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Discharge Criteria for Children Sedated by Nonanesthesiologists

Is "Safe" Really Safe Enough?

ONE of the dilemmas faced by any caregiver providing sedation or anesthesia is the question, When is it safe to send the child home? Most sedation guidelines suggest that the patient should be returned to their baseline status before discharge. In this issue of the Journal, Malviya *et al.* describe a simple but elegant means for assessing the street readiness of infants sedated with chloral hydrate.[1]

One of the reasons that it has been so difficult to make sedation safe is the long-standing battle between specialties regarding definitions, drugs, monitoring, and qualifications. Organizations have modified definitions* or developed guidelines to fit the needs of their specialty.[2] The first guideline for monitoring children sedated for diagnostic procedures was published by the American Academy of Pediatrics (AAP).[3] Unfortunately, we adopted language from the National Institutes of Health regarding dental sedation, especially the misnomer "conscious sedation," an oxymoron in the pediatric population. The AAP later revised the guideline. Pulse oximetry was required for all sedated children and a systematic approach similar to that used by anesthesiologists was developed, *i.e.*, proper fasting, informed consent, focused airway examination, medical and/or surgical history, family history, previous sedation experiences, recommended equipment and medications, proper monitoring and documentation during and after the procedure, and strict discharge criteria.[4] During the following years, the American Society of Anesthesiologists (ASA) became involved with sedation safety, in part because the Joint Commission of Accreditation of Healthcare Organizations (JCAHO) modified their regulations in such a way that made departments of anesthesiology responsible for developing "within institution" sedation guidelines. In response to the JCAHO requirements, and with a strong emphasis on improving safety, the ASA established a task force that developed the guideline for sedation by nonanesthesiologists.[5] The first ASA iteration succeeded in changing the terminology from the oxymoron "conscious sedation" to the more appropriate term "sedation/analgesia," but it did not address deep sedation. In 2002, the ASA published revised sedation guidelines that address all depths of sedation.[6] The ASA, working closely with JCAHO, also developed new language to describe the sedation process,† which was later incorporated by the JCAHO.‡ Now, three stages of sedation are described: minimal, moderate, and deep. Recently, the AAP adopted the ASA definitions for their sedation guidelines[7]; now the AAP, ASA, and JCAHO are speaking the same language. In addition, the JCAHO introduced the essential concept of rescue, *i.e.*, the practitioner must have the skills to rescue should the patient progress to a deeper level of sedation than intended. The JCAHO has been our friend by forcing conformity in the sedation process and the required airway management skills throughout many institutions. Unfortunately, these regulations do not yet apply to private practitioners' offices.

Our specialty is uniquely positioned to improve the sedation process. Now that the language is uniform and the definitions are clear, it is time to examine safety concerns and to explore issues not addressed in any guidelines. What qualifications are needed to administer sedation? How do individuals gain credentials to administer sedation? What drugs have the best efficacy and safety profile? Several years ago, I had the good fortune to be granted access to the adverse medication reports associated with pediatric sedation accidents collected by the Food and Drug Administration.[8] Sixty of 95 cases were associated with death or neurologic injury. Contributory factors included drug overdose, drug interactions (*e.g.*, opioid and benzodiazepine), inadequate monitoring, inadequate medical evaluation, premature discharge, inadequate resuscitation skills, and others. Barbiturates, opioids, benzodiazepines, and sedatives were equally represented, suggesting that one class of drugs did not seem to offer advantage over another.[9] Adverse events were associated with intravenous, intramuscular, oral, rectal, nasal, and inhalational routes of

◆ This Editorial View accompanies the following article: Malviya S, Voepel-Lewis T, Ludomirsky A, Marshall J, Tait AR: Can we improve the assessment of discharge readiness? A comparative study of observational and objective measures of depth of sedation in children. ANESTHESIOLOGY 2004; 100:218–24.

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* Available at the American Academy of Pediatric Dentistry Web site: www.aapd.org/members/reference manual/pdfs/02–03/Sedation.pdf. Accessed August 30, 2003.

† Available at the American Society of Anesthesiologists Web site: www.asahq.org/Standards/20.htm. Accessed August 30, 2003.

‡ Available at the Commission of Accreditation of Healthcare Organizations Web site: www.jcaho.org. Accessed August 30, 2003.

administration. There was a significant association with death and neurologic injury when three or more sedating medications were administered. The majority of events presented with an adverse effect on respiration or oxygenation; however, a large fraction progressed to cardiac arrest, indicating the lack of skills to rescue the patient once a problem developed. Compared with a hospital-like setting, the incidence of death or neurologic injury was threefold higher in an office venue. Two children died in car seats before arriving at the healthcare facility. Ten others (nine who died or had neurologic injury) suffered the event in the automobile or at home after discharge. These patients had each received medications with long half-lives: chloral hydrate (the drug used in the Malviya *et al.* study), promethazine, chlorpromazine, and intramuscular pentobarbital. The majority of these adverse outcomes were clearly preventable, and it was not the drug or the route of administration, but rather the practitioner's lack of rescue skills and inadequate recovery.

The current study has scientifically examined the important safety issue of discharge readiness after sedation with a long-acting drug. They compared their current hospital discharge criteria with a new sedation score (the University of Michigan Sedation Score), combined with a simple Modified Maintenance of Wakefulness score (infants had to be able to stay awake for at least 20 min while observed in a soporific environment). They showed that the use of discharge criteria based on these new scores of alertness ensured that more than 90% of children had returned to baseline, compared with only 55% of children assessed as street-ready according to their current hospital criteria. Malviya *et al.* show very clearly that chloral hydrate can result in prolonged sedation, even after the children reach currently used discharge criteria. In our outcomes study, some of the children died from falling asleep in a car seat with their head falling forward; because of the residual sedating medications, they were unable to spontaneously unobstruct their airway.[8] The current study may represent a turning point in our specialty—a maturing process from looking beyond guidelines and regulations to placing scientific validity on processes that we previously could only assume to be the right thing to do.

The results of this study suggest that the population at greatest risk for prolonged sedation is infants and toddlers (those most likely to return home in a car seat).[1,8,9] It would make sense to implement these new discharge criteria *now*. Because anesthesiologists are central to the development of "within institution" sedation policies, our specialty is in the perfect position to make this happen. Implementation will increase costs, which is why the target population should be focused. The period of observation will be longer (more nursing time [$\sim$ \$7.50/patient hour based on $\sim$ \$30/h nurse salary, observing four patients simultaneously]), and the facility charge will be greater ($\sim$ \$150/h in my institution for phase II recovery observation). Also, finding a quiet venue for this soporific observation period requires more hospital space dedicated to sedation (renovation costs or, alternatively, use of intake areas for both intake and stepdown observation). We all must be proactive and very creative to find the space and resources to sedate children safely. Because a parent's single greatest concern is his or her child's safety, parent participation may be central to rapidly implementing this extended period of observation at a lower cost, provided the reason for "quiet time observation" is explained. It is very likely that such careful assessments of wakefulness will prevent adverse outcomes.

The medical profession, along with the insurance industry and hospital administrators, must progress to the next level to truly make sedation safe. The insurance industry must recognize that some children can be safely cared for only in the hospital setting and by anesthesiologists; they also must compensate us fairly for this service. Hospitals must recognize that developing the proper safety net is expensive in terms of personnel (extra nurses) and facilities (properly equipped sedation and recovery areas). In some hospitals, the administration may need to supplement the income of those in the anesthesiology department to facilitate coverage. Anesthesiologists in turn must recognize that we cannot be present for every patient who requires sedation, and that this process must be provided and supervised by other physicians, *e.g.*, emergency medicine or intensive care specialists who have advanced airway training. In other situations, trained advanced-practice nurses supervised by nonanesthesiologists may provide the sedation. Hospitals must support anesthesiologists' efforts to educate and train these practitioners to do this safely. The current study has addressed one issue, but a number of questions remain to be investigated and clarified. When is it safe to discharge patients sedated with different classes of drugs? What are the safety implications when, as my postanesthesia care nurses describe, it often takes 2 to 4 h longer for the children sedated for magnetic resonance imaging to recover from sedation administered by sedation nurses compared with children anesthetized for the same procedure? What drugs are best for specific procedures? Should the use of certain drugs be restricted to anesthesiologists? Should nonanesthesiologists be taught how to safely use drugs traditionally used as general anesthetics (*e.g.*, propofol)? In times of economic restraint, should nonanesthesiologists with advanced airway training be allowed to bill for anesthesia services? How does the healthcare industry finance the costs of safe sedation? The problem is that far more procedures require sedation than anesthesiologists have the time or desire to cover. How do we as a specialty maintain our vital role in this process without strangling

ourselves with so many sedations that we cannot possibly cover them all?

Our specialty has an amazing track record of examining process and improving safety. We have reduced anesthetic mortality tenfold, we have reduced medication errors, and we have been essential in developing the specialties of Intensive Care and Pain Medicine. Now it is time for our specialty to go beyond the turf battles described above and help develop the best practices for non-anesthesiologists who administer sedation. Through the JCAHO mandate, our specialty can help establish sedation committees that examine quality assurance issues, similar to a code committee. Anesthesiologists do not need to be the "sedation police," but through the committee process (and with the imprimatur of the JCAHO regulations), we can help each hospital develop a commitment to evaluate and change the best practices for sedation. The current study has addressed one issue that could be pivotal in improving the safety of discharging infants sedated with long-acting medications. The simple use of the University of Michigan Sedation Scale and meeting the criteria of "Can the child stay awake for 20 min when undisturbed?" would be wonderfully easy assessments to perform. I applaud Malviya *et al.* for taking an interest in developing better and simple discharge criteria in an area of practice generally avoided by anesthesiologists. I challenge my anesthesiology colleagues to answer the many questions that remain.

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Long-lasting Changes in Brain Protein Expression after Exposure to an Anesthetic

WITHIN the genus of drugs capable of rendering a patient sufficiently unaware to tolerate the anguish of surgery, only a few are compatible with full reintegration of the personality at the conclusion of the procedure. We have become so adept in the delivery of this narrow subset of agents that the marvel of anesthetic reversal is considered commonplace, and is taken for granted by caregivers and researchers alike. Still, can it be that such a dramatic intrusion on the normal function of so complex a system leaves no echo or aftershock? Work reported in this issue of the Journal suggests otherwise.[1]

This Editorial View accompanies the following article: Fütterer CD, Maurer MH, Schmitt A, Feldmann RE Jr, Kuschinsky W, Waschke KF: Alterations in rat brain proteins after desflurane anesthesia. ANESTHESIOLOGY 2004; 100:302–8.

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Fütterer *et al.* exposed rats to 3 h of a single concentration (5.7%) of desflurane in air. Cytosolic proteins isolated from whole brain homogenates at the immediate conclusion of the exposure interval, and at 24 and 48 h thereafter, were separated by two-dimensional polyacrylamide gel electrophoresis (2D PAGE), stained prior to identification of the excised spots by mass spectrometry (MS), and quantified by comparison of spot volumes with those derived from unexposed control animals. Analysis of spots taking up the stain revealed a handful of proteins with either increased or decreased relative abundance persisting 72 h after anesthetic inhalation. The authors' contribution represents the first, albeit preliminary, report of a change in the profile of expressed protein content in the brain after administration of an anesthetic drug in widespread clinical use, and merits consideration in its broader context.

To tackle their novel research question, Fütterer *et al.* have taken a well-traveled proteomic approach. The proteome is generally defined as the complement of pro-

teins expressed by a genome at a particular point in time. Proteomics refers to the qualitative and quantitative comparison of proteomes used to elucidate the differences between two states of a cell, tissue, or organism, *i.e.*, awake and anesthetized. Proteomic research aims to identify and quantify all proteins, protein isoforms and modifications, protein-protein interactions, structural and functional correlates, and higher-order complexities in a specific context. The endeavor is enabled by the advent of high-throughput methodologies permitting the parallel analysis of hundreds to thousands of proteins and peptides. Sorting by 2D PAGE, coupled with detection by MS, is the senior and most widely used method.

Compared to genes, proteins are structurally, functionally, and temporally much more complex. Because the important factor about a gene is its linear sequence, DNA analysis is a relatively straightforward problem of scalability. We know that the number of human genes (transcriptional units) is finite, falling within the range of 30,000–40,000, and well below most estimates made before completion of the Human Genome Project. Conversely, the number of distinct proteins, which function by virtue of their shifting three-dimensional shapes, is thought to exceed 1,000,000. Many factors account for the difference between the number of gene and protein species. Two that predominate are alternate splicing of the transcriptional unit, and posttranslational modifications of the nascent protein (*e.g.*, phosphorylation, glycosylation, methylation, or acetylation). Interestingly, a single gene (*e.g.* neurexin) may encode up to 1,000 different proteins.[2] Thus, the DNA sequence provides a template allowing investigators to compare predicted amino acid sequences from completed genomes with the constellation of measured proteomic data. The hurdle for proteomic research is that whereas each fragment of DNA behaves biochemically much like every other, each protein possesses unique properties, imparting differences in solubility, mass, isoelectric point, presence or absence of cofactors, and folding optima, among others. To complicate matters, the dynamic range of abundance in protein mixtures from biologic sources may span 10 orders of magnitude, with low abundance entities nevertheless subserving essential physiologic functions.

To confront these challenges, Fütterer *et al.* engage in quantitative expression profiling, wherein a biologic sample is characterized by separating, identifying, and quantifying as many proteins as possible, with a focus on those altered in relative abundance with reference to a control sample. In this version of discovery-directed research, investigators often have no idea what will be observed at the conclusion of their efforts. The objective is to generate fresh testable hypotheses and acquire original information about previously recognized proteins, rather than to validate suspected functions and interactions of differentially expressed proteins. As a corollary, results reported from such experimental designs must not be regarded as comprehensive. Failure of a specific protein to make the list does not mean that it is not present in the sample.

Although 263 spots embodying distinct proteins met criteria for analysis in the present investigation, it is reasonable to estimate that the brain as a whole expresses many hundreds of thousands of proteins within any given time frame. Where have the rest of the proteins gone? They have most probably fallen beneath the radar of the methods chosen by Fütterer *et al.* in this inaugural investigation, and their presence and relative abundance remain to be discerned by proteomic techniques and technologies capable of higher resolution of complex and mixed-abundance samples. Solubilization of the protein content of a heterogenous cellular population at the whole organ level, separation of intact proteins, and visualization by silver staining, as done in this study, permit only a limited display of polypeptides that are relatively plentiful in the composite. In particular, such a crude approach precludes detection of lipophilic membrane constituents of great interest to anesthesiologists (*e.g.*, ion channels, alkaline proteins, and multimeric protein complexes). Fortunately, a variety of strategies are available for use preceding the 2D PAGE separation step to reduce complexity, increase sensitivity, and enrich the sample. These include microdissection, ultracentrifugation, sequential extractions with reagents of increasing solubilizing power, pH purifications, isoelectric fractionation, subproteome digestion to signature peptides, and protein tagging.

2D PAGE separates proteins based on their electrical charge in the first dimension and their molecular mass in the second dimension, as reflected by divergent protein mobility in a polyacrylamide gel matrix. The technique enjoys wide popularity because high-affinity detection probes and previous knowledge about specific protein properties are not required. However, 2D PAGE is hampered by many constraints: substantial amounts of sample must be loaded, proteins manifest differential staining sensitivities, manual image analysis is a bottleneck to high-throughput data acquisition, and comigrating proteins confound analysis. Also, 2D PAGE is labor-intensive and exhibits significant experimental variation, as the spot selection protocol devised by Fütterer *et al.* attests. Alternatives to gel-based methods have recently been introduced, including liquid chromatography and protein-detecting microarrays.[3,4] These and related technologies are much more amenable to large-scale, "shotgun" determinations of complex sample admixtures, with advantages over 2D PAGE in sample size, scalability, flexibility, control of ambient conditions, and capacity for automation.

MS as used by Fütterer *et al.* is the detection method of choice in the preponderance of recent proteomic inves-

tigations.[3] The mass spectrometer is able to resolve many tens of thousands of protein and peptide species by measuring the mass to charge ratio (*m/z*) of ions. Matrix-assisted laser desorption/ionization is the process by which proteins refractory to ionization without destruction are converted first to peptides by trypsin digestion, and then to ions by short laser pulses prior to MS. Matrix-assisted laser desorption/ionization–time of flight analysis provides the simplicity, accuracy, and sensitivity necessary for peptide mass mapping, in which peptides are identified by matching a list of observed masses with the archived menu of all masses of each entry in a database. Although matrix-assisted laser desorption/ionization-time of flight is highly efficient in the identification of gel-separated proteins, the measured signal intensity does not correlate with the amount of analyte present in the sample because MS is not an inherently quantitative technique. To draw quantitative conclusions, other methods must be appended, such as the relatively coarse spot-volume estimates used by Fütterer *et al.* Even so, results are not reportable in absolute unit amounts, comprising a major limitation of 2D PAGE-MS methods.

High-throughput proteomics are currently restricted by requisite comparison to incomplete protein sequence databases. Decades may elapse before closure of the human (or, for that matter, any mammalian) proteome is approximated. Moreover, matching observational data to archived data are not failsafe. Rates of false identification, that is, the probability that the candidate peptide has produced the observed spectrum by chance, are not known with precision, underscoring the choice of Fütterer *et al.* to use the Mascot score for this purpose. Because protocols are based on successive iteration between experimental and archived data, a framework to estimate statistical power and appropriate sample size for up to tens of thousands of comparisons has yet to be determined. Statistical methods to estimate the significance of associations between protein expression patterns and sample groups remain close to the drawing board, although tools such as cluster analysis, in which proteins of unknown function clustering consistently with those of defined function become candidates for further validation, hold great promise. In any case, as discovery of protein expression patterns becomes increasingly high-throughput, functional validation at the bench will continue to be painstaking, and low-throughput, for years to come. The huge amounts of data generated by proteomic investigations have led to calls for the standardization of protein identification and quantification, and for the organization of the Proteomics Standards Initiative and Human Brain Proteome Project of the Human Proteome Project.*

Turning to the substance of the research question raised by Fütterer *et al.,* several additional precautions must be borne in mind. Any mapping exercise risks recapitulation of the debacle of phrenology, whether it be correlating traits to DNA sequence, or cellular perturbations to proteomic expression profiles. Great care must be taken in experimental design to minimize and, whenever possible, eliminate systemic, epiphenomenal associations unrelated to primary effects of anesthetic drugs on the nervous system. In this respect, Fütterer *et al.* must be commended for care taken to control the possible confounding influence of hypotension, hypoxia, hypoventilation, gender, and other background variables. Inevitably, recalcitrant variables (*e.g.*, the confounding consequences of immobility and loss of sensorimotor input to the nervous system during anesthetic exposure) will resist the design of even the most sophisticated trials. In the present context, few would argue that 5.7% desflurane (1 minimum alveolar concentration in the rat) represents a full-fledged model of surgical anesthesia. Single doses for single durations of single agents do not support firm or generalizable conclusions; they mandate more sophisticated experimental designs, replication of the authors' observations in other laboratories, and functional validation of the reported protein express fluctuations. Confirmation with corollary methods are also awaited, using, for example, two-dye fluorescent labeling of pooled proteins from anesthetized and awake sources, separated on the same 2D-DIGE (2-D Differential In-Gel Electrophoresis) gel to quantify differential expression on a single platform, and imaging MS of whole brain sections.[5,6]

A small but growing body of literature indicates that anesthetics in clinically relevant concentrations and durations have profoundly detrimental neuronal consequences in those predisposed by environment, age, and genotype.[7–10] That the consequences of drug-induced coma fade without repercussion in the otherwise normal brain may be as much a product of wishful thinking as of collective unwillingness to test the axiom. Indeed, 2D PAGE-MS has been available for several decades, but until Fütterer *et al.*, no one has thought to perform the relevant (and, in retrospect, compelling) investigation. In turn, companion investigations of protein expression profiles after anesthetic exposure in heart, liver, lung, vascular smooth tissue, and other tissues are readily envisioned. Besides confirmation and elaboration of observations made by Fütterer *et al.*, the profession's burden going forward will be to perform the problem-oriented research necessary to transfigure broad, but ultimately shallow, proteomic insights into deeper biological understanding, thereby devising safer, more effective, and possibly more evanescent anesthetic interventions.

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Genetic Testing for Malignant Hyperthermia in North America

CAFFEINE-HALOTHANE contracture testing (CHCT) of fresh, surgically removed skeletal muscle has been the basis for identifying individuals who are susceptible to malignant hyperthermia (MH). CHCT is invasive, expensive, and currently performed at five specialized centers in the United States, two in Canada, and one in Brazil, and has a sensitivity and specificity of 97% and 78%, respectively.[1] Similar *in vitro* contracture testing has been used across Europe to identify MH-susceptible individuals and has sensitivity and specificity values of 99% and 94%, respectively.[2] A less invasive, highly sensitive, and specific diagnostic test for MH has been actively sought for many years. Recent studies, coupling functional and genetic causes for MH, have brought genetic testing for this anesthetic-induced, life-threatening disease to the forefront.[3] In this issue of the Journal, the report of a September 2002 meeting sponsored by the Malignant Hyperthermia Association of the United States represents an important first step toward a better diagnostic test for MH in North America.[4]

Volatile anesthetics are the primary trigger of MH, causing an abnormally increased release of calcium within skeletal muscle cells.[3] Mutations in the gene (RYR1) encoding the skeletal muscle calcium release channel (ryanodine receptor protein RyR1), are linked to MH susceptibility in humans,[5,6] pigs,[7] and dogs.[8] In each species, skeletal muscle is characterized by abnormal *in vitro* contracture responses to caffeine and halothane. The MH syndrome is effectively prevented and treated by dantrolene, which inhibits intracellular Ca^{2+} release from the sarcoplasmic reticulum Ca^{2+} stores by binding to, and thus decreasing, the RyR1 channel open-state probability.[9,10] Many RYR1 mutations have been expressed in heterologous systems (myotubes, COS-1, or human embryonic kidney cells) that show enhanced calcium fluxes when treated with RyR1 agonists.[11–13] Collectively, this might seem to encompass and resolve the genetic basis for MH, but unfortunately, complicating issues exist. RyR1 is a homotetrameric protein. Each subunit has a molecular weight of 560 kDa (5,038 amino acids), making it one of the largest proteins known. Located on chromosome 19, the RYR1 gene spans 160,000 nucleotide bases, consists of 106 exons; as such, it is one of the most complex human genes. Consequently, most laboratories can only look at small pieces of the RYR1 gene when searching for mutations that might link to MH. Despite these technologic barriers, over 40 different MH-associated RYR1 mutations have been found in three different regions of the gene. Another complicating factor is that MH is genetically heterogenous; *i.e.*, mutations in RYR1 have not been identified in all MH families. Nevertheless, it is expected that once all mutations in RYR1 are identified, they may account for up to 70% of MH among all susceptible families.[14] As for the other non-RYR1 genes associated with MH, mutations in the gene encoding the alpha subunit of the dihydropyridine receptor have been reported, but these seem to be very rare.[15,16] Five other chromosomal loci (17q21–24, 1q32, 3q13, 7q21–24, and 5p) have linkage to MH, but the genes are not yet identified.

Diagnostic genetic screening for MH was initiated in Europe over 2 yr ago. The European Malignant Hyperthermia Group established guidelines for RYR1 mutation screening with 15 causative RYR1 mutations selected for initial testing.[17,18] The first step is identification of an MH-susceptible individual using the validated *in vitro*

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contracture test. Screening with a panel of 15 different RYR1 mutations follows. If an RYR1 mutation is detected, then other first-degree relatives of that individual can be tested; those in whom the mutation is found are diagnosed as having MH without undergoing *in vitro* contracture testing. However, if the particular familial mutation is not found, the muscle (*in vitro*) contracture test is required for MH diagnosis. This policy avoids false-negative diagnoses. In one European center, introduction of genetic testing allowed the diagnosis of MH susceptibility to be confirmed in approximately 50% of the proband's relatives.[14]

The search for a less invasive method than CHCT to diagnose MH has been ongoing for many years. Many approaches have been tried, but none have supplanted the muscle contracture test.[19] Newer tests based on advances in molecular genetics and cellular physiology have the potential to be effective. The newer tests include measurements in Ca^{2+} fluxes studied either in cultured skeletal muscle cells or in lymphoblastoid cells naturally expressing RYR1. Censier *et al.*[20] reported enhanced intracellular calcium release from muscle cultured from MH-susceptible patients. Sei *et al.*[21,22] identified and characterized the RYR1 in human B-lymphocytes and reported that Ca^{2+} release induced by caffeine and 4-chloro-m-cresol was greater in cells from individuals susceptible to MH than in normal individuals or patients testing negative for CHCT. Also, Loke *et al.*[23] recently demonstrated that direct sequencing of RYR1 transcripts from viable leukocytes could be used to analyze the complete RYR1 in blood samples. Further studies are required to determine the diagnostic potential of these tests.

Another approach has been the use of nuclear magnetic resonance spectroscopy to noninvasively measure adenosine triphosphate, pH, creatine phosphate, and other high-energy phosphates.[24] With exercise, MH-susceptible individuals demonstrate greater depletion of high-energy phosphates and a decrease in pH compared to people without MH. Yet other investigators have shown that *in vivo* microinjection of caffeine in muscle produces an increase in carbon dioxide output and hydrogen ion production in MH-susceptible indiviuals.[25] A multicenter study to evaluate this test in a larger number of patients is in the planning stages in European MH centers.

The disadvantages of contracture testing are that CHCT must be performed on fresh, surgically removed skeletal muscle (usually vastus lateralis), and total costs at one of the MH diagnostic centers (including testing, anesthesia, preoperative surgical assessment, and hospital charges) can range from $5,000 to $6,000. With the reduced number of MH testing centers, patients can incur losses in time, travel, and housing costs. In 2002, the North American MH Genetics Group[4] developed guidelines for genetic MH diagnosis, taking advantage of the European Malignant Hyperthermia Group model. In addition, for the past 5 yr, the North American MH group's active research program has screened patients diagnosed as having MH by CHCT for RYR1 mutations and has found most results to be consistent with the European data.[26] However, some mutations appear to be specific to the North American population.[27,28] On the basis of these results, the North American MH Genetics Group has identified the priorities for initial MH genetic screening. The panel of 17 RYR1 mutations proposed at the recent genetic workshop will be used. This panel will be updated as new causative mutations are discovered. Families to be tested must be identified by a CHCT-positive result or by a strong history for MH and will be referred from a MH Diagnostic Center. The North American MH Registry database can be used to identify potential families and to maintain the results of genetic testing. Samples of DNA (blood or buccal cells) will be obtained and sent to the genetic testing laboratory. The genetic MH testing laboratory must be a Clinical Laboratory Improvement Act–certified laboratory to receive Medicare and Medicaid payments. For each family, initial testing will involve screening for the 17 mutations recommended in individuals determined to have MH and will be the most costly (estimates are unavailable). Once a mutation is identified in the affected member, then family members will be offered testing for the presence of the family-specific mutation. Mutation-positive members would be regarded as MH-positive without further CHCT testing, and the cost will be considerably less. To avoid the danger of a false-negative diagnosis, it will remain necessary to continue performance of CHCT for diagnosis of those family members who do not carry the familial RYR1 mutation. The initial genetic screening for MH will be limited by low sensitivity because the recommended panel of mutations does not cover all potential mutations. At this time, it is not practical to screen the entire RYR1 gene or all RYR1 mutations in each individual with MH. However, the panel of 17 mutations having the highest frequency of occurrence among North American MH families is a starting point. This step introduces new diagnostic tools to the MH centers and, in those MH-positive families in which a causative RYR1 mutation is identified, many individuals will be spared the expensive and invasive CHCT test. As is standard practice in the diagnosis of other genetic diseases, genetic counseling will be necessary; initially, this may be performed through the MH Diagnostic Center from which the patient was referred. Future developments of MH genetic screening will be documented on the Web site of the Malignant Hyperthermia Association of the United States.§

§ Web site: http://www.mhaus.org. Accessed June 16, 2003.

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