpretation and report) or, when a consult regarding an abnormal test result is requested by the patient's attending physician, using codes 80500 (clinical pathology consultation, limited) or 80502 (comprehensive consultation of complex diagnostic problem). Analytic interpretation is billable by a physician using code 83912-26 except for certain microbiology tests for which interpretation is already included in the molecular test code for that organism. Physicians as well as nonphysician scientists who interpret molecular results can bill the technical component of 83912. One should visit www.clinbioinformatics.org for an update on how laboratory information systems might be used to codify molecular and cytogenetic test results for purposes of billing as well as for linking to larger medical nomenclature systems (eg, SNOMED-CT and LOINC).

REPORTING RESEARCH TEST RESULTS

Research test results should not be included in clinical reports. If an assay is not yet validated for clinical use, then test results should be shared with a subject's physician only in the context of an institutional review board–approved study protocol. One should consider reporting unvalidated test results in a spreadsheet or in a letter in which it is clearly stated that the test was done on a research basis. Just because an assay is validated for use on 1 sample type does not imply that the assay is valid on other sample types, and it is up to the laboratory director to determine when validation work is sufficient to justify application of an assay to a particular sample type.

There are some grey areas that warrant the discretion of the professionals involved, analogous to the off-label use of a drug. An individual laboratory director or clinician can choose to make an exception to the standard policy or to take risks that are perceived to benefit an individual patient, for example when an orphan disease is involved and there is no option to send the sample out for validated testing. However, such exceptions should be handled with caution because federal law defines minimal standards that must be met by clinical labs,[13,14] and the public expects that relevant laws are followed.

Clinical trials often include laboratory testing in the research protocol. Any laboratory test whose results are used for patient management, even as part of a clinical trial that is paid for by the trial sponsor, must be validated, performed, and reported by a CLIA-certified clinical laboratory (www.ncrr.nih.gov/clinical/gcrcpatientsafety20010622.asp#XI).

GENE NOMENCLATURE

Because genes were discovered and named in many different research laboratories worldwide, sometimes after years of concurrent research, it was inevitable that there would be multiple names for the same gene. Because it is confusing to have multiple names, it was equally inevitable that a process would be developed to select a single name. Each gene is also assigned a shortened name, called a symbol, that is frequently used in reports and publications.

Which of the following gene symbols is correct, bcl-2, Bcl-2, bcl2, Bcl2, *BCL2,* *BCL2,* BCL2, *bcl*II, or *Bcl*II? According to the Human Genome Organisation Gene Nomenclature Committee, the correct symbol for the gene named ''B cell CLL/lymphoma 2'' is *BCL2.*

Gene symbols are in upper case Latin letters ± Arabic numerals. Gene symbols are italicized (*BCL2*), whereas the protein is not (BCL2). To distinguish between mRNA and cDNA, the relevant prefix is shown in parentheses: (mRNA)*BCL2* or (cDNA)*BCL2.* No superscripts or subscripts are accepted. No Roman numerals (I, II, III, IV, V, etc) or Greek letters (α, β, γ, . . . , κ, λ) are accepted. Oncogenes should have no prefix (*MYC*, not *c-MYC; MLH1,* not *hMLH1*). No punctuation is permitted within a gene name (except for HLA and antigen receptor genes). HLA alleles are assigned by the World Health Organization Nomenclature Committee for Factors of the HLA System, whereas immunoglobulin and T-cell receptor genes are named by the International ImMunoGeneTics (IMGT) Information System Nomenclature Committee (http://imgt.cines.fr).

A searchable database of correct gene names and symbols is found on the Web site of the Human Genome Organisation Gene Nomenclature Committee (www.gene.ucl.ac.uk/nomenclature). Selected gene symbols associated with leukemia, lymphoma, and sarcoma are shown in Table 4. The correct gene name and symbol is not necessarily the same as the common one used the medical literature; one should note that *ABL1* (not *ABL*) is the gene associated with chronic myelogenous leukemia, and *ETV6/RUNX1* (not *TEL/AML1*) is the translocation commonly found in childhood acute lymphoblastic leukemia.

Now that the rules have been summarized as set forth by the professional groups that are responsible for naming genes, it is important to address how to maximize their utility in the health care system. First, it is important that proper gene nomenclature be used in clinical documents to facilitate communication among health care providers and those who interface with providers. Laboratory information system programmers are aware of this and are working to facilitate the process. A change in the way gene tests are reported may cause confusion in the short term, but it will ultimately serve the health care system well in the long term. Second, manufacturers of kits and analyte-specific reagents must use proper gene nomenclature. Third, researchers and journal editors should be encouraged to adopt the same nomenclature system that is being adopted in clinical settings, so that all scientists are communicating in the same ''language.'' These are lofty goals that most will agree on in principle, but several obstacles must first be overcome. Some of the problems as well potential solutions are discussed later.

ANTIGEN RECEPTOR GENES

The immunoglobulin and T-cell receptor genes are among the most complex human genes because they somatically rearrange during physiologic development of B and T lymphocytes. Proposed symbols for these genes are shown in Table 5. In the T-cell receptor γ locus, *TRGV1* through *TRGV11* are the 11 variable regions, whereas *TRGJ1, TRGJ2, TRGJP, TRGJP1,* and *TRGJP2* are the 5 joining regions. One should note that some approved gene symbols end with the ''at'' symbol, for example, *IGH@,* to designate a locus that encompasses several domains (eg, V, D, J, and/or C). Our consensus recommendation is to drop the ''@'' in clinical reports, in part because of the risk that information systems will not process this symbol properly.

TRANSLOCATIONS

Translocations are common in hematopoietic malignancies and in sarcomas. A translocation can influence disease pathogenesis by encoding a fusion protein as with *PML/RARA* or by juxtaposing promoter and enhancer elements of 1 gene with another gene whose transcription is then dysregulated, as with *IGH/BCL2.* Translocation nomenclature is already well established in the cytogenetics arena. The chronic myelogenous leukemia translocation is expressed as t(9;22)(q34;q11.2) for the karyotype designation, or as ''nuc ish 9q34(ABL1x3,),22q11.2(BCRx3)(ABL1conBCRx2)'' when interphase fluorescence in situ hybridization using a dual fusion strategy is used. An International System for Human Cytogenetic Nomenclature encompasses recommendations for karyotype and fluorescence in situ hybridization nomenclature,[15] but the guidelines for fluorescence in situ hybridization are sometimes difficult to apply in clinical situations,[16] thus emphasizing the need for analytic and clinical interpretation of test results in patient reports.

When a translocation is identified by amplification methods such as RT-PCR, it seems reasonable to list the 2 juxtaposed or fused genes separated by a slash (*PML/RARA*). Some have argued that a hyphen should be used (*PML-RARA*) but a hyphen means ''deleted'' in cytogenetic terminology. A semicolon (*PML;RARA*) would be more consistent with karyotype nomenclature. One group has recommended no syntax at all between 2 recombined genes (*PMLRARA*).[17] However, it is essential to use some form of punctuation so that it is clear when one gene name ends and when the next one starts. Our consensus recommendation is that a slash be used to separate the 2 gene names.

As for which gene is listed first, some have recommended that the genes be listed in order of their appearance in the karyotype, as is done for cytogenetic nomenclature. In this scheme, genes on chromosome 1 are listed before those on chromosome 2, p arm before q arm, and centromeric before telomeric. Others argue that karyotypic location is less important than oncogenic mechanism when describing an event at the molecular level, and therefore fused genes should be listed starting 5′ and ending at the 3′ terminus of a fusion transcript. Thus, chronic myelogenous leukemia has a *BCR/ABL1* translocation because the 5′ portion of *BCR* is fused with the tyrosine kinase domain of the 3′ portion of *ABL1.* In fact, the leukemic cells also produce *ABL1/BCR* fusion product, but these reciprocal transcripts are not known to be oncogenic, in contrast to *BCR/ABL1* transcripts. Is it imperative to know the structure and oncogenicity of the genes involved in the translocation before deciding how to report it? In many cases this information is known, although there are some translocations for which the affected gene may be

Table 4. Common Translocations in Hematopoietic Neoplasms and Sarcomas

Tumor Type*	Karyotype	Karyotype Order	Mechanistic Order
Myeloid leukemias			
CML and pre-B-ALL	t(9;22)(q34.1; q11.2)	*ABL1/BCR*	*BCR/ABL1*
AML-M2	t(8;21)(q22; q22.3)	*RUNX1T1/RUNX1*	*RUNX1/RUNX1T1*
AML-M3	t(15;17)(q24;21.1)	*PML/RARA*	*PML/RARA*
AML-M3, atypical	t(5;17)(q35; q21.1)	*NPM1/RARA*	*NPM1/RARA*
AML-M3, atypical	t(11;17)(q23; q12)	*ZBTB16/RARA*	*ZBTB16/RARA*
AML-M3, atypical	t(11;17)(q13; q12)	*NUMA1/RARA*	*NUMA1/RARA*
AML-M3, atypical	t(17;17)(q11.2; q12)	*STAT5B/RARA*	*STAT5B/RARA*
AML-M4eo	inv16(p13.1q22)	*MYH11/CBFB*	*CBFB/MYH11*
CMML	t(5;12)(q32; p13)	*PDGFRB/ETV6*	*ETV6/PDGFRB*
Chronic eosinophilic leukemia	t(4;4)(q12; q11)	*FIP1L1/PDGFRA*	*FIP1L1/PDGFRA*
B-cell leukemias and lymphomas			
Pre-B ALL	t(12;21)(p13; q22.3)	*ETV6/RUNX1*	*ETV6/RUNX1*
Pre-B ALL	t(1;19)(q23; p13.3)	*PBX1/TCF3*	*TCF3/PBX1*
Mixed lineage leukemia	11q23 translocations	partner/*MLL*	*MLL*/partner
Burkitt lymphoma	t(8;14)(q24; q32.3)	*MYC/IGH*	*IGH/MYC*
Burkitt lymphoma	t(2;8)(p12; q24)	*IGK/MYC*	*IGK/MYC*
Burkitt lymphoma	t(8;22)(q24; q11.2)	*MYC/IGL*	*IGL/MYC*
Mantle cell lymphoma	t(11;14)(q13; q32.3)	*CCND1/IGH*	*IGH/CCND1*
Follicular lymphoma	t(14;18)(q32.3; q21.3)	*IGH/BCL2*	*IGH/BCL2*
Diffuse large B cell lymphoma	t(3;14)(q27; q32.3)	*BCL6/IGH*	*IGH/BCL6*
Lymphocytic lymphoma	t(9;14)(p13; q32.3)	*PAX5/IGH*	*IGH/PAX5*
MALT lymphoma	t(14;18)(q32.3; q21)	*IGH/MALT1*	*IGH/MALT1*
MALT lymphoma	t(11;18)(q21; q21)	*BIRC3/MALT1*	*BIRC3/MALT1*
MALT lymphoma	t(1;14)(p22; q32.3)	*BCL10/IGH*	*IGH/BCL10*
Splenic lymphoma, villous lymphocytes	t(7;14)(q21; q32.3)	*CDK6/IGH*	*IGH/CDK6*
Plasma cell myeloma			
Myeloma	t(4;14)(p16.3; q32.3)	*WHSC1/IGH*	*IGH/WHSC1*
Myeloma	t(6;14)(p25; q32.3)	*IRF4/IGH*	*IGH/IRF4*
Myeloma	t(14;16)(q32; q22)	*IGH/MAF*	*IGH/MAF*
Myeloma	t(16;22)(q22; q11.2)	*MAF/IGL*	*IGL/MAF*
T-cell leukemias/lymphomas			
T-ALL	del(1)(p32p32)	*STIL/TAL1*	*STIL/TAL1*
T-ALL	t(7;11)(q35; p13)	*TRB/LMO2*	*TRB/LM02*
ALCL	t(2;5)(p23; q35)	*ALK/NPM1*	*NPM1/ALK*
Sarcomas of bone and soft tissue			
Alveolar rhabdomyosarcoma	t(2;13)(q35; q14.1)	*PAX3/FOXO1A*	*PAX3/FOXO1A*
Alveolar rhabdomyosarcoma	t(1;13)(p36; q14.1)	*PAX7/FOXO1A*	*PAX7/FOXO1A*
Ewing sarcoma/PNET	t(11;22)(q24; q12)	*FLI1/EWSR1*	*EWSR1/FLI1*
Ewing sarcoma/ PNET	t(21;22)(q22.3; q12)	*ERG/EWSR1*	*EWSR1/ERG*
Desmoplastic round cell tumor	t(11;22)(p13; q12)	*WT1/EWSR1*	*EWSR1/WT1*
Clear cell sarcoma	t(12;22)(q13; q12)	*ATF1/EWSR1*	*EWSR1/ATF1*
Low-grade fibromyxoid sarcoma	t(7;16)(q33; p11)	*CREB3L2/FUS*	*FUS/CREB3L2*
Myxoid liposarcoma	t(12;16)(q13; p11)	*DDIT3/FUS*	*FUS/DDIT3*
Myxoid liposarcoma	t(12;22)(q13; q12)	*DDIT3/EWSR1*	*EWSR1/DDIT3*
Myxoid chondrosarcoma, extraskeletal	t(9;22)(q22; q12)	*NR4A3/EWSR1*	*EWSR1/NR4A3*
Synovial sarcoma	t(X;18)(p11.2; q11.2)	*SSX1/SS18*	*SS18/SSX1*
Synovial sarcoma	t(X;18)(p11.2; q11.2)	*SSX2/SS18*	*SS18/SSX2*
Dermatofibrosarcoma protuberans	t(17;22)(q22; q13)	*COL1A1/PDGFB*	*COL1A1/PDGFB*
Alveolar soft part sarcoma	t(X;17)(p11.22; q25)	*TFE3/ASPSCR1*	*ASPSCR1/TFE3*
Infantile fibrosarcoma	t(12;15)(p13; q25)	*ETV6/NTRK3*	*ETV6/NTRK3*
Endometrial stromal sarcoma	t(7;17)(p15; q11.2)	*JAZF1/SUZ12*	*JAZF1/SUZ12*
Inflammatory myofibroblastic tumor	2p23 translocations	*ALK*/partner	partner/*ALK*

* CML indicates chronic myelogenous leukemia; ALL, acute lymphoblastic leukemia; AML, acute myelogenous leukemia; CMML, chronic myelomonocytic leukemia; MALT, mucosa-associated lymphoid tissue; ALCL, anaplastic large cell lymphoma; and PNET, primitive neuroectodermal tumor.

quite distant from the translocation breakpoint (as with CCND1 dysregulation in mantle cell lymphoma) or for which the affected genes remain uncertain, or for which multiple genes are affected by the chromosomal rearrangement. Common translocations are listed in Table 4 in ''karyotype order'' and, in a separate column, in ''mechanistic order.'' Until further consensus is achieved on this issue, it is recommended that laboratory directors and scientists use their own judgment in listing the order of translocated gene partners.

When the affected genes have not yet been defined, then the locus that is probed should be designated. For example, BCL1 or 11q13 were commonly used to describe the targeted locus before the *CCND1* gene was identified as being consistently dysregulated in mantle cell lymphoma and myeloma bearing t(11;14). This raises another ques-

Table 5. Nomenclature of Selected Antigen Receptor Genes

Official Gene Symbol	Proposed for Clinical Reports*
IGH@	*IGH*
IGHJ@ (IGH joining group)	*IGHJ*
IGK@	*IGK*
IGKJ@	*IGKJ*
IGKC (κ constant region)	*IGKC*
IGL@	*IGL*
TRA@ (T-cell receptor α locus)	*TRA*
TRB@	*TRB*
TRG@	*TRG*
TRD@	*TRD*

* Caution: The Human Genome Organisation Gene Nomenclature Committee proposes *TRD* as the official symbol of the tRNA aspartic acid gene. Our consensus recommendation is *TRD* also be used as the symbol for the T-cell receptor δ gene in the context of reporting molecular test results for lymphoid neoplasia.

tion: When amplifying across the translocation breakpoint, is it reasonable to report that PCR for the ''*CCND1/IGHJ*'' was done, even though no probe for the *CCND1* gene itself was used in the test? Or would clinicians understand a report better if the traditional BCL1 terminology were used? Or is it preferable to use cytogenetic nomenclature, even when karyotype was not performed, by reporting that PCR for the ''t(11;14)(q13;q32)'' was done? The consensus recommendation is that the term *CCND1/IGHJ* be used to describe this molecular assay. The corresponding analytic interpretation might be worded as ''a translocation was identified between the joining region of the *IGH* gene and the major translocation cluster near the *CCND1* gene, consistent with the presence of t(11;14)(q13;q32).'' The ultimate decision on how to report this and other molecular findings remains largely at the discretion of the interpreting laboratory scientist.

When Southern blot analysis is used to detect a translocation, only 1 of the gene partners is typically probed, whereas a putative partner gene juxtaposition may be inferred. For example, *BCR* gene rearrangement by Southern blot analysis implies *ABL1/BCR* translocation. *MLL* gene rearrangement, on the other hand, may be caused by a myriad of genetic events including translocation, inversion, insertion, partial deletion, or duplication, and the implications of such rearrangement for prognosis and detecting minimal residual disease warrant comment in the report.

MUTATIONS, DELETIONS, AND OTHER GENETIC VARIATIONS

Nomenclature for allele variation is the purview of the Human Genome Variation Society (www.hgvs.org). Their Web site describes the rules, gives examples, and provides links to databases in which numerous disease-related gene mutations are cataloged. Nomenclature rules are complex and depend at least in part on the consequence of the nucleotide-level defect on translation of the protein. It is unrealistic to determine the consequence of the defect in every patient sample, so one depends on research, preferably published in the medical literature or cataloged in reliable public databases, to infer the biochemical consequence and interpret the clinical significance of a given variant. Progress has been made in establishing databases to catalog disease-causing mutations, benign polymorphisms, and variants of undetermined significance.[18–20]

Nomenclature for genetic variation is generally based on sequences deposited in GenBank. It must be made clear which reference sequence is used by specifying the accession number from a public database, and whether this sequence represents cDNA or genomic DNA, which impacts on whether c. or g. should proceed the nucleotide number.[21] A Web resource called RefSeq found at www.ncbi.nlm.nih.gov/RefSeq seeks to establish reference sequences for each human gene and for the genomes of selected microorganisms. This is being accomplished by synthesizing the data in GenBank to create a standard gene sequence along with corresponding RNA and protein sequences. Because provisional sequences may change before they are validated and finalized, this database is a moving target. Furthermore, a single gene may have multiple RefSeqs reflecting cDNA splice variants. For example, the *HFE* gene currently has 11 different cDNA RefSeqs in GenBank, each of which encodes a different isoform of the protein. The longest cDNA is 2222 bp (NM_000410.3), whereas the full gene sequence is 269 712 bp (NG_001335.1). The best reference sequence is not always evident, but choosing the longest cDNA sequence seems reasonable for most clinical applications. The consensus coding DNA sequence shows the nucleotide numbering for each cDNA RefSeq, with number 1 being A of the initiating ATG codon. The RefSeq project demonstrates progress toward establishing a common reference by which all other sequences are compared. In clinical reports, it remains important to designate the reference sequence number (accession number and version number) by which the patient's variations are compared.[23]

The historical designation of a particular mutation often differs from the correct one. For the *MTHFR* gene, NM_005957 is the GenBank accession number. *MTHFR* NM_005957.3:c.665C>T is the current recommended nomenclature for a cDNA-level mutation that is more commonly referred to in older literature as C677T. Because the *MTHFR* gene mutation is actually tested for at the DNA level rather than the cDNA level, it is reasonable to avoid splicing problems altogether by reporting the result at the genomic DNA level; however, an appropriate genomic DNA reference sequence does not yet exist in GenBank. The corresponding protein level change can be inferred and described in the interpretation section of the report, if desired, by adding the phrase ''encoding p.Ala222Val.''

Some examples of nomenclature for common genetic defects and their associated diseases are shown in Table 6. The laboratory findings should reflect the type of testing that was done, that is, a DNA-based test should describe any nucleotide-level defect that is found. It is recommended that the inferred amino acid change also be described, using brackets if the protein level change is deduced but not experimentally proved. We favor using the 3-letter amino acid nomenclature, for example, Cys282Tyr rather than C282Y, to avoid confusion about whether A, G, C, and T stand for the amino acids alanine, glycine, cysteine, and threonine or the nucleotides adenine, guanosine, cytosine, and thymine. One should use the prefix ''g.'' for genomic DNA, ''c.'' for cDNA or coding DNA, or ''p.'' for protein.

Molecular tests may yield results that represent unexpected byproducts of the way that test was performed, such as a base change adjacent to the one that is the primary target of the assay. These are reported at the discre-

Table 6. Nomenclature for Commonly Tested Inherited Disease Genes

Correct Nomenclature	Common or Prior Name	Associated Disease
F2 AF478696.1:g.21538G>A	*Prothrombin* 20210G>A or G20210A	Venous thrombosis*
F5 NM_000130.3:c.1601G>A [p.Arg534Gln]	*FV* Leiden or 1691G>A or R506Q	Venous thrombosis
HFE NM_000410.3:c.845G>A [p.Cys282Tyr]	*HFE* C282Y	Hemochromatosis
HFE NM_000410.3:c.187C>G [p.His63Asp]	*HFE H63D*	Hemochromatosis
SERPINA1 NM_000295.3:c.863A>T [p.Glu288Val]	*A1AT* PI*S allele, Glu264Val	α_1-Antitrypsin deficiency
SERPINA1 NM_000295.3:c.1096G>A [p.Glu366Lys]	*A1AT* PI*Z allele, Glu342Lys	α_1-Antitrypsin deficiency
CFTR NM_000492.3:c.1521_1523delCTT [p.Phe508del]	*CFTR* deltaF508 or 508delF or ΔF508	Cystic fibrosis

* Note: The F2 guanine to adenine substitution is in the 3′ portion of the gene that is transcribed but not translated, so an alternate way to express it would be c.*97G>A, which indicates that the mutation site is 97 bases beyond the end of the stop codon in the cDNA sequence.

Table 7. Partial Report of a Positive DNA Methylation Test Result

Results: The methylated (maternal) *SNRPN* allele was detected, but the unmethylated (paternal) *SNRPN* allele was not detected.
Interpretation: Only the methylated (maternal) *SNRPN* allele was detected, consistent with a diagnosis of Prader-Willi syndrome (see comment).
Comment: Methylation studies revealed the presence of only the methylated (maternal) allele of the *SNRPN* gene. This result suggests that there is either a deletion involving the paternally derived chromosome 15, maternal uniparental disomy (UPD) 15, or an error in imprinting (methylation) of the paternal chromosome 15. Any of these alterations is consistent with the diagnosis of Prader-Willi syndrome. Genetic counseling is strongly recommended to discuss the implications of this result and to discuss further testing that can be done to determine which genetic mechanism is responsible for the condition in this patient. A test for deletion of the relevant region of chromosome 15 is available in the cytogenetics laboratory (www. genetests.org).

tion of the laboratory director or designee. Like karyotyping, DNA sequencing is likely to yield more information than can be fully interpreted. In general, the sequencing report should list all of the variants that are found, categorizing them as mutations, polymorphisms, and ''variants of undetermined significance.''[22]

METHYLATION AND OTHER EPIGENETIC PHENOMENA

There are no established rules on how to report DNA methylation or other epigenetic events. We recommend a practical approach whereby the target gene or promoter is identified, the molecular findings are stated, and the report is kept concise but medically informative. Targeted CpG sites can be designated by a range of nucleotide numbers in a specified GenBank sequence. Table 7 is a sample report of a methylation study for a patient suspected of having Prader-Willi syndrome.

MICROSATELLITES AND NONCODING REGIONS

Clinical testing often involves analysis of highly polymorphic regions of DNA. Minisatellites, short tandem repeats (also known as microsatellites), and single nucleotide polymorphisms are commonly interrogated. Thousands of these polymorphic regions have been identified in human DNA, and selected loci are targeted by the forensic community for DNA fingerprinting of individuals in the national convicted offender database. The same polymorphic loci (and many others; see Table 8) are used by health care professionals for assessing donor cell engraftment, chimerism, allele loss, microsatellite instability, parentage, zygosity in twins, and matching samples to their human source. A searchable catalog of single nucleotide polymorphisms is available at the NCBI Web site and at www.hapmap.org. Nomenclature for describing single nucleotide polymorphisms is found in the guidelines of the Human Genome Variation Society at www.genomic.unimelb.edu.au/mdi/mutnomen. A list of common short tandem repeats has been compiled by the National Institute of Standards and Technology (www.cstl.nist.gov/biotech/strbase/index.htm). Further work is needed to identify the official gene symbol or NCBI accession number for each of the commonly tested loci.

Table 8. Common Polymorphic Short Tandem Repeat Loci Used for Identity Testing or Microsatellite Instability Testing

Locus, Common Name	Chromosome	Repeat Motif	Range of Common Repeat Alleles
Loci used for identity testing			
CSF1PO	5	TAGA	6–15
D3S1358	3	[TCTG][TCTA]	12–20
D5S818	5	AGAT	7–16
D7S820	7	GATA	6–14
D8S1170	8	[TCTA][TCTG]	7–18
D13S317	13	TATC	7–15
D16S539	16	GATA	5, 8–15
D18S51	18	AGAA	8–27
D21S11	21	[TCTA][TCTG]	24–38
FGA	4	CTTT	17–46.2
TH01	11	TCAT	4–13.3
TPOX	2	GAAT	6–13
vWA	12	[TCTG][TCTA]	10–22
Penta D	21	[AAAGA]	2.2–17
Penta E	15	[AAAGA]	5–24
D2S1338	2	[TGCC][TTCC]	15–28
D19S433	19	[AAGG]	9–18.2
Loci used for microsatellite instability testing			
BAT-26	2	[A]	26
NR-21	14	[A]	21
NR-24	2	[A]	24
MONO-27	2	[A]	27
BAT-25	4	[A]	25

MITOCHONDRIAL DNA ANALYSIS

A good source of information on mitochondrial DNA and associated clinical disorders is www.mitomap.org. The mitochondrial DNA sequences in the NCBI database should be used as a reference by which to report sequence variants, analogous to what is recommended for reporting alterations of chromosomal DNA. Interpretation of mito-

Table 9. Recommendations for Reporting Molecular Tests

1. Follow reporting guidelines in College of American Pathologists checklists including descriptions of the patient, sample, testing laboratory, method, and result.
2. Limit abbreviations to those described herein or to those defined on first use in a given report.
3. Raw data should be sufficiently interpreted to facilitate clinical decision making.
4. Discuss pertinent limitations of the test.
5. Keep the report concise and effective.
6. Report only validated assays and sample types.
7. Do not report results for controls except if relevant to interpretation.
8. Encourage clinical research so decisions are evidence-based.
9. Use proper gene nomenclature and encourage its use system-wide.
10. Encourage development of ''RefSeqGene'' to create a single reference sequence for each human gene (as well as for each human pathogen's genes).

chondrial test results is complicated by the variable numbers of targets per cell or per anatomic site and by the fact that point mutations or deletions in mitochondrial DNA usually increase exponentially with age and are inherited from the mother.

MICROORGANISMS

Decades ago, the American Society for Microbiology developed standards for bacterial gene names, and these rules are still used today in most clinical and research applications. Typically, bacterial gene names have 3 small letters representing the function or class of gene and a fourth capital letter for the specific gene (eg, *dnaG* encodes a DNA primase). Homologous genes in other organisms may have the same name, and therefore it is important to designate the bacterial genus and species. A compilation of bacteria names along with their genomic reference sequences (RefSeqs) is found at www.ncbi.nlm.nih.gov/genomes/lproks.cgi. Each RefSeq document specifies the gene name, DNA sequence, and amino acid translation. Mutations are designated by the nucleotide number, whereas the inferred amino acid substitution is reported by the codon number. It is unacceptable to report only the inferred amino acid change when the more specific nucleotide-level change was actually tested for in the laboratory.

Fungal genes have similar designations, and a list of fungal RefSeqs is found at www.ncbi.nlm.nih.gov/genomes/FUNGI/funtab.html. In the list of viral RefSeqs at www.ncbi.nlm.nih.gov/genomes/static/vis.html, one should note that Epstein-Barr virus is called by its official name of human herpesvirus 4, whereas human cytomegalovirus is called human herpesvirus 5. In clinical reports, it is sufficient to use the common medical name for each organism.

For human immunodeficiency virus mutations and genotypes, laboratories may consult the compilation of mutations updated by the International AIDS Society at http://iasusa.org/resistance_mutations/index.html. Mutations identified in the HIV1 reverse transcriptase or protease genes are commonly reported in conjunction with the inferred amino acid substitution. Multiple mutations should be listed individually.

PRACTICAL CONSIDERATIONS AND PROBLEMATIC ISSUES

The rules set forth by professional groups are not always practical when applied in clinical laboratories. To improve compliance and applicability, this article synthesizes the prior work of various professional groups, identifies areas where further work and consensus is needed, and makes recommendations as summarized in Table 9.

It will be difficult to adopt new nomenclature for certain genes or certain defects whose common names are entrenched in the literature and in patient records. It may even be dangerous if a patient report is worded in a way that is misunderstood by a clinician. However, the longer the delay in adopting consensus nomenclature, the more entrenched colloquial names become, and the harder it becomes to transition to a standard nomenclature system. During the transition period, it seems reasonable to use the correct terminology followed by colloquial terminology in parentheses, for example, *RUNX1T1/RUNX1* (*ETO/AML1*).

Although it is proper to display gene names in italics, the use of italics may be impractical if electronic information systems cannot process such formatting. Furthermore, it seems cumbersome to describe transcripts as ''(mRNA) PML/RARA'' when instead they could be termed ''PML/RARA transcripts.'' Thus, it is recommended that laboratories balance the previously published rules with the practicalities of implementing them. It is certainly acceptable to omit italics and to use the terms ''PML/RARA translocation,'' ''PML/RARA transcripts,'' ''PML/RARA cDNA,'' and ''PML/RARA protein'' to describe the various analytes and products that are relevant in clinical laboratory reports.

It seems reasonable to develop a single reference sequence as the gold standard by which all gene variants are described. This gold standard sequence will likely include designation of introns, exons, and 3′ and 5′ untranslated regions as well as promoter and enhancer regions and CpG islands, so that virtually all known DNA-based elements affecting structure or function or transcriptional regulation are captured. Likewise, a single cDNA sequence could be designated for each gene to use as a reference for describing changes in the coding region. The IMGT recommends that the sequence of the IG and TR genes be gauged against their ''allele*01'' reference sequences. For small nonhuman genomes such as viruses, it might be more straightforward to use the whole genome as the DNA reference sequence (eg, accession numbers having an ''NC_'' prefix in the NCBI database). Given the diversity of ''normal'' sequences, arguments can be avoided as to which variant is most representative of ''wild type'' by agreeing that it is more important to have a reasonable sequence ''written in stone'' than it is to have a representative sequence that takes years to devise. A project is being initiated at NCBI to develop a reference sequence that can be used for clinical reporting, a product

that is coined ''RefSeqGene.'' Until such time as ''RefSeqGene'' is available, it seems reasonable to continue to express the location of certain genetic variants by the nomenclature that clinicians are accustomed to, so that only 1 transition to a new numbering system is required.

Many other problematic issues in molecular reporting will undoubtedly emerge as progress continues in our understanding of genomics and disease. Such issues are commonly addressed by medical professionals working in teams and in conjunction with pertinent professional societies to gain consensus.

The evolving infrastructure for harmonizing gene nomenclature is taking the scientific community in the right direction. The groups responsible for assigning gene names are urged to restrain from changing names and symbols despite new discoveries elucidating each gene's structure, function, and clinical significance. Previous models demonstrate the benefits of a nomenclature system. The immunology community thrives 2 decades after developing the Cluster Designation system for naming surface antigens on hematopoietic cells. Another precedent dates back to the 1930s when it was recognized that the ABO blood group antigens needed consistent nomenclature. However, history also teaches us that widespread consensus is not always achieved as evidenced by persistent disagreement on an International System of Units (SI). Nevertheless, it is timely and important for patient care that we now move forward with a structure for reporting genetic findings in a systematic fashion. This requires collaboration with basic and clinical scientists from all areas of medicine.

FEEDBACK

To provide feedback, please send correspondence to jkachin@cap.org. Updated versions of these recommendations are posted on the CAP Web site at www.cap.org.

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